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ORIGINAL PAPER

Seroprevalence and Associated Risk Factors for *Toxoplasma gondii* Infections Among Urban Poor Communities in Peninsular Malaysia

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Abstract

Purpose Toxoplasmosis is a zoonotic infection linked to compromised hygiene and sanitation via the handling of infected cat faeces, eating undercooked contaminated meat or transplacental transmission. We conducted a study to determine seroprevalence and risk factors associated with toxoplasmosis among the urban poor communities in Malaysia.

Methods The demographic profiles for each participant were obtained through a questionnaire survey prior to blood collection. A total of 389 participants were recruited and blood samples screened for the presence of anti-*Toxoplasma* IgG and IgM antibody using an ELISA commercial kit, SERION ELISA classic *Toxoplasma gondii* IgG and IgM.

Results The overall *T. gondii* seroprevalence was 69.6% with 56.8% seropositive for anti-*Toxoplasma* IgG, 7.7% seropositive for anti-*Toxoplasma* IgM and 5.1% seropositive for both IgG and IgM antibodies. The presence of both antibody classes in blood samples indicated high avidity, suggesting latent infection. Univariate analysis revealed significant associations that included; age, ethnicity, location and employment status while, significant lifestyle factors included source of drinking water and eating style. A multifactorial statistical model that incorporated all the significant effects from the first-stage univariate analyses listed above revealed that age and ethnicity were the two dominant and independent effects on IgG seroprevalence. For seroprevalence of IgM, the multifactorial model revealed a significant interaction between work and accommodation. IgM seroprevalence was higher among the unemployed inhabitants of PPR (Program Perumahan Rakyat) than those living in non-PPR accommodation, and higher than among the employed irrespective of their accommodation.

Conclusion High seroprevalence of Toxoplasmosis in the community calls for increased awareness of disease transmission and improvements in hygiene and sanitation.

Keywords Toxoplasmosis · *Toxoplasma gondii* · Seroprevalence · Urban poor communities · Malaysia

Introduction

Toxoplasmosis is caused by the ubiquitous intracellular protozoan parasite, *Toxoplasma gondii,* and has been recognized as one of the most common diseases affecting up to one third of the world population [5, 12]. The capacity of *T. gondii* for invasion of both animal and human hosts emphasizes its medical and veterinary importance as the causative agent of toxoplasmosis [4, 34]. Human transmission occurs via ingestion of food or water contaminated with oocysts shed from infected cat faeces; consumption of undercooked or raw meat, oysters, clams, or mussels containing tissue cysts [1, 13, 17], exposure to contaminated soil through activities such as gardening or children playing in sandpits [29] and vertical transmission from mother to foetus [11].

Most human infections are asymptomatic. Severe clinical symptoms such as general malaise, fever and lymphadenopathy occurring only rarely. The risk of developing clinical illness is higher particularly, for the unborn foetus in *Toxoplasma*-infected pregnant women, immunosuppressed patients following tissue/organ transplantation, those infected with HIV or undergoing cancer treatment [44]. The

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standard method for diagnosis is via serological testing, and currently mostly based on the detection of antibodies for *Toxoplasma*-specific immunoglobulin IgG and IgM antibodies by the ELISA method [20, 25]. Other methods previously applied have included, indirect hemagglutination (IHA) test, Sabin-Feldman dye test and indirect fluorescent antibody test [45].

Toxoplasma infection is common in populations where hygiene and sanitation are compromised. Serological surveys in Malaysia have provided evidence that this parasite is widespread, with inconsistent incidences among groups with different demographical profiles [23]. This includes healthy individuals (13.9–30.2%) [35], pregnant women (23.0–31.6%) [16, 30], HIV patients (21.0%-41.2%) [25–28], newborn babies (2.0%) [38], indigenous communities (10.6%-37.0%) [22], animal handlers and farmers (19.9–27.0%) [2], schizophrenia patients (51.0%) [14], hospital patients (44.12%) [18] and migrant workers (5.2–57.4%) [32].

Malaysia has undergone recently cumulative growth of urbanization from 43.2% (1989) to 76% (2018) resulting in an urban population boom from 27% (1970) to 71% (2010) with the incidence of urban poverty on a downward trend over the past three decades from 16.5% in 1970 to 1.0% in 2012 [33, 36]. The population boom over such a short time span has created various problems for local governments especially, in fulfilling provision of services, housing and infrastructures and creating job opportunities for urban dwellers [6]. Inadequate amenities and a shortage of housing have led to deterioration of the living standards in the urban population and increased diseconomies, e.g., environmental deterioration, pollution, congestion, diseases, squatters, improper dumping of garbage and mismanagement in sanitation services [19]. These factors underlying poverty have been highlighted also by UNICEF [41] which concluded that 99.7% of children living in low-cost flats in Kuala Lumpur live in poverty and 7% in absolute poverty.

