

## Research Article

# SEROPREVALENCE AND RISK FACTORS ASSOCIATED WITH TOXOPLASMA GONDII INFECTION AMONG PREGNANT WOMEN ATTENDING ANTENATAL CARE IN PLATEAU STATE SPECIALIST HOSPITAL, JOS, NIGERIA

<sup>1</sup>DANUAN, M.B, <sup>1</sup>DAKUL, A.D, <sup>1\*</sup>ADELEKE, E.A, <sup>2</sup>LUMI, E.B, <sup>3</sup>CHUNDUNG, N.D

<sup>1</sup>Department of Zoology, University of Jos, Plateau State, Nigeria.

<sup>2</sup>Department of Science Laboratory Technology, University of Jos, Plateau State, Nigeria.

<sup>3</sup>Department of Microbiology, University of Jos, Plateau State, Nigeria.

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### ABSTRACT

Toxoplasmosis has been famously described as a cosmopolitan disease caused by the zoonotic parasite known as *Toxoplasma gondii* with the capacity to induce miscarriage and congenital defects during pregnancy. Despite its public health significance, there is a dearth of information about the epidemiology of the infection in pregnant women in Nigeria, consequently this study aimed to determine the burden and risk factors associated with *T. gondii* infection in pregnant women attending Ante-Natal Clinic (ANC) at Plateau State Specialist Hospital, Jos, Nigeria. 3mls venous blood was collected from 268 pregnant women within the reproductive age groups of 15 to 50 years. Structured questionnaire was used to obtain demographic and *Toxoplasma* infection risk related data. Sera were assayed using a commercial ELISA kit (Novo Tec Immunodiagnostica® Germany). 46 out of the 268 women were positive with an overall prevalence rate of 17.16%. The highest prevalence was recorded among women within the age group  $\leq 20$  years (28.00%), urban settlers (18.18%), women in their second trimester 18.75%, those who owned cats (25.00%) and those who have contacts with cat's faeces (29.10%). There was a significant association ( $p < 0.05$ ) between the prevalence of *T. gondii* antibodies (IgG) with age, ethnic group and blood transfusion history. While other factors show no significant relationship ( $p > 0.05$ ). Asymptomatic Toxoplasmosis common amongst pregnant women thus it is imperative for policy makers and caregivers to consider introducing routine screening of toxoplasmosis especially as part of antenatal program for pregnant women.

**Keywords:** *Toxoplasma gondii*, Seroprevalence, Pregnant Women, Risk Factors, Plateau State.

### INTRODUCTION

*Toxoplasma gondii* is a microscopic, obligate intracellular alveolate parasite which is responsible for the disease toxoplasmosis with a world-wide distribution (Tenter *et al.*, 2000). Toxoplasmosis occurs in a wide range of warm-blooded animals including humans. In 2010 *T. gondii* was implicated to have caused more than 20 million new cases worldwide (Torgerson, 2013) and CDC (2015) reported that the disease has infected over 30–50% of the world human population whereas Foroutan and Ghaffarifar, (2018) reported the disease to have infected more than  $\frac{1}{3}$  of human population globally. The transmission of *T. gondii* is mainly through accidental ingestion of infective stages in contaminated soil, food or water and the eating of infected undercooked or raw meat (Awobode and Olubi, 2014). Some studies have identified water as a potential source of the infection in both humans and animals (Bowe *et al.*, 1997, Miller *et al.*, 2002, Bahia-Oliveira *et al.*, 2003, De Moura *et al.*, 2006). Robert-Gangneux & Darde, (2012) reported that though rarely, unpasteurized milk containing the parasite's cyst are capable of transmitting the parasite and also blood transfusion or organ transplant from infected individuals. If pregnant women become infected with *T. gondii*, one of the major consequences is vertical transmission to the foetus also known as congenital toxoplasmosis, the disease condition that may associate with miscarriages, severe neurological or ocular disease (leading to blindness), as well as cardiac and cerebral anomalies in the newborn and even fetal death (Jones *et al.*, 2003, Jones *et al.*, 2010 and Paul *et al.*, 2018). Congenital toxoplasmosis usually occurs when the mother acquires a new infection just prior to or during pregnancy with a higher risk

