SEROPREVALENCE TOXOPLASMA INFECTION AMONG HIV/AIDS PATIENTS IN KATHMANDU, NEPAL

Bhandari S,¹ KC S,¹ Devkota L,² Khadka S,³ Rai G,⁴ Bastola A,² Rai SK⁴

¹ShiGan International College of Science and Technology, Narayangopal Chok, ²Sukraraj Tropical & Infectious Disease Hospital, Teku, ³HIV Reference Unit, National Public Health Laboratory, Teku, ^{1,4}Nepal Medical College and Teaching Hospital, Attarkhel, Gokarneswor-8, Kathmandu, Nepal

ABSTRACT

Toxoplasmosis is a zoonotic disease caused by *Toxoplasma gondii* affecting about one third of the world's population. It can be asymptomatic to fatal toxoplasmic encephalitis depending on the immune status of infected individuals. Among HIV/AIDS patients, it usually manifest as lifethreatening condition. We, therefore, studied the seroprevalence of *T. gondii* infection among HIV/AIDS patients in Nepal and this report constitutes the first report from Nepal. A total of 45 HIV/AIDS patients were included in this study. The serum samples collected and stored at -20°C were tested for *Toxoplasma* IgG and IgM antibodies at National Public Health Laboratory, Teku, Kathmandu using Snibe Maglumi 1000 Fully Automated Immunoassay Analyzer and the results were expressed in AU/ml. The blood put into the EDTA tube was used for CD4 count using BD FACS Calibur Flow Cytometer. In this study, 33.3% (15/45) HIV infected patients were seropositive for anti-*T. gondii* IgG. However, none of them were positive to anti-*T. gondii* IgM. Most of the patients (36 out of 45 patients) had <200/mm³ CD4 cell count. However, out of them 36.1% (13/36) were seropositive to anti-*T. gondii* IgG whereas 22.2% (2/9) patients with \geq 200 CD4 cell counts had *Toxoplasma* antibodies (p >0.05).

KEYWORDS

Toxoplasma antibody, IgG, IgM, HIV/AIDS patients, Nepal

CORRESPONDING AUTHOR

Mr. Swatantra Bhandari ShiGan International College of Science and Technology, Narayangopal Chowk, Kathmandu, Nepal Email: swatantra.bhandari@outlook.com Orcid No: https://orcid.org/0000-0002-4625-9099 DOI: https://www.doi.org/10.3126/nmcj.v23i1.36236

RESEARCH NOTE

Toxoplasmosis is a global zoonotic disease caused by Toxoplasma gondii (an ubiquitous coccidian parasite that completes life-cycle in two hosts: cat and other feline group of animals as definitive host and animals including birds as intermediate hosts) affecting about one third of the world's population. 1,2 Infection in human takes place by various means, namely, ingestion of oocysts (released in cat feces) through contaminated food materials, ingestions of tachyzoites and bradyzoites present in raw and undercooked meat and meat products, transmissions from blood transfusion and organ transplantation and through transplacental route (mother to fetus). Most infections in human are asymptomatic or appear as flu-like manifestations (fever, malaise, fatigue, muscle pains, sore throat, and headache) with lymphadenopathy; however, severe and fatal condition such as toxoplasmic encephalitis may occur depending on the immune status of infected individuals. Severe opportunistic fatal manifestations occur in HIV/AIDS patients.3 Congenitally transmitted toxoplasmosis results into various irreparable deformities including stillbirth and abortion. It has been estimated that about one third of the world's population is infected with T. gondii with the reported seropositivity rates in different country ranging from less than 10% to over 90%.4 In this short communication, we report the *Toxoplasma* seroprevalence in HIV/ AIDS patients in Nepal.

This study was done in 45 HIV/AIDS patients attending Antiretroviral Treatment (ART) Centre, Teku, Kathmandu (n=41) and Sparsha Nepal, Sanepa, Lalitpur (n=4) during the period of May 2017 to January 2018. Consent was first taken from each patient and 4 ml of blood was taken aseptically and out of which 1 ml was put into EDTA tube and remaining 3 ml was put into plain tube. The serum separated, in the plain test tube, was stored at -20°C until tested. Test was performed at National Public Health Laboratory, Teku, Kathmandu using Snibe Maglumi 1000 Fully Automated Immunoassay Analyzer and the results were expressed in AU/ml. The blood put into the EDTA tube was used for CD4 count using BD FACS Calibur Flow Cytometer. Ethical approval was taken from the Institutional Review Committee of Shi-Gan Health Foundation prior this study.

In this study, 33.3% (15/45) HIV infected patients were seropositive for anti-*T. gondii* IgG. However, none of them were positive to anti- *T. gondii* IgM. There was no significant difference in seroprevalence in males (27.3%)

and female (50.0%) (p >0.05) and same was true with regard to the age of the patients (<40 year: 32.0% and ≥40 years: 35.0%) (p >05) (Table-1). Most of the patients (36 out of 45 patients) had $<200/\text{mm}^3$ CD4 cell count. However, out of them 36.1% (13/36) were seropositive to anti-*T. gondii* IgG whereas 22.2% (2/9) patients with ≥200 CD4 cell counts had *Toxoplasma* antibodies (p >0.05) (Table-2).