With this background, the current study was conducted to determine the seroprevalence of toxoplasmosis in the states of Malacca, Selangor and Wilayah Persekutuan, Kuala Lumpur and to highlight intrinsic, extrinsic and lifestyle factors associated with infection in the targeted population.

Materials and Methods

Study Population and Ethical Consideration

This study was carried out between October 2016 and March 2017 among selected urban poor communities living in villages in town, care homes and low-cost high-rise flats developed by the National Housing Department of Selangor and Wilayah Persekutuan Kuala Lumpur or commonly known as People's Housing Project (Program Perumahan Rakyat or PPR) (Fig. 1). A total of 389 inhabitants participated as part of a community well-being program. The inclusion criteria for the urban poor cohort were based on non-monetary aspects and focused on the standard of accommodation and living areas. Participants were asked to complete a questionnaire which provided relevant information on each subject including socio-demographics, environmental health, lifestyle habits, eating habits and any recent illnesses. All participants were fully informed about the nature of the study and completed consent forms.

This study was approved by the Medical Ethics Committee of the University of Malaya Medical Centre (UMMC) (MECID NO: 20143-40). Completed, signed consent forms were obtained from each participant prior to screening, while consent for children was obtained from their guardians in a similar manner before the collection of samples.

Samples Collection

Approximately 5 mL of blood were drawn by trained medical personnel and placed into a blood collection tube, BD Vacutainer for serum (red top cap without anticoagulant). The collected blood samples were immediately placed into an ice box and subsequently transported to the Parasitology Laboratory, Faculty of Science, University of Malaya. Blood samples were spun at 1500 × g for 10 min and the serum samples were kept at − 20 °C until further use.

Detection of Immunoglobulin G and M Antibodies to *T. gondii*

The presence of anti-*Toxoplasma gondii* immunoglobulin G (IgG) and immunoglobulin M (IgM) in the serum was screened for using commercial kits comprising the standard enzyme-linked immunosorbent assay (ELISA), the SERION ELISA classic *Toxoplasma gondii* IgG and IgM (IgG-Institut Virion/Serion, Germany and IgM-Institut Virion/Serion, Germany) following the manufacturer's instruction.

For the IgG assay, a positive result was defined by readings > 20 IU/ml, indicating latent or pre-existing *Toxoplasma* infection and > 350 IU/ml indicating recent infection for the IgM assay. Samples that were both IgG-positive and IgM-positive were tested using an IgG avidity assay (IgG; Institut Virion/Serion, Germany) according to the manufacturer's instruction and the level of avidity was then calculated using a SERION evaluation spread sheet prepared on demand.

Statistical Analysis

All data obtained from both questionnaire and laboratory assays were entered, edited and analysed statistically using the software SPSS version 22. Summary data are provided