at the third trimester of pregnancy towards maturity than if acquired during the first trimester. However, the earlier the foetus is infected, the greater the effects to the foetus and newborn (Al-Hamdani & Mahdi, 1997, Dunn *et al.*, 1999 and Foulon *et al.*, 1999). Ante-natal serological screening of *T. gondii* infection based on IgG and IgM detection is the mainstay in monitoring the risk for congenital toxoplasmosis, which indicates previous exposure. Epidemiological studies recording prevalence of *T. gondii* infection in pregnant women around the world indicate considerable variation between countries, ranging, for example, from 9% to 67% in European countries (Lopes *et al.*, 2013). In Africa, the seroprevalence of *T. gondii* during pregnancy is generally as high as 80% (Zemene *et al.*, 2012). In Ghana Ayi *et al.*, (2009) reported a prevalence of 92.5%. In Nigeria, the prevalence of Toxoplasmosis among pregnant women have been reported in various studies. Deji-Agboola *et al.*, 2011 Showed 40.4% in Lagos, South-West Nigeria, Nasir *et al.*, (2015) presented 48.9% prevalence in Maiduguri, North-East Nigeria, Oboro *et al.*, (2016) revealed 65.6% in Port Harcourt South-South Nigeria and Ibrahim *et al.*, (2017) showed 43.4% in Kano North-West Nigeria. With this enormous burden of toxoplasmosis amongst pregnant women in Nigeria, it underscores the need for early diagnosis and appropriate treatment as the most reliable ways of reducing the risk of trans-placental transmission and subsequent sequelae in the newborn (Pomares *et al.*, 2016, Remington *et al.*, 2004). Although cases of toxoplasmosis have been reported in Nigeria, yet it is obvious that in many parts of the country there is significant paucity of information on Toxoplasmosis amongst pregnant women and even in the areas where it has been described, these cases are under reported possibly because of the absence or ineffective screening program during antenatal and postnatal visits. In this study however we sought to determine the seroprevalence of Toxoplasmosis *gondii* and its associated risks

\*Corresponding Author: ADELEKE, E.A,

<sup>1</sup>Department Of Zoology, University Of Jos, Plateau State, Nigeria.

factors amongst pregnant women following Ante-natal clinic (ANC) services at Plateau State Specialist Hospital, Jos, North Central Nigeria.

## MATERIALS AND METHODS

### Study Area

The study was conducted at the Plateau State Specialist Hospital (PLSSH), located at the metropolitan city of Jos - North LGA of Plateau State, North Central Nigeria. Jos is the capital of Plateau State located on Latitude 9° 55'42.6"N and Longitude 8° 53' 31.6"E with an area of about 291Km<sup>2</sup> with average elevation of 4,000 ft.(1,220m). According to National Population Commission (2006), it has an estimated population of about 900,000 people.

### Ethical Consideration

The study protocol was approved by the Health Research and Ethics Committee of Plateau State Specialist Hospital (PSSH) with the Registration number NHREC/05/01/2010b. All the study participants gave their informed consent for inclusion before they were included in the study. A total of Two hundred and sixty eight (268) pregnant women were recruited for this study within the reproductive age groups of 15 to 50 years old, receiving antenatal at PLSSH, Jos. Volunteers were made to fill a structured questionnaire where demographic data and information on individual risk factors were obtained.

### Sample collections and preparations:

3mls of venous blood was collected using a sterile disposable syringe from each consenting participant into plain vacutainer tubes. The tubes were then appropriately labeled with patients' laboratory number. Collected blood were allowed to coagulate at room temperature for about 10min before serum was separated by centrifugation and later centrifuged at 3000 rpm for 10 minutes. The serum was separated using clean Pasteur pipettes and stored at -20°C before serological tests.

### Detection of *Toxoplasma gondii* IgG antibody

Qualitative detection of *T. gondii*-specific IgG was performed using ELISA commercially prepared kit obtained from Novo Tec Immunodiagnostica® (Germany) according to manufacturer's instructions. Optical densities were measured by spectrophotometer at a wavelength of 450nm. Results were defined semi-quantitatively by calculating a ratio of the extinction value for test samples (optical density) to the extinction value of the calibrator positive and negative control sera. Values higher than the cut-off  $\geq 0.09$  were considered positive while Values  $< 0.09$  was considered negative for TOX IgG

### Statistical analysis

Data obtained in this study were analyzed using the Statistical Package for Social Sciences (SPSS) version 23.0. Continues variables were summarized descriptively using the measure of central

tendencies and categorical data are reported as tabulation of proportions. Association between various risk factors and the prevalence of *Toxoplasma* infection was determined using Chi-square test, where  $p < 0.05$  was considered statistically significant.

