

Sexual Behavior and History of Injecting Narcotics Use as Predictors of *Toxoplasma gondii* Infection in Patients with *Human Immunodeficiency Virus Infection*

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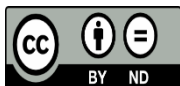


Keywords:

Toxoplasma, infection, serologi, seks, perilaku, risiko

ABSTRACT

Sexual behavior is one of the factors thought to be associated with *Toxoplasma gondii* infection. Several studies have shown that sexual behavior such as oral sex, a history of injuries to the genital organs during sexual intercourse, and working as a sex worker, is associated with *Toxoplasma gondii* infection. In addition, a history of injecting narcotic use is also associated with *Toxoplasma gondii* infection. One method to determine infection status is the serological examination through the measurement of immunoglobulin G (IgG). IgG seropositivity is the basis for determining the status of *Toxoplasma gondii* infection in the chronic or latent phase. This study aims to determine the relationship between sexual behavior and a history of injecting drug use with *Toxoplasma gondii* infection based on IgG serological status. This study's population was 62 Human Immunodeficiency Virus patients in DKI Jakarta, Bogor Regency and City, and Depok City. In this study, the infection status of *Toxoplasma gondii* was measured by ELISA method at the Department of Parasitology, Faculty of Medicine, and University of Indonesia. Sexual behavior and history of injection drug use were measured using a questionnaire. The results showed that 43 samples had positive serology status, 18 had negative serology status, and 1 was in the borderline range. Based on the analysis using logistic regression test, age, education, getting paid for having sex, using condoms during oral sex, the habit of swallowing ejaculated fluid during oral sex, history of injuries to the genital organs during sex, and history of injection drug use are factors Risk of *Toxoplasma gondii* infection.



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1. Introduction

Toxoplasma gondii (*T. gondii*) is a zoonotic intracellular protozoan. Cats are the definitive host for *T. gondii*, while all warm-blooded animals are intermediate hosts [1- 3]. Most *T. gondii* infections originate from ingesting *T. gondii* oocysts, which are found in undercooked livestock meat. In addition, *T. gondii*

ocysts can contaminate the soil, so that people with high-intensity of contact with the soil, such as farmers, and street cleaners, become a vulnerable group [4].

In immunocompetent conditions, *T. gondii* infection does not cause severe problems and symptoms. However, in immunocompromised or immunosuppressed people, *T. gondii* infection can be fatal. Including people with *Human Immunodeficiency Virus* (HIV) infection. *T. gondii* infection is one of the fatal opportunistic infections in HIV patients [5]. Some of these include inflammation of the lining of the brain (meningitis), *encephalitis* (*encephalitis*) [6], and eye infection or *ocular toxoplasmosis* [6].

The hypothesis is developing that transmission of *T. gondii* occurs through sexual intercourse. This hypothesis is based on the route of sexual infection of *T. gondii* in animals. *T. gondii* tachyzoites are found in semen and testicular tissue in humans and some animals, such as goats and pigs. In some animals, such as rabbits, sheep, and dogs, infection in female animals occurs due to the artificial insemination process with semen from male cattle infected with *T. gondii* [7- 9]. Although at this time it is not known whether the appearance of tachyzoites in the semen occurs during the acute or latent phase. For example, the latent phase coincides with the incidence of infection and therapy lowers the body's immunity [10].

Studies from several European countries have shown a correlation between *T. gondii* infection and the incidence of sexually transmitted diseases, especially *gonorrhoea*, *sypilis*, and *chlamydia*. So it appears that the correlation occurs because of the similarity of risk factors, namely unprotected sex behavior [10].

The hypothesis that sexual behavior was associated with *T. gondii* infection was the difference in immunoglobulin G (IgG) levels between the female sex worker group and the control group (OR = 4.05; 95% CI: 1.84–8.89; P = 0.0001). Female sex workers who are at risk of infection with *T. gondii* and have IgG seropositive are women who serve more than 20 customers per week. In addition, a history of wounds experienced during sexual intercourse is also a risk factor for *T. gondii* infection in female sex workers [11].