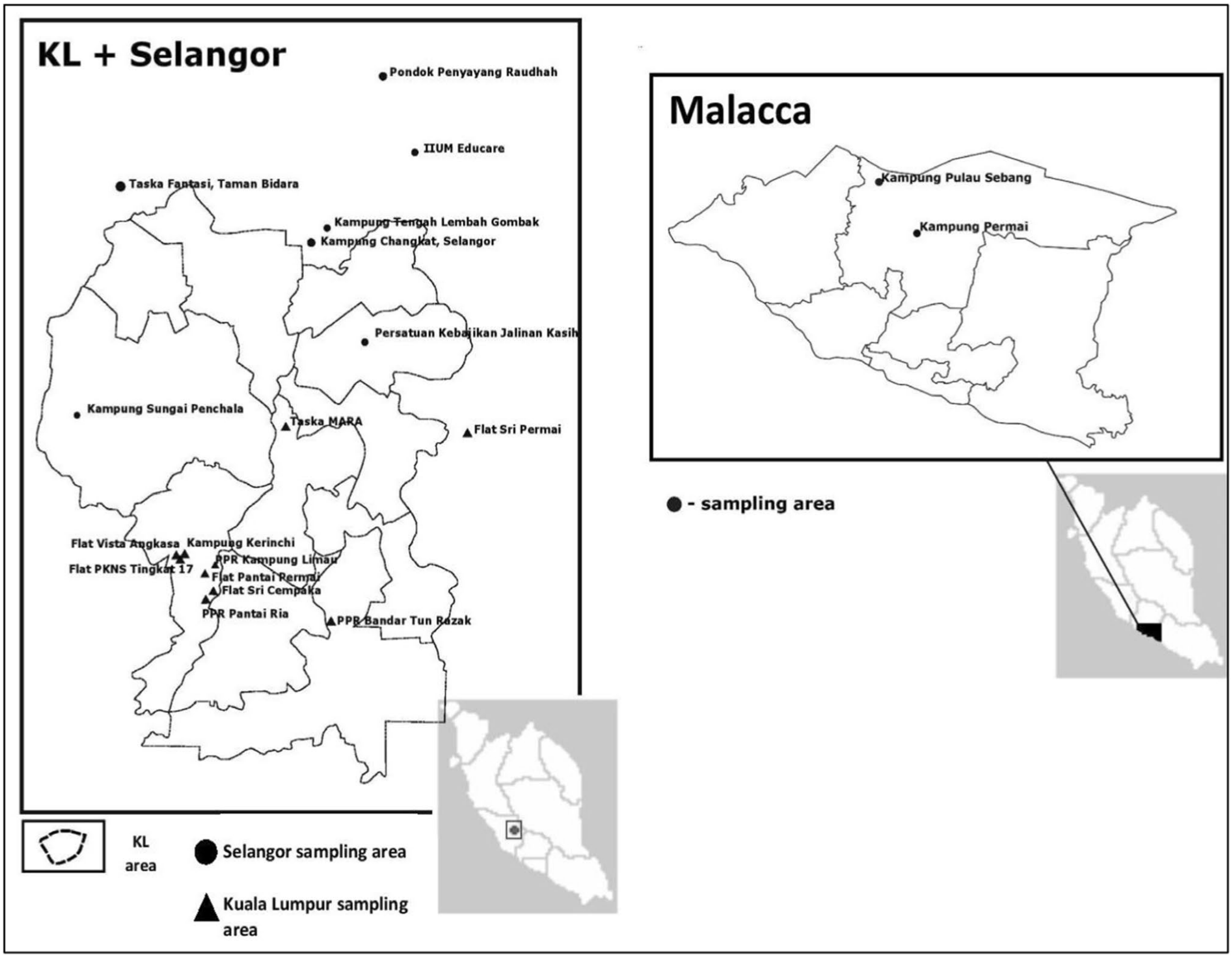

Fig. 1 Study locations

for prevalence (percentage infected in relevant factor levels) plus the 95% confidence limits calculated by bespoke software based on the tables of Rohlf and Sokal [31]. We provide also odds ratios + 95% confidence limits calculated by bespoke software for levels within each factor, using one level as the reference point in each case.

Analysis was conducted using maximum likelihood techniques based on log linear analysis of contingency tables. In the first phase univariate models were fitted with either presence/absence of IgG or IgM and one of the following factors in turn: sex (males and females), age (seven age classes comprising those < 12 years old, 12–18 years old, 18–25 years old, 25–34 years old, 35–44 years old, 45–54 years old and > 55 years old) and ethnicity (Malay, Chinese, Indian and others). Extrinsic factors included; location (Kuala Lumpur, Selangor and Malacca), types of accommodation (flats and other types of settlements), employment status (employed and unemployed) and education attainment (primary, secondary, tertiary and no formal education). The lifestyle habits recorded included; food intake (home cooked or other sources), source of drinking water (boiled or water filtered), meat consumption (yes or no), eating habit with hand (yes or no), hand washing frequency (four levels: none, less than three times, three to five times, more than five times) and pet ownership (no pets, cats, dogs and rabbits).

For each level of analysis in turn, beginning with the most complex model, involving all possible main effects and interactions, those combinations that did not contribute significantly to explaining variation in the data were eliminated in a stepwise fashion beginning with the highest-level interaction (backward selection procedure in SPSS). Finally, in a third step, multifactorial models were fitted comprising all the intrinsic and extrinsic factors that had been found to be statistically significantly associated with infection status in the initial univariate phases of analysis.

Results

Socio-Demographic Characteristics

A total of 389 volunteers were recruited, with slightly more than half (56.0%) females, and the rest males (44.0%). The majority were aged between 35 to 44 years (19.5%), followed by 25 to 34 years (16.2%), less than 12 years (16.2%), 55 years old or older (15.7%), 19 to 24 years (13.1%), 45 to 54 years (12.1%) and 12 to 18 years (7.2%). The study cohort comprised predominantly Malays (82.0%), followed by Indians (11.1%), Chinese (6.7%) and a Sikh (0.3%). Based on location, most participants resided in Kuala Lumpur (52.4%), followed by Selangor (33.9%) and Malacca (13.6%). The majority inhabited low-cost PPR flats (54.2%) compared to other types of settlements (45.8%), were not employed (58.4%) with most educated to secondary level (52.2%) followed by primary level (22.6%) and tertiary level (19.8%) while 21 participants (5.4%) had no formal education.

Seroprevalence of *T. gondii*

The overall seroprevalence for *T. gondii* was 69.7% with 56.8% seropositive for anti-*Toxoplasma* IgG only, 7.7% for anti-*Toxoplasma* IgM only and 5.1% for both IgG and IgM antibodies (Table 1). Samples with both IgG and IgM antibodies showed high avidity, suggesting latent infection.