## RESULTS

Of the two hundred and sixty-eight pregnant women (268) examined in this study, forty-six (46) of them tested positive to *T. gondii* IgG antibodies giving an overall prevalence of 17.16%. Table 1 below showed the association between age and *Toxoplasma* antibodies. In this table the highest number of respondents (49.3%) of the 268 women that indicated their age group were between the age group 21 to 30 years, followed closely by those between the ages 31 to 40 years (39.2%). The mean age of the participants was found to be  $26.37 \pm 0.66$  years. The disease distribution in relation to age groups showed that the women with ages less than or equal to twenty ( $\leq 20$ ) years old had more of the infection with a burden of 28.00%, followed closely by those women between the ages of 21-30 years old who had a prevalence of 19.69%. In this study we observed that women of ages 41-50 years had no toxoplasma infection (0.0%). T-test analysis showed a significant association amongst the different age groups in this study ( $p < 0.05$ ).

**Table 1: Distribution of *Toxoplasma* Antibody among the study subjects in relation to age**

Age (yrs)	IgG ANTIBODY				
	No. Examined	No. Positive(%)	T	df	P-value
$\leq 20$	25	07 (28.00)	2.884	63.96	0.005
21 – 30	132	26 (19.69)			
31 – 40	105	12 (11.43)			
41 – 50	06	0 (0.00)			
Total	268	46 (17.16)			

Table 2 showed the association between *Toxoplasma* antibody and socio-demographic factors. Those women who reside in the urban centers (18.18%) were positive for IgG while those who reside in the rural centers had no *T. gondii* infection. 17.8% of the married women were infected with the parasite while those who are single had none. While considering educational background of participants, we observed that those with primary level of education had highest number of *T. gondii* infection with 25.00%, trailed closely by those who had no formal education with 23.08% burden of the infection. In relation to occupation, women that are unemployed showed the highest infection with 18.39%, Artisans and market women showed a prevalence of 17.86% and 12.50% respectively. The least infection 11.11% was found amongst women that engage in farming/abattoir market. In this study the Hausa/Fulani ethnic group demonstrated the highest *T. gondii* infection (29.51%), than other ethnic groups. Chi square analysis revealed no significant association between all the above demographic factors i.e. residence, marital status, educational background and occupation ( $p > 0.05$ ) except for ethnicity ( $p < 0.05$ ).

**Table 2. Distribution of *Toxoplasma* Antibody Among the Study Subjects in Relation to Socio-Demographic Factors.**

Sociodemographic Factors	IgG ANTIBODY				
	No. Examined	No. Positive (%)	$\chi^2$	df	p-value
<b>Residence</b>					
Urban	253	46 (18.18)	3.292	1	0.07
Rural	15	0 (0.00)			
Total	268	46 (17.16)			

<b>Marital Status</b>					
Single	10	0 (0.00)	2.152	1	0.14
Married	258	46 (17.82)			
Total	268	46 (17.16)			
<b>Educational Background</b>					
Informal	13	3 (23.08)	2.095	3	0.55
Primary	28	7 (25.00)			
Secondary	122	20 (16.40)			
Tertiary	105	16 (15.24)			
Total	268	46 (17.16)			
<b>Occupation</b>					
Employed	52	8 (15.38)	0.473	4	0.98
Unemployed	87	16 (18.39)			
Garden/Farmer/Abattoir	09	1 (11.11)			
Market	08	1 (12.50)			
Artisan	112	20 (17.86)			
Total	268	46 (17.16)			
<b>Ethnic Group</b>					
Hausa /Fulani	61	18 (29.51)	8.827	3	0.03
Igbo	3	0 (0.00)			
Yoruba	5	0 (0.00)			
Others	199	28 (14.07)			
Total	268	46 (17.16)			