Table 1: Gender and age wise distribution of seroprevalence of anti-*T. gondii* IgG (n=45)

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Gender	Total n	IgG Pos n	%	p value
Male	33	9	27.3	p >0.05
Female	12	6	50.0	p >0.05
Age group				
<40 years	25	8	32.0	p >0.05
≥40 years	20	7	35.0	h >0.03

Table 2: Seroprevalence of *Toxoplasma* antibody based on CD4 count

CD4 count	Total n	IgG Pos n	%	p value
<200	36	13	36.1	
≥200	9	2	22.2	p >0.05
Total	45	15	33.3	

The studies conducted in Nepal include the seroprevalence in general population in different places,^{5,6} ranging from 24.0% to 76.1% (with an average of 45.6%),⁷⁻⁹ pregnant women and women with bad obstetric history¹⁰ and selected patients in Kathmandu.¹¹ High seroprevalence in common meat animals (pigs, goats, buffalos and chickens) has been reported¹² and the high seroprevalence in man is associated with the raw meat eating habits of locals.¹³ Among the selected patients, patients with malignancy (cancer) had highest prevalence (68.7%)¹¹ however, till now only one case of congenital toxoplasmosis has been reported.¹⁴

Present study constitutes the first report on seroprevalence of toxoplasmosis among HIV/AIDS patients in Nepal. Though *T. gondii* infects primarily immunocompromised individuals, the seroprevalence among HIV infected patients was found to be less (33.3%) than the average 45.6% of general population. However, this findings was lower or similar to those reported in hospital staffs and apparently healthy individuals from Nepal. On the contrary, present finding was much higher than (9.8%) that reported in HIV positive individuals from

South Africa,¹⁵ Nigeria (10.8%),¹⁶ Western Iran (19.1%)¹⁷ and Eastern China.¹⁸ Present finding was lower than from Mozambique (46.0%)¹⁹ and from Iran (49.7%).²⁰ It was much lower than those reported in HIV/AIDS positive patients from Northwest Ethiopia (76.5%)²¹ and Northern Iran (96.3%).²² In this study, none of the patients were found to be positive for *Toxoplasma* IgM antibody indicating that all IgG antibody patients were not infected recently. In a study done by Mohraj *et al*²⁰ also have shown only 1.0% of the patients had *Toxoplasma* IgM antibodies. In another study, however, 2.3%¹⁸ have been reported to be positive for *Toxoplasma* IgM antibody.

In this study, though, the sample size was small, females were infected marginally higher than in males. This was in agreement with the general population in the country. However, similar prevalence in males and females has been reported in other studies. In this study, the samples size was divided only two groups (40 years and 240 years). The seroprevalence was marginally higher in the age group of 240 years. An increasing rate of seroprevalence with age has also been reported among general population in Nepal. A higher prevalence of infection in older age groups HIV/AIDS patients has also been reported.

The CD4 cell count in this study was lower than those reported from different studies 17,20,21 except one study in which the median CD4 cells count was 25 cells/mm 3 . 23 Most of patients had <200/mm 3 CD4 cell counts. However, just over one third of them (36.1%) had *Toxoplasma* IgG antibodies whereas 22.2% (2/9) patients with ≥ 200 CD4 cell counts had *Toxoplasma* antibodies. Though, CD4 cell count was lower in majority of the patients, none of the patients had symptoms of toxoplasmic encephalitis as has been reported by other investigators. 20,23

This study, showed that one-third of HIV/AIDS patients in Nepal are infected with *T. gondii*. However, none of them showed the signs and symptoms of toxoplasmosis. It is therefore, recommended to conduct a study with a larger sample size in future.