Risk Factors Associated with the Seroprevalence of *T. gondii* Infections

Seropositivity of *T. gondii* was analysed statistically in relation to sociodemographic factors. In the first univariate phase of analysis only two intrinsic factors were found to be significantly associated with IgG seropositivity to *T. gondii*, i.e., age ($X^2_6 = 45.389$, $P < 0.001$) and ethnicity ($X^2_3 = 27.577$, $P < 0.001$) (Table 2). Seroprevalence was significantly higher among Malays and those aged > 12 years old. Analyses for anti-*Toxoplasma* IgM antibody and combination with both IgG and IgM antibodies did not show any association with any of the three intrinsic factors.

Of the four extrinsic factors considered, only one factor was found to be significantly associated with seropositivity for *T. gondii* IgG, i.e., location namely, Kuala Lumpur ($X^2_2 = 23.537$, $P < 0.001$) (Table 2). Analyses for anti-*Toxoplasma* IgM showed that the residents from PPR flats ($X^2_1 = 6.995$, $P < 0.05$) and those unemployed ($X^2_1 = 4.789$, $P < 0.05$) were significantly associated with infection (Table 3).

Of the lifestyle factors considered, only two factors were found to be significantly associated with seropositivity for *T. gondii* IgG, i.e., source of drinking water ($X^2_1 = 8.200$, $P < 0.05$) with higher seroprevalence among those consuming filtered drinking water and routinely eating using hands ($X^2_1 = 8.718$, $P < 0.05$) (Table 2). However, analyses for seropositivity based on anti-*Toxoplasma* IgM, and both IgG and IgM antibodies, showed that none of these factors were significantly associated with infection (Tables 3 and 4).

Finally, in a multifactorial model for IgG seroprevalence in which we fitted only the significant effects from the initial univariate phase of analysis (age, ethnicity, location, source of drinking water and eating habits with *T. gondii* IgG seropositivity), two factors retained significance namely; age ($X^2_6 = 39.58$, $P < 0.001$) and ethnicity ($X^2_3 = 19.875$, $P < 0.001$). However, the analysis did not find any significant interactions between these factors, indicating that they were independent effects on seroprevalence (Table 5). For IgM seroprevalence, we fitted type of accommodation, employment status and hand washing frequency, and the model revealed only one significant interaction (type of accommodation × employment status; $X^2_1 = 4.18$, $P = 0.042$). As Fig. 2 shows IgM seroprevalence was higher among the unemployed living in PPR accommodation than among unemployed living in non-PPR accommodation. A post hoc analysis of IgM seroprevalence restricted to the unemployed subjects ($n = 227$), confirmed that prevalence was significantly higher among those in PPR accommodation ($X^2_1 = 8.37$, $P = 0.004$). The type of accommodation made no difference to IgM seroprevalence for the employed and was relatively low (< 5%) and similar to that of the unemployed subjects living in non-PPR accommodation.

Table 1 Overall seroprevalence of IgG and IgM antibodies to *Toxoplasma gondii* among urban poor communities ($N = 389$)

Antibodies	*Toxoplasma gondii* seropositive		
	No. of seropositive	Seropositive (%)	95% confidence limits (%)
IgG + only	221	56.8	50.8–62.7
IgM + only	30	7.7	5.1–11.6
IgG + IgM +	20[a]	5.1	3.0–8.5
Total	271	69.7	63.9–74.9

[a]All 20 volunteers positive for both IgG + and IgM + were found to have latent infections based on the avidity test

Discussion

Urban poverty is a dynamic condition arising from weaknesses in the management of cities. Despite the decline of the overall incidence of poverty in Malaysia, rapid urbanization has created pockets of urban poor communities. In

Table 2 Potential risk factors on socio-demographic and lifestyle factors associated with IgG seropositivity of *T. gondii* among the studied population (univariate analysis; $N=389$)