In Table 3, the association between *Toxoplasma* antibodies and possible risk factors was highlighted. Those women who had never experienced any of abortion, still birth or miscarriage showed high infection rate of 19.88%, while those with the experience more than twice had the least infection rate at 9.09%. Statistical analysis amongst the various groups showed insignificant relationship ( $p>0.05$ ). The age of pregnancy revealed that the infection was recorded more 18.75% amongst women in their second trimester than the others. There was also no significant relationship ( $p>0.05$ ). In relation to Cat (pet) ownership, the results displayed that women who own cat showed more burden of the toxoplasmosis disease at 25.00% though without a statistical significance ( $p>0.05$ ). Furthermore, contact with cat's faeces as a risk factor also showed that infection is higher (29.16%) in women who have had contact with cat's litter 29.2%, than those have no such history 16.1% though there was no significant relation between the groups. Handling of raw meat and consumption of undercooked meat indicated that more infection was recorded among those that do not handle raw meat 23.73% than those who handled. Also women that consume meat also showed more infection rate (17.42%). Likewise results demonstrated more *T. gondii* infection 17.6% in women who were in the habit frequent consumption of suya than those who do not. In all of these, statistical analysis revealed no significant association among all the groups compared above. Another factor was the mode of vegetable consumption which showed that women who preferred their vegetables cooked recorded more *T. gondii* infection with a prevalence of 24.14%, while those who would rather consume them fresh and raw had a burden of 17.05%. There was also no significant relationship amongst them ( $p>0.05$ ). The type of drinking water as another factor revealed more infection 18.56% among those who drink sachet water and those who drink pipe borne water with 16.00%. No significant association between both groups. Considering the Blood transfusion history in the last 12 months amongst the study participants as a factor revealed that women who had history of blood transfusion in the last 12 months had the highest disease burden 351.58% than those without such history 16.06%. Statistical relationship between the two groups indicated a significant association between them ( $p>0.05$ ). Preference in the consumption of beef had the highest number 24.00% of positive cases for *T. gondii* IgG antibodies, while those who consumed goat/mutton and dog meat had the least number of positive cases, 10.6% and 14.7% respectively. There was no significant association among the group ( $p>0.05$ ).

**Table 3. Distribution of *Toxoplasma* Antibody among the study subjects in relation to Possible Risk Factors.**

Risk Factors	No. Examined	IgG ANTIBODY		$\chi^2$	df	p- value
		No. Positive (%)				
<b>Still Birth, Abortion, Miscarriage</b>						
Once	65	09 (13.85)		2.318	3	0.51
Twice	21	02 (9.52)				
More than Twice	11	01 (9.09)				
Never	171	34 (19.88)				
Total	268	46 (17.16)				
<b>Trimester</b>						
First	34	05 (14.71)		0.820	2	0.66
Second	176	33 (18.75)				
Third	58	08 (13.79)				
Total	268	46 (17.16)				

<b>Ownership of Cats</b>					
Yes	64	16 (25.00)	3.500	1	0.06
No	204	30 (14.71)			
Total	268	46 (17.16)			
<b>Contact with Cats Feaces/Litter</b>					
Yes	24	07 (29.10)	1.801	1	0.18
No	56	09 (16.07)			
Total	80	16 (20.00)			
<b>Only 80 responded</b>					
<b>Handling of Raw Meat</b>					
Yes	209	32 (15.31)	2.195	1	0.14
No	59	14 (23.73)			
Total	268	46 (17.16)			
<b>Meat Consumption</b>					
Yes	264	46 (17.42)	0.421	1	0.52
No	04	0 (0.00)			
Total	268	46 (17.16)			
<b>Blood Transfusion in the Last 12 Months</b>					
Yes	19	06 (31.58)	4.114	1	0.04
No	249	40 (16.06)			
Total	268	46 (17.16)			
<b>Form in Which Meat is Consumed</b>					
Mildly Cooked	23	04 (17.39)	2.388	2	0.30
Cooked Till Soft	236	39 (16.52)			
Cooked but Tough	09	3 (33.33)			
Total	268	46 (17.16)			
<b>Taste Meat While Cooking</b>					
Yes	169	26 (15.38)	0.933	1	0.33
No	99	20 (20.20)			
Total	268	46 (17.16)			
<b>Consumption of Suya</b>					
Yes	238	42 (17.65)	0.198	1	0.656
No	30	04 (13.33)			
Total	268	46 (17.16)			
<b>Type of Drinking Water</b>					
Pipe Borne Water	133	21 (15.79)	0.288	2	0.87
Well Water	38	07 (18.42)			
Sachet Water	97	18 (18.56)			
Total	268	46 (17.16)			
<b>Consumption of Vegetables</b>					
Yes	261	44 (16.86)	0.639	1	0.42
No	07	02 (28.57)			
Total	268	46 (17.16)			
<b>Form of Cooking Meat</b>					
Frying	175	29 (16.57)	0.281	2	0.87
Roasting	19	04 (21.05)			
Stewing	74	13 (17.57)			
Total	268	46 (17.16)			
<b>State of Vegetables Before Consumption</b>					
Fresh and Raw	88	15 (17.05)	3.516	2	0.17
Steamed	122	17 (13.93)			
Cooked	58	14 (25.14)			
Total	268	46 (17.16)			
<b>Type of Meat Consumed</b>					
Pork	37	07 (18.92)	4.812	4	0.31
Goat/Mutton	47	05 (10.64)			
Beef	75	18 (24.00)			
Chicken	75	11 (14.66)			
Dog Meat	32	05 (14.71)			
Total	268	46 (17.16)			