Proof of sexual transmission of *T. gondii* can only be done indirectly because ethical problems constrain the natural proof process. However, several analyses showed a transmission pattern of *T. gondii* in some pairs. A study revealed that women could contract *T. gondii* from their husbands or male partners. Infected partners are a risk factor for transmitting *T. gondii* in women with heterosexual sexual behavior. But not a risk factor for men with heterosexual sexual behavior. This has led to the hypothesis that transmitting *T. gondii* is associated with oral sex behavior [10].

Men with homosexual orientation have a high prevalence of *T. gondii* infection. IgG seroprevalence in homosexual men reached 30.8%, while IgM seroprevalence was 1.4%. One of the risk factors for *T. gondii* infection is the behavior of using saliva as a lubricant, which is one of the factors associated with *Toxoplasma* infection, with an Odds Ratio (OR) value of 1.6 (95% CI: 0.66-4.12). The population in this study was 143 men with same-sex behavior (MSM) or homosexuals [12].

This study aims to determine whether some sexual behavior is associated with *T. gondii* infection based on IgG serologic status. In this study, several behaviors, such as getting paid during sexual intercourse, the frequency of oral and anal sex, the habit of swallowing ejaculated fluid during sexual intercourse, and injuries to the genitals during sexual intercourse, became the variables studied.

Variables that are proven to be associated with *T. gondii* infection based on IgG serologic status will be predictor variables in the prediction model of *T. gondii* IgG seropositivity in HIV/AIDS patients. Hopefully,

this model can be used to predict and become one of the instruments in screening procedures for *T. gondii* infection in HIV patients.

2. Methods

2.1 Population and Sample

This research has obtained a certificate of *ethical approval* from the Research and Community Health Service Ethics Committee, Faculty of Public Health, University of Indonesia (The *Research And Community Engagement Ethical Committee, Faculty of Public Health, University of Indonesia*). The population in this study were HIV patients who were actively undergoing *Anti Retrovirus* (ARV) treatment at health facilities in the DKI Jakarta Province, Bogor Regency and City, and Depok City.

The inclusion criteria in this study are:

1. People living with HIV carry out HIV examination, care and treatment at health centres, hospitals, and health service centers located in DKI Jakarta Province, Depok City, and Bogor City during the study period.
2. 18 – 59 years old
3. Willing to undergo serological examination for *T. gondii antibodies*
4. Willing to be a research respondent.
5. Have CD4 levels above 350 cells/mm³

While the exclusion criteria in this study are:

1. HIV patients have blood or vascular disorders, so taking blood is impossible.
2. People with HIV are at stages 3 and 4, according to the World Health Organization (WHO).
3. Have had cotrimoxazole prophylactic therapy.
4. Is pregnant

The research sample comprised 62 people who met the inclusion and exclusion criteria. All samples underwent the process of taking blood through a vein. The amount of blood taken from each respondent ranged from 2.5 - 3cc. The blood specimen was then transferred into a 3 cc EDTA tube. Then the blood plasma specimen was taken to the Parasitology Laboratory, Faculty of Medicine, and University of Indonesia.

2.2 Research methods

Measurement of IgG *T. Gondii* using the ELISA method. The result of measuring IgG levels of *T. gondii* is in the form of an index measure in IU per ml. *T. gondii* infection was declared positive if the index value was 10 IU/ml. Meanwhile, *T. gondii* infection was declared negative if the index was <8 IU/ml. If the index value is in the range 8 < to < 10 it is declared as borderline. In this study, there was 1 sample that was in the borderline range, so it was not included in the sample group being analyzed.

Measurement of sexual behavior and history of using narcotics and injecting dangerous objects (drugs) as predictors of *T. gondii* infection was carried out using a questionnaire instrument. The analytical test used is a Linear Regression analysis test, using the IBM SPSS version 27 application with a license validity period until December 31, 2037.