Factors	%	95% CL	OR (95% CI)	*P* value
Socio-demographic factors				
Sex				
Male (*n* = 171)	57.3	48.2, 66.1	1.037 (0.345, 0.517)	0.39285
Female (*n* = 218)	56.4	51.9, 60.9	1	
Age class (years)				
12–18 (*n* = 28)	60.7	40.9, 77.1	5.944 (3.698, 9.790)	**0.00064**
19–24 (*n* = 51)	60.8	50.7, 70.0	5.962 (3.361, 7.706)	**0.00005**
25–34 (*n* = 63)	57.1	46.1, 67.6	5.128 (2.797, 6.151)	**0.00010**
35–44 (*n* = 76)	64.5	52.4, 75.2	6.980 (3.748, 8.096)	**0.00000**
45–54 (*n* = 47)	63.8	46.1, 78.7	6.787 (3.893, 9.129)	**0.00002**
> 55 (*n* = 61)	73.8	63.3, 82.3	10.817 (6.126, 14.123)	**< 0.00001**
< 12 (*n* = 63)	20.6	12.9, 30.9	1	
Ethnicity				
Malay (*n* = 319)	62.7	57.3, 67.8	4.342 (2.194, 4.436)	**0.00009**
Chinese (*n* = 26)	30.8	15.4, 50.6	1.148 (0.753, 2.188)	0.38629
Others/Sikh (*n* = 1)	100	5.0, 100.0		
Indian (*n* = 43)	27.9	15.1, 44.7	1	
Location				
Kuala Lumpur (*n* = 204)	66.2	61.9, 70.3	4.524 (2.173, 4.179)	**0.00001**
Selangor (*n* = 132)	53.0	45.1, 60.9	2.611 (1.287, 2.536)	**0.00858**
Malacca (*n* = 53)	30.2	21.4, 40.4	1	
Accommodation				
PPR (*n* = 211)	58.8	54.3, 63.2	1.190 (0.395, 0.590)	0.27859
Non-PPR (*n* = 178)	54.5	45.1, 63.5	1	
Employment				
Employed (*n* = 162)	61.7	52.9, 69.8	1.413 (0.476, 0.717)	0.10206
Unemployed (*n* = 227)	53.3	48.7, 57.9	1	
Education attainment				
Secondary (*n* = 203)	60.6	56.2, 64.8	1.684 (0.667, 1.104)	0.05126
University (*n* = 77)	57.1	45.0, 68.6	1.460 (0.671, 1.242)	0.19279
No formal education (*n* = 21)	57.1	35.4, 76.7	1.460 (0.901, 2.354)	0.29592
Primary (n = 88)	47.7	34.7, 60.8	1	
Lifestyle factors				
Food				
Other sources (*n* = 45)	60.0	42.7, 75.6	1.160 (0.544, 1.025)	0.35909
Home cooked (*n* = 344)	56.4	50.8, 61.9	1	
Source of drinking water				
Water filter (*n* = 92)	69.6	56.2, 80.5	2.038 (0.801, 1.319)	**0.00800**
Boil (*n* = 297)	52.9	47.6, 58.1	1	
Meat preparation				
Fully cooked (*n* = 380)	56.8	50.9, 62.6	1.054 (0.775, 2.932)	0.39776
Half cooked (*n* = 9)	55.6	25.1, 83.1	1	
Eating habits				
Hands (*n* = 351)	59.3	53.6, 64.8	2.797 (1.413, 2.854)	**0.00655**
Others (*n* = 38)	34.2	20.7, 49.9	1	
Daily hand washing frequency				
None (*n* = 3)	66.7	13.5, 98.3	1.625 (1.493, 18.362)	0.37127
Less than three times (*n* = 15)	73.3	46.6, 90.3	2.234 (1.660, 6.457)	0.20357
More than five times (*n* = 342)	56.1	50.5, 61.6	1.040 (0.555, 1.189)	0.39692
Three to five times (*n* = 29)	55.2	36.0, 72.8	1	

Table 2 (continued)

Factors	%	95% CL	OR (95% CI)	*P* value
Pet ownership				
None (*n* = 340)	57.1	51.5, 62.5	1.329 (1.144, 8.216)	0.38333
Cat (*n* = 44)	56.8	39.7, 72.6	1.316 (1.146, 8.893)	0.38543
Rabbit (*n* = 4)	50.0	9.8, 90.2	1	

PPR Program Perumahan Rakyat

the country, poverty is measured quantitatively using the Poverty Line Index (PLI) to differentiate between the poor and non-poor households. More precisely, urban poor are defined as households with an income of less than RM940 (Ringgit Malaysia 940) per month (or approximately USD 220) [3] and unable to fulfil the minimum basic necessities. These include non-monetary indicators such as; food, education, health, housing, possession of household appliances and availability of amenities [6, 36].

Overcrowding and poor living conditions can contribute to the spread of diseases associated with poor sanitation and hygiene. Moreover, individuals may be less informed of the benefits of a healthy lifestyle in addition to having restricted access to quality healthcare. The UN 2030 Agenda [42] for sustainable development promotes improvements in health and well-being of all ages in a population, particularly for women and children.

In view of this, a well-being community program was conducted among selected urban poor residents in the vicinity of Kuala Lumpur, Selangor and Malacca. The seroprevalences of *T. gondii* with anti-*Toxoplasma* IgG and IgM antibodies revealed that more than half of the study population had experienced infection. Seroprevalence values based on anti-*Toxoplasma* IgG antibodies have been found to vary worldwide. In Southeast Asian countries, seroprevalence varies between < 2% up to 70% [23] while studies from high-income countries, such as USA and UK, have reported between 10 and 40% [37]. Meanwhile, values for seroprevalence from Central and South America and continental Europe have ranged from 50 to 80% [13]. This is not surprising as human infection is widely reported, with nearly one-third of the world population chronically infected.