## DISCUSSION

The infection trend with *T. gondii* in Nigeria from available documents has shown a consistent high prevalence pattern, this is particularly worrisome because transmissions of the disease seem uninterrupted since majority of the populace are ignorant of the disease. Montoya and Liesenfeld, (2004) argued that because acute infection in most infected persons is asymptomatic or mild, infection during pregnancy go unnoticed and may result in transmission to the foetus with severe signs and symptoms. In this study, a lower prevalence (17.19%) of IgG antibodies was recorded which is in contrast with other findings where higher seroprevalence levels (78%) was reported by Onadeko et al., (1992) in Ibadan, 40.8% reported by Akinbami et al.,(2010) in Lagos, 65.6% by Oboro et al., (2016) in Port Harcourt, 48.9% by Nasir et al.,(2015) in Maiduguri and 43.4% by Ibrahim et al., (2017) in Kano. Elsewhere outside Nigeria are the reports of Ayi et al., (2010), who reported a prevalence of 92.5% in Ghana, Shimelis et al., (2009) in Addis Ababa, Ethiopia who recorded 93.3%. However our findings corroborated the report of Cong et al, (2015), a case control study in eastern China among pregnant women where a 17.3% prevalence was reported. Jenum et al., (1998) and Nash et al., (2005) both reported (10%) in Norway and United Kingdom respectively. In Nigeria, Ogo (2016) also reported a lower prevalence (12.1%) amongst pregnant women in Jos. Several factors are responsible for the marked differences in the burden of *T. gondii* as observed in these different regions (even among those in the same country), among which may perhaps be due to variation in geographical and climatic conditions, since according to Logar et al., (2002), oocyst sporulation is enhanced by hot and wet conditions. Also, different techniques and sample size employed in these studies may pay a significant role in the results they present and finally differences in cultural and religious believes etc., may have also impacted in the burden of the infection in the different study areas (Ballah et al., 2017) In relation to age, *T. gondii* infection showed a significant increase with age in other words the younger women are likely to acquire the disease than their older counterparts. Though in our study older women between the ages 41 to 50 years showed no single infection with toxoplasmosis, it is convenient to conclude that the number of individuals in this group may have affected this finding. Conversely some studies corroborated our findings that increase in age reduces the vulnerability of individuals from acquiring the disease, amongst which are the studies of and Al-Qurashi et al., (2001), and Ogo (2016). However, Ayi et al., (2010) reported that the acquisition of toxoplasmosis infection has nothing to do with age. The conceivable reason we suppose, why *T. gondii* infection was higher among younger women than older ones may be a reflection of possible behavioral differences, eating characteristics and exposure by this age group of women to the environment which might have been contaminated with sporulated oocysts and hence serves as a medium that is critical for the transmission of parasites (Ballah et al., 2017, Al-Qurashi et al., 2001).Erstwhile studies had reported that being rural dweller, having low education/illiterate, married and unemployed (Kamal et al., 2015; Nasir et al., 2015), were all found to be risk factors for contracting *T. gondii* infection. However, none of these factors were significantly associated with *T. gondii* infection. Puccioet al., (2014) also corroborated this finding while Paul et al, (2018), explained that this was likely so because the study was a hospital-based study, involving a special group of women, who may not represent the wider community. Thus the need to conduct more studies that involve pregnant women from different settings to establish and compare the *T. gondii* seroprevalence among these different groups cannot be overemphasized. Also they opined that there is also a need to conduct longitudinal studies across seasons of the year to determine the seasonality of toxoplasmosis in pregnancy to establish these finding. In our study, the Hausa/Fulani ethnic group