3. Results and Discussion

3.1 Results

Based on serological testing using the ELISA method, 43 (69.4%) had positive *T. Gondii* IgG serology, 18 (29.4%) had negative serology, and 1 (1.6%) were in borderline conditions. One sample in the borderline condition was not analyzed, so the data analysis was carried out on 61 samples.

This study's gender, age, educational status, and socioeconomic status were suspected as risk factors for *T. gondii* infection based on IgG serological status. To prove whether these four variables are risk factors and are associated with *T. gondii* infection status based on IgG serological status, statistical tests were carried out using logistic regression tests.

The educational status variable consists of 4 categories. Namely the status of low education (SD), junior secondary education (SMP), senior secondary education (SMA), and higher education (college). At the same time, the socioeconomic status variable consists of 4 categories. Namely poor, lower middle, middle, and upper middle. Classification of research respondents into a socioeconomic classification based on ownership of 13 valuables or assets. Namely household energy sources, refrigerators, air conditioners, water heaters, wireless internet networks (WiFi), gold, computers, motorcycles, cars, flat-screen televisions more than 32 inches, land/land/garden ownership, electric power, and sources of drinking water.

In this study, the relationship between risk factors and *T. gondii* infection status was considered significant if the p-value was less than (0.05). In addition, the logistic regression test can also calculate the value of the Odds Ratio (OR). Where the OR is more significant than 2 indicates a relationship between the risk factor variables and the infection status of *T. gondii*. This study's confidence interval range with an accuracy of 95% (95% CI) was also considered. This value is one of the parameters to determine the existence of random error caused by the sampling process.

Based on demographic background, age is one of the variables significantly associated with *T. gondii* infection status. Respondents less than 28 years old have a 10.11-fold higher risk of being infected with *T. gondii* than those who are more than 39 years old.

In addition, high school education (high school graduation) is also a risk factor for *T. gondii* infection. Where respondents with a high school education level, the risk of infection with *T. gondii* is 4 times higher than respondents with higher education (college graduate).

Table 1 Demographics of Respondents

Variable	Positive IgG (n=43)	Negative IgG (n=18)	OR (95% CI)	
Gender	Man	35	16	1.37 (0.13 – 14.23)
	Woman	5	1	0.60 (0.03 – 13.58)
	she male	3	1	1
Age	<28 Years	9	7	10.11 (1.05 – 97.02)*
	28-34 Years	14	5	4.64 (0.48 – 45.21)
	34-39 Years	7	5	9.29 (0.90 – 95.95)
	>39 Years	13	1	1
Education	Low education	4	1	1.54 (0.14 – 17.33)
	First Secondary Education	13	5	0.50 (0.36 – 7.00)
	High School Education	16	2	4.00 (0.38 – 42.37)
	higher education	10	10	1
Socio-	Poor	11	3	0.82 (0.11 - 6.34)

Economic Status	Middle down	16	9	1.69 (0.28 – 10.17)
	Intermediate	10	4	1.20 (0.67 – 8.66)
	Middle to Upper	6	2	1

*=Significant at (0.05)

Sexual behavior that is a risk factor associated with the status of *T. gondii infection* based on IgG serologic status is a history of injuries to the genital organs (penis/vagina) during sexual intercourse. However, in this study, the OR value could not be calculated because there was 1 empty cell in the respondent's IgG *T. gondii seronegative condition*.

Based on the calculation of the OR value, HIV patients who get paid for having sex (sex workers) have 4.5 times the risk of being infected with *T. gondii* compared to HIV patients who are non-sex workers. However, judging from the 95%CI value, there is a reasonably wide range, which is probably caused by *sampling*.

In addition, sexual behavior that is a risk factor for *T. gondii* infection is never using condoms during oral intercourse (OR: 5.33 95% CI: 0.56 – 51.27); and behavior of swallowing ejaculated fluid.

