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Original Article

Screening for the Detection of IgG anti-Toxoplasma gondii Antibodies in Female Donors at Semarang Regency

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ABSTRACT

Toxoplasmosis is a widespread zoonotic disease caused by Toxoplasma gondii (T.gondii). Blood and blood products are possible routes of transmission of T. gondii, especially in patients who have undergone multiple transfusions. The aim of this study is to determine the seroprevalence of toxoplasmosis in female donors using blood serum which is analyzed serologically to detect ImmunoglobulinG (IgG) using the Enzyme-Linked Fluorescent Assay (ELFA) method, as well as the relationship between risk factors for toxoplasmosis, such as keeping cats, receiving blood transfusions, eating undercooked meat and eating raw vegetables. This research is an analytic observational study using a cross-sectional study design. The sample taken is female donor candidates who are willing to participate during the study period, from March to September 2023, and have met the inclusion criteria to become research subjects by explaining the purpose and procedure of the study and the consent letter signed by each research subject before blood samples were taken and filling out the research questionnaire. 177 blood serum samples of female donors were collected from blood donor candidates using consecutive sampling method and a questionnaire to obtain risk factor data that influence the incidence of toxoplasmosis. The results showed that the seroprevalence of toxoplasmosis in the blood of female donor at Indonesian Red Cross Blood Donor Unit (UDD PMI) Semarang Regency, Central Java Province was 26.6%. In this study the risk factors that influence the incidence of toxoplasmosis are consuming raw vegetables with 5.835 times more risk than women who do not consume raw vegetables, while other factors such as keeping cats, receiving blood transfusions, and eating undercooked meat, have no significant effect. The seroprevalence of toxoplasmosis in female donors at UDD PMI of Semarang Regency is quite high due to the habit of eating raw vegetables or fresh vegetables with a very high transmission potential.

Keywords: *Anti-Toxoplasma gondii, Female Donors, IgG Screening.*

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INTRODUCTION

Toxoplasmosis is one of the most widespread diseases and is a zoonotic disease in humans and animals worldwide caused by single cell obligate intracellular opportunistic protozoa belonging to the phylum *Apicomplexa*[1].

Epidemiological surveys show that toxoplasmosis has a global distribution with a highly variable prevalence from 1% to 90% in different regions of the country. It is estimated

that about 25% to 30% of the world's population is chronically infected with Toxoplasma gondii[2,3]. Toxoplasmosis affects one third of the world's population with prevalence rates in developing countries varying between 30-60%[4]. At the global level, the seroprevalence is 40-70% in South American and Caribbean countries. El Salvador, Germany, and France show 75% seropositive Indonesia and Ivory Coast have a prevalence of 60%[5]. There are few reports of voluntary blood donors with positive T. gondii antibodies. In Africa, prevalence varying from 7-80% is observed which drops to 6.4% in South Africa. Namibia recorded a low positivity of 0.96, New Zealand reported 42.9% seroprevalence, China, prevalence ranged from 0.4 to 20.2%, and prevalence of 63.9% in Balinese women (Indonesia)[5]. High seroprevalence in some countries, positive serological tests can be used as an effective method for donor screening[4].

Human toxoplasmosis occurs after birth, through ingestion or appropriate handling of tissue cysts from undercooked or raw meat, consumption of contaminated food (unwashed raw fruit and vegetables), and drinking water contaminated with sporulated oocysts or unpasteurized milk containing decent tachyzoites. Moreover, tachyzoites in the blood can be crossed to the fetus via the placenta when an uninfected woman acquires the infection during pregnancy[2,6].

T. gondii requires two hosts to complete its life cycle: the *Felidae* family as the definitive host and vertebrates such as birds and mammals as the intermediate host[4]. Toxoplasma infection can also be transmitted through blood transfusions and organ transplants[4].

T. gondii which is transmitted from blood donors to recipient patients becomes our concern[7]. Toxoplasmosis in healthy adults is a chronic disease without clinical symptoms or with self-limited symptoms. Although the safety of the blood supply is always under strict control and supervision, toxoplasmosis transmitted by transfusion from asymptomatic donors remains a concern for patients receiving blood transfusions, especially donor recipients with low immunity. *T. gondii* infection lasts a lifetime and most people who are infected are asymptomatic, testing is necessary to identify toxoplasmosis in potential female blood donors[4]. The aim of this study is to determine the seroprevalence of toxoplasmosis in female donors using blood serum.

METHOD

This research is an analytic observational study using a *cross-sectional* study design. The sample taken is female donor candidates who are willing to participate during the study period, from March to September 2023 by explaining the purpose and procedure of the study and the consent letter signed by each research subject before blood samples were taken and filling out the research questionnaire.