was more vulnerable to the *T. gondii* infection than the other ethnic groups. Bessong & Mathomu, (2010) reported ethnicity may not play any significant role in the disease transmission, referring to other work in Northwest Ethiopia and South Africa, though with smaller sample size being the reason their study showed no statistical significance. Nevertheless we understand even by corroborating other studies that ethnic groups could become vulnerable to *T. gondii* infection as a result of cultural practices, eating habits, and food preferences, and also largely religious believes (Logaret et al., 2002).The current study observed more *T. gondii* infection among pregnant women who have had history of abortion, miscarriage or still birth than those women without. Although we understand that miscarriage, abortion or still birth may be conditions associated to congenital toxoplasmosis even though our study could not establish any significant association between women who had experience any of these conditions in previous pregnancies and those who did not. Previous studies have corroborated this finding, e.g., Gogate et al., (1994) in India, Attia et al., (1995) in Egypt, Pal et al., (1996) in Pakistan, Alvarado-Esquivel et al, (2014) in Mexico, Saki et al., (2015) in Iran and Laure et al, (2018) in Cameroon all have reported high rate of *T. gondii* infection about women with history of miscarriages, abortions or still births. Foulon et al., (1999) and Dunn et al., (1999) reported that the risk of congenital transmission increases with gestational age, with the highest rates in the third trimester compared with in the first trimester, although disease severity decreases with gestational age, with first trimester infection resulting in miscarriage. From our study however, more infection was recorded with women at their second trimester, without any significant association with the age of pregnancy otherwise referred to as gestational age. This finding is consistent with that reported by Ballahet al., (2017) and Ogo, (2016). According to Montoya and Remington, (2008), risk of fetal transmission is highest in the second trimester stage of pregnancy; infact Holliman, (1990) placed the risk of fetal transmission at this stage of pregnancy to up 68%, however with less severe consequences to the infected baby. Contrasting report from Nasir et al., (2015) revealed pregnant women at third trimester had the highest IgGseropositivity. Nevertheless, like in our study Nasir et al., (2015) showed no significant difference between anti-toxoplasma antibodies and gestational age.Oocysts excreted by cats could be an important source of contamination. From this study we showed that majority of the women with the *T.gondii* antibodies kept cats at home but there was no established significant association between *T. gondii* infection and history of cat contact. Thus we went further to investigate the role of contact with catfaeces as possible risk for *T. gondii* infection. In our findings, we observed that the infection was more among those women that have had contact with cat faeces even though there was still no significant relationship between the infection and handling of catfaeces. These finding was consistence with studies which was conducted in Mwanza Tanzania (Doehring et al., 1995). However a study from Ethiopia (Zemene et al., 2012) showed significant association between contact with cats and seroprevalence of *T. gondii*. Our study also demonstrated that *T. gondii* infection was not associated with consumption of raw vegetables, handling and consumption of raw/undercooked meat, or specific source of drinking water even though such factors have been found to be significant by Torrey &Yolen, (2013). Furthermore in our study, we were able to establish that blood transfusion history is a risk factor in the transmission of *T. gondii* infection and is consistent with the reports of Robert-Gangneux and Darde, (2012).

## CONCLUSION

The results obtained from this study further illustrate the magnitude of Toxoplasmosis in Nigeria and the consequences it poses on the populace especially the pregnant women. The disease is common

among pregnant women and our study highlighted age, ethnic group and blood transfusion as important factors that could impact on the disease. Other factors such as ignorance, gestation stage, ownership of cats and contacts with their faeces, handling and consumption of raw meat and vegetables amongst others are likely to significantly contribute to the burden of the infection. Consequently it is imperative that policies like including laboratory testing for *T. gondii* infection during antenatal investigations and proper education of the populace especially the pregnant women by Government and relevant stake holder would go a long way to curb *T. gondii* spread.

### Competing Interests

Authors declare that this study was carried out without any existing competing interests.

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