Table 2. Sexual Behavior and History of Injecting Drug Use

Variable	Positive IgG (n=43)	Negative IgG (n=18)	OR (95% CI)
Getting Paid for Having Sex	Yes	9	4.50 (0.53 – 38.50)
	Not	34	1
Using Condoms During Oral Sex	Always Use Condoms	6	4.00 (0.32 – 48.66)
	Frequent use of condoms	4	4.00 (0.27 – 58.56)
	Rarely Use Condoms	13	2.46 (0.23 – 26.11)
	Never Use a Condom	12	5.33 (0.56 – 51.27)
	Never Have Oral Sex	8	1
Frequency of Swallowing Ejaculate Fluid	Always Swallowing Ejaculate Fluid	2	N/A
	Frequently Swallowing Ejaculate Fluid	1	N/A
	Rarely Swallow Ejaculate Fluid	9	8.89 (0.92 – 85.66)
	Never Swallow Ejaculate Fluid	23	2.44 (0.26 – 22.97)

	Never Have Oral Sex	8	1	1
Injured genital organs during sexual intercourse	Yes	8	0	N/A*
	Not	35	18	
History of Injecting Drugs	Yes	4	0	N/A
	Not	39	18	

*=Significant at (0.05)

N/A= Not Available – because there are empty cells

Once known, several risk factors associated with *T. gondii* infection, then performed a multivariate analysis using logistic regression test. This analysis aims to produce a predictive model of *T. gondii* infection based on IgG serological status.

The 7 risk factors analyzed to produce this predictive model are; age, education, getting paid for typing sex, using condoms during oral sex, habit of swallowing ejaculated fluid during oral sex, history of injuries to genital organs during sex, and history of injection drug use.

The results of the logistic regression test yielded a Chi-square value of 16.628 with a p-value of 0.020. So it can be concluded that there is a significant relationship between the 7 risk factors analyzed with *T. gondii* infection based on IgG serological status. Based on the Nagelkerke R Square value, the result is 0.339. This value concludes that the magnitude of the influence of the seven risk factors on *T. gondii* infection is 33.9%.

Prediction model of *T. gondii* infection based on IgG serologic status is as follows:

$$L_n \frac{P}{1-P} = -84,513 - 0,612 * X1 + 0,366 * X2 + 0,928 * X3 + 0,285 * X4 + 0,064 * X5 + 20,489 * X6 + 19,967 * X7$$

Where:

- X1 = Respondent Age Group
- X2 = Respondent's Education Level
- X3 = Getting Paid During Sex
- X4 = the Habit of Using Condoms during Oral Sex
- X5 = Frequency of Swallowing Ejaculate Fluid
- X6 = History of Injury to the Sex Organs during Sex
- X7 = History of Injecting Drug Use

And each score for each category in each risk factor variable is as follows:

Variable	Category	Score
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Age (X1)	<28 Years	1
	28-34 Years	2
	34-39 Years	3
	>39 Years	4
Education (X2)	Low education	1
	First Secondary Education	2
	High School Education	3
	higher education	4
Getting Paid During Sex (X3)	Yes	1
	Not	0
Condom Use During Oral Sex (X4)	Always Use Condoms	1
	Frequent use of condoms	2
	Rarely Use Condoms	3
	Never Use a Condom	4
	Never Have Oral Sex	0
Frequency of Swallowing Ejaculate Fluid (X5)	Always Swallow Ejaculate Fluid	4
	Frequently Swallowing Ejaculate Fluid	3
	Rarely Swallow Ejaculate Fluid	2
	Never Swallow Ejaculate Fluid	1
	Never Have Oral Sex	0
Injured Sex Organs During Sexual Intercourse (X6)	Yes	1
	Not	0
History of Using Injecting Drugs (X7)	Yes	1
	Not	0

3.2 Discussion

This study proves that sexual behavior is a risk factor for *T. gondii* infection based on IgG serologic status. This is in line with the discovery of *T. gondii* cysts in reproductive tissue. *T. gondii* tissue cysts can be found in semen and ejaculate in men based on a study conducted by Disco et al. in 1971. Evidence of sexual transmission of *T. gondii* can only be done indirectly because ethical problems constrain the natural proof process. However, several analyses have shown a transmission pattern of *T. gondii* in some pairs. Hlaváčová J, in his analysis entitled, "Male to Female Presumed Transmission of Toxoplasmosis between Sexual Partners" reveals that women can contract *T. gondii* from their husbands or male partners. In the analysis, the authors stated that an infected partner was a risk factor for transmitting *T. gondii* in women with heterosexual sexual behavior. But not a risk factor for men with heterosexual sexual behavior. These results prove that *T. gondii* can be transmitted from infected men to their partners [10]. The notion that *T. gondii* can be transmitted by men to their partners was revealed in studies with a population of homosexual men.