The questionnaire given to respondents had been tested for validity and reliability on 30 respondents because the test results were close to a normal curve. The instrument is said to be valid if it has a correlation significance value of 95% or a=0.05. This research has received ethical approval from the Health Research Ethics Committee of the Health Polytechnic of the Ministry of Health, Semarang.

A total sampling of 177 female donor serum samples were collected and aged 18-60 years old. Sample collection was carried out at UDD PMI, Semarang Regency, Central Java Province. The sample examination was carried out at the Health Analyst Department Laboratory of Poltekkes Kemenkes Semarang.

The primary data collection was collected directly by the researcher by carrying out a serological blood serum examination to detect Immunoglobulin G (IgG) using the *Enzyme Linked Fluorescent Assay* (ELFA) method, with a standard assessment of the anti-toxoplasmosis immunoglobulin G (IgG) reaction consisting of 3 (three) conditions, namely , nonreactive (< 4 IU/mL), equivocal (≥ 4 and < 8 IU/mL), and reactive (≥ 8 IU/mL)[8]. Respondents were asked to fill out a questionnaire whose results were used as data on risk factors that influence the incidence of toxoplasmosis.

RESULTS

The total sample in the study were 177 blood serum of female donors, with a donor age range of 18-60 years old, examination of IgG Anti-Toxoplasma using the ELISA method. The results in table 1 showed that 47 people (26.6%) were positive for IgG toxoplasma and 130 people (73.4%) were negative for IgG toxoplasma.

Table 1. Examination results of IgG Toxo ELISA method

Female donors	Frequency	Percent
Positive	47	26.6%
Negative	130	73.4%
Total	177	100%

Statistical analysis was performed using the *Chi-square test*, using categorical data to find out the relationships between variables and risk factors that are significantly related to toxoplasmosis. Then, it was analyzed multivariate, and the level of significance (p) was set at a value limit of 0.05 and a confidence interval of 95%. All statistical analysis was done by software.

Table 2. Test Results for the Relationship Between Risk Factors and Toxoplasmosis Seroprevalence Using the *Chi-Square Test*

Risk factor	Female Donors	
	p-value	**OR (Meaningful)**
Keeping cats	0.184 (No)	1,648
Receiving Blood Transfusions	0.344 (No)	0.725
Eating Undercooked Meat	0.078 (No)	2.008
Eating Raw Vegetables	0.002 (Yes)	5,835

Description: p-value = significance at α <0.05, OR = Odds ratio

Based on table 2, it can be seen that cat owners, a history of blood transfusions and eating undercooked meat are not significantly associated with positive *T.gondii* IgG incidence.

The risk factor that has a significant relationship with toxoplasmosis seroprevalence in female donors (the largest OR) is only in eating raw vegetables with OR=5.835. It can be interpreted that eating raw vegetables is significantly associated with positive IgG *T.gondii* incidence so women who consume raw vegetables increase the risk of positive IgG toxo on average as much as 5.835 times greater than women who do not consume raw vegetables with an increased susceptibility from 1.706 times to 19.963 times.

DISCUSSION

Based on research results, the prevalence of toxoplasma infection in Indonesia ranges from 40-60%, compared to Malaysia which is 42.5%. This prevalence rate is also greater compared to countries in Africa (61.4%), Oceania (38.5%), South America (31.2%) and European countries (29.6%)[9].

The tropical climate of Indonesia which becomes the main breeding ground for the Toxoplasma parasite, which is more commonly found in subtropical and tropical regions, is the reason for this high prevalence. This is because the parasite reproduces sexually in the cat's host, and the oocysts are then excreted from the body along with the feces. Oocysts can survive up to a year in warm, moist soil when fully developed and not exposed to sunlight. Intermediate hosts such as rats, birds, pigs, goats, and humans can be infected with this disease by consuming contaminated adult oocysts[10]. The temperature in Indonesia is considered ideal because toxoplasma oocysts need temperatures between 4 and 37 degrees Celsius to survive[10].

According to the Regulation of the Minister of Health Number 91 of 2015, blood transfusions are only performed on blood that has passed the infectious disease screening test. However, the detection of positive Toxoplasma samples in donor blood indicates the need for additional testing with Anti-Toxoplasma characteristics. In particular, the use of whole blood which must be limited or reduced for donor blood recipients with certain conditions and replaced with leucoreduce blood products to prevent the transmission of toxoplasmosis through opportunistic infections, can be used as a preventive action for the transmission of Toxoplasma gondii through blood transfusions. Immunosuppressed patients, particularly those with HIV, cancer, or organ transplant recipients, are considered particularly susceptible to Toxoplasma gondii[11,12]. However, the risk factors for receiving blood transfusions in this study did not significantly influence positive IgG toxo incidence.