Seroprevalence of anti-*Toxoplasma* IgG antibodies in the current study was marginally higher (56.8%) compared to previous local studies (13.9–30.2%) [35]. In addition, 30 volunteers (7.7%) were positive for specific IgM but negative for IgG antibodies, indicating possible acute infection. Two intrinsic factors (those aged > 12 years old and Malay ethnicity) showed particularly significant associations with *T. gondii* infection, and were the only two factors that were retained in the final model when all other factors that had shown significance in initial univariate analyses, had been taken into account. The higher seroprevalence particularly among older individuals is generally in agreement with previous studies [22, 24, 39] and the overall view that the cumulative probability of exposure to infection increases with age. A study among indigenous communities (Orang Asli) showed significantly higher seroprevalence ($P \leq 0.001$) among those aged 12 years and older (70/133; 52.6%), compared to younger participants (113/362; 31.2%) [22]. While a study among veterinary personnel (veterinarian, technicians and students) and pet owners in the area of Klang Valley also identified the age group ≥ 30 years old, as more likely to have been exposed to *Toxoplasma* infection [2]. Recently, it has been suggested that toxoplasmosis infection among older individuals may also be transmitted sexually via the ejaculate of infected men to uninfected men/women [7, 8]. Evidence also suggests that there is a high risk of infection from unprotected oral sex when the infected ejaculate is swallowed [15].

Malays recorded the highest seroprevalence in the current study. This finding was also in agreement with other local studies [35, 40] and studies conducted in South East Asia including Indonesia [9] and Singapore [43]. One possible explanation is that Malays are more inclined to keep cats as pets compared to other animals and therefore more likely to be exposed to oocysts deposited by cats [21, 22]. However, the results of this study may be biased as most participants recruited were Malays. Brandon-Mong et al., [2] recorded higher prevalence among Indians (29.0%) and rationalized that Indians compared to other ethnic groups work more closely with animals. Chinese were least exposed to infection and similar observations have been reported also in Singapore (Chinese: 16- 22%,Malay: 20.1–39.0%; Indian: 19.3–37.0%) and Indonesia (Chinese: 2.3–7.0%; Indonesian: 14.3–18.0%) [10].

Significant relationships were also observed in univariate analyses of IgM seroprevalence among the residents inhabiting low-cost PPR flats and those who were unemployed. These findings were substantiated in a multifactorial model, which showed clearly that IgM seroprevalence

Table 3 Potential risk factors on socio-demographic and lifestyle factors associated with IgM seropositivity of *T. gondii* among the studied population (univariate analysis; $N=389$)

Factors	%	95% CL	OR (95% CI)	*P* value
Socio-demographic factors				
Sex				
Female ($n=218$)	8.3	6.0, 11.2	1.193 (0.635, 1.356)	0.35983
Male ($n=171$)	7.0	3.6, 13.2	1	
Age class (years)				
< 12 ($n=63$)	12.7	6.8, 21.7	4.436 (3.533, 17.358)	0.07416
12–18 ($n=28$)	3.6	0.2, 17.5	1.130 (1.031, 11.868)	0.39704
19–24 ($n=51$)	7.8	3.7, 15.2	2.596 (2.140, 12.186)	0.22391
35–44 ($n=76$)	5.3	1.7, 13.7	1.694 (1.394, 7.876)	0.33382
45–54 ($n=47$)	12.8	4.6, 28.6	4.463 (3.605, 18.744)	0.08204
> 55 ($n=61$)	8.2	3.7, 16.3	2.723 (2.215, 11.881)	0.20142
25–34 ($n=63$)	3.2	0.8, 9.6	1	
Ethnicity				
Malay ($n=319$)	7.5	5.1, 10.9	2.034 (1.770, 13.634)	0.31626
Indian ($n=43$)	11.6	4.2, 26.4	3.289 (2.927, 26.565)	0.22792
Chinese ($n=26$)	3.8	0.2, 18.8	1	
Location				
Kuala Lumpur ($n=204$)	8.8	6.6, 11.7	1.728 (1.027, 2.531)	0.19683
Malacca ($n=53$)	9.4	4.8, 17.1	1.860 (1.297, 4.285)	0.23761
Selangor ($n=132$)	5.3	2.7, 10.1	1	
Accommodation				
PPR ($n=211$)	10.9	8.3, 14.0	2.989 (1.738, 4.152)	**0.01919**
Non-PPR ($n=178$)	3.9	1.5, 9.3	1	
Employment				
Unemployed ($n=227$)	10.1	7.6, 13.3	2.496 (1.452, 3.471)	**0.04803**
Employed ($n=162$)	4.3	1.8, 9.5	1	
Education attainment				
Primary (n=88)	10.2	4.5, 21.0	2.079 (1.465, 4.964)	0.19983
Secondary ($n=203$)	7.4	5.3, 10.1	1.456 (0.988, 3.077)	0.32327
No formal education ($n=21$)	9.5	1.7, 30.5	1.921 (1.594, 9.367)	0.30728
University ($n=77$)	5.2	1.7, 13.7	1	
Lifestyle factors				
Food				
Other sources ($n=45$)	11.1	3.8, 26.3	1.595 (1.017, 2.806)	0.26570
Home cooked ($n=344$)	7.3	4.8, 10.8	1	
Source of drinking water				
Boil ($n=297$)	8.4	5.9, 11.9	1.599 (1.005, 2.705)	0.25900
Water filter ($n=92$)	5.4	1.5, 15.2	1	
Meat preparation				
Half cooked ($n=9$)	11.1	0.6, 44.3	1.513 (1.330, 11.005)	0.37058
Fully cooked ($n=380$)	7.6	5.0, 11.5	1	
Eating habits				
Others ($n=38$)	13.2	5.5, 27.2	1.976 (1.267, 3.530)	0.17084
Hands ($n=351$)	7.1	4.7, 10.7	1	
Daily hand washing frequency				
None ($n=3$)	33.3	1.7, 86.5	7.643 (6.977, 80.110)	0.10513
Less than three times ($n=15$)	20.0	5.7, 46.6	3.821 (2.821, 10.773)	0.05833
Three to five times ($n=29$)	17.2	7.1, 36.0	3.185 (2.081, 6.006)	**0.04023**
More than five times ($n=342$)	6.1	3.9, 9.4	1	
Pet ownership				