This research is also following a study conducted on 143 homosexual men. This study found that men with a homosexual orientation had a high prevalence of *T. gondii* infection. IgG seroprevalence in homosexual men reached 30.8%, while IgM seroprevalence was 1.4%. In this study, oral sex behavior, in the form of using saliva as a lubricant, was one of the factors associated with *Toxoplasma* infection, with an Odds Ratio (OR) value of 1.6 (95% CI: 0.66-4.12) [12].

The hypothesis of sexual transmission of *T. gondii* stems from evidence in animals. In a study in Brazil, sexual transmission of *T. gondii* was identified from natural mating behavior in sheep. When mating with a male, a female sheep are infected with *T. gondii*, and transmits the infection to the fetus in her womb [13]. In an experimental study in dogs, *T. gondii* was detected in the testes and epididymal tissue by

immunohistochemical examination. In this study, *T. gondii* can be transmitted through sexual intercourse, which impacts the transmission of *T. gondii* to their offspring [14].

This study also showed a relationship between sexual behavior and *Toxoplasma gondii* infection in a population of 1,865 individuals in the Czech Republic. This study identified several risk factors associated with *Toxoplasma gondii* infection in the Czech Republic. The prevalence of *Toxoplasma gondii* in women in this study was 30.1% (434/1443), while the prevalence of *Toxoplasma gondii* in men was 20.1% (85/422) [15].

In this study, statistical analysis was performed using the Spearman Correlation test to determine the relationship between risky sexual behavior and the incidence of *Toxoplasma gondii* infection. *Risky sexual behavior* is defined as having sex with multiple partners without a condom. Based on the analysis results, there was a significant relationship between risky sexual behavior and the incidence of *Toxoplasma gondii* infection, with a p-value of <0.01. However, based on the analysis of stratification by sex, a significant relationship between risky sexual behavior and *Toxoplasma gondii* infection only occurred in women, with p<0.005. At the same time, the analysis of the relationship between risky sexual behavior and *Toxoplasma gondii* infection in men showed insignificant results, with a p of 0.96.

The results of multivariate analysis using the Logistics Regression test show the Odds Ratio (OR) value of 1.22 with 95% *Confidence Interval* of 0.79 - 1.90. So it can be seen that people with risky sexual behavior have a risk of infection with *Toxoplasma gondii* 1.22 times higher than people who do not have risky sexual behavior. Based on the analysis of stratification by sex, it is known that the OR value for women is 1.38. At the same time, the OR for men is 0.83. Based on the results of the analysis, although not statistically significant, the suspicion of a relationship between risky sexual behavior and *Toxoplasma gondii* infection is open [15].

There is a study to determine the factors associated with *Toxoplasma gondii* infection in people with a mental health condition who are hospitalized at the Durango City Mental Hospital, Mexico. To diagnose *Toxoplasma gondii* infection is, serological examination to detect Immunoglobulin G (IgG) and Immunoglobulin M (IgM). The serological examination showed that 18.2% (25/17) of people with a mental health condition had *Toxoplasma gondii* infection, which was diagnosed based on the detection of IgG in blood serum. Based on multivariate analysis using the Logistic Regression test, it is known that *sexual promiscuity* is a risk factor with an OR value of 15.8 with a 95% *Confidence Interval* of 3.8 to 64.8. The results of this analysis indicate that a hypothesis regarding the relationship between sexual behavior and *Toxoplasma gondii* infection can be developed [16].