The prevalence of mammals that act as a source of transmission, such as cattle and pets, as well as the tendency to eat undercooked meat and raw vegetables are risk factors for this disease. Environmental hygiene and sanitation issues are also incriminating factors. Pregnant women and those with low immune systems are among the groups of people most at risk of infecting this disease[13]. The risk factors for eating undercooked meat in this study also did not have a significant effect on positive IgG toxo incidence, but a high-risk factor that has a significant effect is eating raw vegetables on the

positive incidence of IgG toxo, so women who consume raw vegetables are 5.8 times more likely to be infected with toxoplasma than women who don't consume it.

Foods that are often consumed in Indonesia that have the potential to carry this dangerous parasite include satay, grilled/grilled chicken, and steak. This food is an example of processed meat that should not be eaten raw. About 33.9% of Indonesia's population consumes grilled food 1-6 times per week. Based on information, it is known that this parasite will die if cooked at 74°C for chicken meat and 71°C for ground beef[10].

Transmission of Toxoplasma gondii through food has traditionally referred to the ingestion of cystic tissue in raw form or undercooked meat[14]. The toxoplasmosis epidemic associated with the consumption of fresh produce or juice describes an acute toxoplasmosis epidemic in an industrial plant in Brazil, which was associated with the consumption of green vegetables[15], identifying 73 cases of acute toxoplasmosis associated with acai juice consumption[16]. It is concluded that the reported cases of toxoplasmosis associated with consumption of salad vegetables may be due to many potential routes of infection for this parasite and the fact that infection is often asymptomatic[13].

Research based on samples collected from donors who were treated at the blood center of the university hospital in Ankara, Turkey in 790 male donors (90.0%), 89 female donors (10.0%), with an age range of 18-65 years old and mean age 34 years. Number of donors with Toxo IgG. (+) as many as 213 (24.2%)[17]. In line with this study, where donor subjects were only women at UDD PMI, Semarang Regency, Central Java Province, out of 177 samples, 47 (26.6%) samples were positive for IgG Toxo.

The seroprevalence rate in healthy blood donors is very high such as in Egypt (59.6%), Kenya (54%), Saudi Arabia (52.1%), North India (51.8%), Czech Republic (32%), Iraq (30%), South India (20.3%), Turkey (19.5%), Southern Iran (12.3%), Taiwan (9.3%), Mexico (7.4%) and Northeastern Thailand (4.1%)[18].

The results of the study reported that the cities with the highest prevalence of toxoplasmosis in Indonesian women were Jambi (60%), Bantul (55.6%), Minahasa (50%) Samarinda (43.5%), and Makassar (40%)[10]. Compared with the results of the prevalence of toxoplasmosis in UDD PMI Semarang Regency (26.6%), it is still below the prevalence of the city mentioned earlier. The seroprevalence of T. gondii in Central Java is 20%[19].

According to the highest risk factors for toxoplasmosis in Indonesian women are the presence of feral cats (OR=14.153), occupation (OR=7.97), habit of eating raw or undercooked meat (OR=5.667), parity status (OR=5.33), not wearing shoes in outdoors (OR=4.684), and raise birds (OR= 0.177)(10). In contrast to the risk factors assessed in this study were keeping cats (OR=1.648), receiving blood transfusions (OR=0.725), eating undercooked meat (OR=2.008), and eating raw vegetables (OR=5.835), indicating risk factors that a significant effect on donor subjects at UDD PMI Semarang Regency is eating raw vegetables. Promotive and preventive measures are needed to fight toxoplasma infection, especially in women who have the habit of eating raw vegetables, environmental sanitation, and poor personal hygiene habits[20].

CONCLUSION

Seroprevalence of toxoplasmosis in female donors in Semarang Regency is quite high (26.6%) this is related to the dietary habit of consuming raw vegetables (fresh vegetables). Possible transmission of infection happens through blood transfusion, while screening tests are not performed to detect toxoplasma in UDD PMI Semarang Regency.

The detection of Toxoplasma samples in donor blood that is positive indicates the need for additional testing with Anti-Toxoplasma characteristics to prevent the transmission of *T. gondii* to people at risk, especially multi-transfusion patients can be used as a preventive measure for the transmission of *T.gondii* through blood transfusions.

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CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

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