Table 3 (continued)

Factors	%	95% CL	OR (95% CI)	*P* value
Dog (*n* = 1)	100	5.0, 100.0	1.688 (1.079, 0.2434)	0.24034
Cat (*n* = 44)	11.4	4.0, 26.4		
None (*n* = 340)	7.1	4.7, 10.5	1	

PPR Program Perumahan Rakyat

Table 4 Potential risk factors on socio-demographic and lifestyle factors associated with IgG and IgM seropositivity of *T. gondii* among the studied population (univariate analysis; *N* = 389)

Factors	%	95% CL	OR (95% CI)	*P* value
Socio-demographic factors				
Sex				
Female (*n* = 218)	6.4	4.5, 9.0	1.887 (1.178, 3.132)	0.17751
Male (*n* = 171)	3.5	1.3, 8.5	1	
Age class (years)				
< 12 (*n* = 63)	9.5	4.7, 18.0	6.316 (5.579, 47.788)	0.09698
19–24 (*n* = 51)	3.9	1.3, 9.8	2.449 (2.233, 25.368)	0.30731
25–34 (*n* = 63)	3.2	0.8, 9.6	1.967 (1.793, 20.307)	0.34361
35–44 (*n* = 76)	5.3	1.7, 13.7	3.333 (2.971, 27.295)	0.22652
45–54 (*n* = 47)	10.6	3.3, 26.3	7.143 (6.338, 56.234)	0.08399
> 55 (*n* = 61)	1.6	0.2, 7.3	1	
Ethnicity				
Malay (*n* = 319)	5.3	3.4, 8.3	1.407 (1.228, 9.608)	0.37836
Indian (*n* = 43)	4.7	0.7, 17.6	1.220 (1.114, 12.934)	0.39395
Chinese (*n* = 26)	3.8	0.2, 18.8	1	
Location				
Selangor (*n* = 132)	5.3	2.7, 10.1	1.086 (0.683, 1.842)	0.39363
Malacca (*n* = 53)	5.7	2.4, 12.3	1.164 (0.855, 3.225)	0.38903
Kuala Lumpur (*n* = 204)	4.9	3.3, 7.2	1	
Accommodation				
PPR (*n* = 211)	7.1	5.1, 9.8	2.648 (1.705, 4.788)	0.07228
Non-PPR (*n* = 178)	2.8	0.8, 7.7	1	
Employment				
Unemployed (*n* = 227)	6.2	4.2, 8.8	1.709 (1.067, 2.837)	0.22423
Employed (*n* = 162)	3.7	1.4, 8.6	1	
Education attainment				
Primary (n = 88)	8.0	3.0, 17.9	3.241 (2.588, 12.852)	0.14186
Secondary (*n* = 203)	4.9	3.3, 7.2	1.943 (1.527, 7.134)	0.27926
No formal education (*n* = 21)	4.8	0.3, 23.3	1.875 (1.713, 19.867)	0.35158
University (*n* = 77)	2.6	0.5, 9.9	1	
Lifestyle factors				
Food				
Other sources (*n* = 45)	8.9	2.5, 23.5	2.000 (1.362, 4.271)	0.19679
Home cooked (*n* = 344)	4.7	2.8, 7.6	1	
Source of drinking water				
Water filter (*n* = 92)	5.4	1.5, 15.2	1.080 (0.699, 1.977)	0.39472
Boil (*n* = 297)	5.1	3.2, 7.9	1	
Meat preparation				
Half cooked (*n* = 9)	11.1	0.6, 44.3	2.375 (2.093, 17.600)	0.29058
Fully cooked (*n* = 380)	5.0	3.0, 8.3	1	
Eating habits				