Another evidence of an association between sexual behavior and *T. gondii* infection is a case-control study using a study population of 408 women. The case group in this study were 136 women who worked as commercial sex workers (CSWs), in Durango City, Mexico. The analysis showed that *Toxoplasma gondii* infection was associated with status as a CSW. The calculation of the OR value produces a relationship strength of 4.05 with a 95% *Confidence Interval* of 1.84 - 8.89. These calculations indicate that the incidence of *Toxoplasma gondii* infection in CSWs is 4.05 times higher than in women who are not CSWs. [11].

This study reveals that the respondent's habit of serving more than 20 customers per week has an OR value of 2.76 with a 95% *Confidence Interval* of 0.6 - 11.00. These results indicate that CSWs who serve more than 20 customers per week have a 2.76 times higher risk of *Toxoplasma gondii* infection than CSWs who

serve 20 customers or less. This study's results align with the intensity of sexual intercourse and working as sex workers or getting paid when having sex, which is one of the risk factors for *Toxoplasma gondii* infection.

Toxoplasma gondii infection was a study of 119 couples who underwent health consultations provided by the Mexican Ministry of Health. This study aimed to determine the similarity of infection status (*concordance*) between the 119 couples. *Toxoplasma gondii* infection was diagnosed using blood serum examination to determine IgG and IgM levels. The results showed that 40 out of 119 couples, both husband and wife, showed positive IgG titers. Furthermore, there were 12 couples where both husband and wife had high IgG titers (>150 IU/ml). However, in the analysis results using the Kappa Index calculation method, the degree of *concordance* between 119 couples was low [17].

The theory put forward to explain the low concordance is because each partner only has sex with his partner. Thus, it is hypothesized that sexual transmission of *Toxoplasma gondii* is related to the intensity of sexual intercourse with several partners. As the results of research from [16], [15]. In addition, the low concordance was caused because all individuals in this study did not have immune problems. So the chance of *Toxoplasma gondii* cysts in the genital organs is low. This study suspects that sexual transmission of *Toxoplasma gondii* occurs when *Toxoplasma gondii* is in the reproductive organs. In individuals without immune problems (immunocompetent), the risk of the presence of *Toxoplasma gondii* is low. This results in a lower risk of transmission of *Toxoplasma gondii* infection through sexual intercourse in immunocompetent individuals.

Immunocompromised status due to the influence of infection with Human Immunodeficiency Virus (HIV) and Acquired Immunodeficiency Syndrome (AIDS) is associated with *Toxoplasma gondii* in the reproductive organs. A literature study revealed that *Toxoplasma gondii* was found in the testes of HIV/AIDS patients [18], [19]. A case study reported that *Toxoplasma gondii* was found in the testes of an immunocompetent man [20]. The evidence that *Toxoplasma gondii* can develop in the testes strengthens the notion that *Toxoplasma gondii* infection is related to sexual intercourse. Especially in individuals with high sexual intensity, multiple partners, and who do not use condoms.

In a study involving 143 homosexual men, it was found that the incidence of *T. gondii* infection was associated with injecting narcotic use (OR=3.8, 95% CI: 2.413-5.996; aOR=3.1, 95% CI: 1.905-5.093, $p < 0.001$) [21]. The use of injection narcotics is associated with *T. gondii* infection because blood cells are a predilection for the parasite. Tachyzoites and sporozoites can be found in blood cells such as mononuclear cells and leukocytes [22].

4. Conclusion

This study showed age, education, getting paid for typing sex, using condoms during oral sex, the habit of swallowing ejaculated fluid during oral sex, history of injuries to the genital organs during sex, and history of injection drug use. This study also reveals that the magnitude of the influence of the seven risk factor variables is 33.9%. This study also produced a predictive model of *T. gondii* infection status based on IgG serologic status.

However, further research with a larger sample size is needed to increase the validity of the research results. Further research also needs to include eating behavior and clean and healthy living behavior (PHBS) to increase the magnitude of the influence of risk factors on *T. gondii* infection status based on IgG status.

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