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REFERENCES

- Montoya JG, Leisenfeld O. Toxoplasmosis, Lancet 2004: 12: 363: 1965-76. DOI: 10.1016/S0140-6736(04)16412-X.
- Flegr J, Prandota J, Sovickova M, Israili ZH. Toxoplasmosis - a global threat. Correlation of latent toxoplasmosis with specific disease burden in a set of 88 countries. *Plos One* 2014; 9: e90203; DOI: 10.1371/journal.pone.0090203
- Rezanezhad H, Sayad F, Shadmand E et al. Seroprevalence of Toxoplasma gondii among HIV Patients in Jahrom, Southern Iran. Korean J Parasitol 2017; 55: 99-103. doi: 10.3347/ kjp.2017.55.1.99.
- Pappas G, Roussos N, Falagas ME. Toxoplasmosis snapshots: global status of Toxoplasma gondii seroprevalence and implications for pregnancy and congenital toxoplasmosis. *Int'l J Parasitol* 2009; 39: 1385-94 DOI: http://dx.doi.org/10.1016/j. ijpara.2009.04.003.
- Rai SK, Shibata H, Sumi K et al. Seroepidemiological study of toxoplasmosis in two different geographical areas in Nepal. Southeast Asian J Trop Med Public Health 1994; 25: 479-84.
- Rai SK, Kubo T, Yano K et al. Toxoplasma gondii infection in eastern Nepal- a seroepidemiological study. J Infect Dis Antimicrob Agents 1998; 15: 105-9.
- Rai SK. Toxoplasma, toxoplasmosis and its possible implication as possible opportunistic pathogen in (review paper) Nepal Med Coll J 1999; 1: 81-6. [Google Scholar]
- 8. Rai SK. Toxoplasma Infection in Nepal: an overview. In: Yano A, Nam HW, Anuar AK et al (eds) Asian Parasitology, Toxoplasmosis and Babesiosis in Asia; Edit-in-chief: Yano A, vol 4, Federation of Asian Parasitologists, Japan 2005: 82-96.
- Rai SK. Changing trend of infectious diseases in Nepal. In: Adhikari R., Thapa S. (eds) Infectious Diseases and Nanomedicine III. Advances in Experimental Medicine and Biology, Vol 1052. Springer, Singapore 2018. DOI: https://doi. org/10.1007/978-981-10-7572-8_3
- Rai SK, Shibata H, Sumi K et al. Toxoplasma antibody prevalence in Nepalese pregnant women and women with bad obsteric history. Southeast Asian J Trop Med Public Health 1998; 25: 739-743.
- Rai SK, Upadhyay MP, Shrestha HG. Toxoplasma infection in selected patients in Kathmandu. Nepal. Nepal Med Coll J 2003; 5: 89-91. [Google Scholar]
- 12. Rai SK, Kubo T, Yano K *et al.* Seroprevalence of Toxoplasma gondii infection in common meat animals and its public health importance in Nepal. *J Inst Med (Nepal)* 1996; 18: 55-60. [Google Scholar]
- 13. Rai SK, Matsumura T, Ono K, *et al.* High Toxoplasma seroprevalence associated with meat eating habit of locals in Nepal. *Asia-Pacific J Public Health* 1999; 11: 89-93. DOI: 10.1177/101053959901100207. [Google Scholar]

- 14. Rai SK, Sharma A, Shrestha RK, Pradhan P. First case of congenital toxoplasmosis from Nepal. *Nepal Med Coll J* 2011; 13: 64-6. [Google Scholar]
- 15. Kistiah K, Barragan A, Winiecka-Krusnell J, Karstaedt A and Frean J. Seroprevalence of Toxoplasma gondii infection in HIV-positive and HIV-negative subjects in Gauteng, South Africa. South Afr J Epidemiol Infect 2011; 26: 225-8.
- 16. Amuta EU, Amali O, Jacob SE, Houmsou RS. Toxoplasma gondii IgG antibodies in HIV/ AIDS patients attending hospitals in Makurdi Metropolis, Benue State, Nigeria. *Int'l J Med Biomed Res* 2012; 1: 186-92.
- 17. Rostani A, Keshavarz H, Shojaee S, Mohebali M, Meamar AR. Frequency of Toxoplasma gondii in HIV positive patients from West of Iran by ELISA and PCR. *Iran J Parasitol* 2014; 9: 474-81.
- Wang L, He L, Meng D et al. Seroprevalence and genetic characterization of Toxoplasma gondii in cancer patients in Anhui Province, Eastern China. Parasite Vector 2015; 8: 162. DOI: 10.1186/ s13071-015-0778-5.

- 19. Domingos A, Ito LS, Coelho E, Lúcio JM, Matida LH, Ramos AN Jr. Seroprevalence of Toxoplasma gondii IgG antibody in HIV/AIDS-infected individuals in Maputo, Mozambique. *Revista de Saúde Pública* 2013; 47: 890-6. DOI: 10.1590/s0034-8910.2013047004661
- 20. Mohraz M, Mehrkhani F, Jam S *et al.* Seroprevalence of toxoplasmosis in HIV(+)/AIDS patients in Iran. *Acta Medica Iranica* 2011; 49: 213-8.
- 21. Muluye D, Wondimeneh Y, Belyhun Y *et al.* Prevalence of Toxoplasma gondii and associated risk factors among people living with HIV at Gondar University Hospital, Northwest Ethiopia. *Hindawi Publishing Corporation ISRN Trop Med* 2013; DOI: http://dx.doi.org/10.1155/2013/123858
- 22. Rahimi MT, Mahdavi SA, Javadian B *et al.* High eeroprevalence of Toxoplasma gondii antibody in HIV/AIDS individuals from North of Iran. *Iran J Parasitol* 2015; 10: 584-9.
- 23. Nissapatorn V, Lee C, Quek KF, Leong CL Mahmud R, Abdullah KA. Toxoplasmosis in HIV/AIDS patients: a current situation. *Japanese J Infect Dis* 2004; 57: 160-5.