Table 3 (continued)

Factors	%	95% CL	OR (95% CI)	*P* value
Others (*n* = 38)	7.9	2.3, 20.5	1.684 (1.214, 4.348)	0.28953
Hands (*n* = 351)	4.8	2.9, 7.9	1	
Daily hand washing frequency				
Less than three times (*n* = 15)	20.0	5.7, 46.6	5.450 (4.061, 15.932)	**0.02076**
Three to five times (*n* = 29)	6.9	1.2, 22.1	1.615 (1.264, 5.818)	0.33016
More than five times (*n* = 342)	4.4	2.6, 7.3	1	
Pet ownership				
Cat (*n* = 44)	9.1	2.7, 23.6	2.025 (1.380, 4.331)	0.19209
None (*n* = 340)	4.7	2.8, 7.7	1	

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Table 5 Maximum likelihood log linear analysis of contingency table including all significant effects from the univariate analyses

Step	Effect	Chi-square	Degrees of freedom	Significance *p*
35	IgG × location	0.210	2	0.900
42	IgG × Eating	0.505	1	0.477
47	IgG × drinking	0.572	1	0.450
48	Final minimum sufficient model			
	IgG × ethnicity	**19.875**	**3**	**< 0.001**
	IgG × age	**39.580**	**6**	**< 0.001**
	Location × drinking	49.830	2	< 0.001
	Location × age	127.582	12	< 0.001
	Location × ethnicity	224.326	6	< 0.001
	Drinking × eating	26.993	1	< 0.001
	Drinking × age	28.704	6	< 0.001
	Eating × location	84.314	2	< 0.001
	Eating × ethnicity	80.393	3	< 0.001
Goodness of fit of the final model to the data		104.543	616	0.99

The initially fitted model was IgG × location × eating[a] × drinking[b] × ethnicity × age. At each step, based on backward stepwise selection, the least significant interaction between fitted effects was eliminated to eventually generate the minimum sufficient model. We show only the steps at which interactions with IgG were eliminated and their probabilities at that stage, and then all remaining effects in the final model, plus the overall goodness of fit of the final model to the data

[a]Eating = eating habit with hands or other

[b]Drinking = source of drinking water

was higher among the unemployed subjects living in PPR accommodation compared to those living in non-PPR accommodation and among the employed irrespective of their accommodation. The unemployed in PPR have higher prevalence of IgM probably due to more of them having time to keep cats as pets. The Ministry of Housing & Local Government in Malaysia initially established PPR to fulfil the need for low cost housing, following demolition of squatter dwellings previously occupied by lower income groups. However, this type of settlement is vastly overcrowded with poor waste management and poor civic consciousness. A decline in services and attitude have impacted the overall quality of life of residents especially through exposure to a range of communicable and non-communicable diseases [33]. Bad waste management purportedly in these developments, with rubbish collected once a month only, has driven residents to dispose trash over their balconies to the surrounding areas, attracting rats and stray animals to roam and scavenge below.

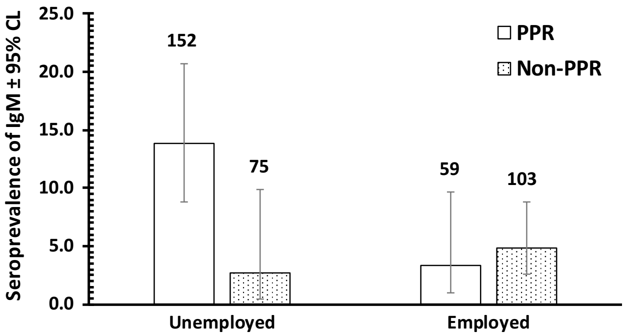

Fig. 2 IgM seroprevalence among employed and unemployed subjects living in PPR (Program Perumahan Rakyat) and non-PPR accommodation. The sample size for each group is given above the relevant column

In addition, bad attitude and low educational background are also responsible for the perpetuation of disease transmission through little or lack of awareness of good hygiene practices. A significant relationship was recorded among residents who consumed filtered water and preferred eating with their hands in the initial univariate analyses. Transmission of these parasitic protozoans can occur via contaminated water supplies particularly through the consumption of filtered water rather than drinking boiled water, and lack of cleanliness during eating is also likely to predispose individuals to infection, but when we controlled for other factors, these two were not retained in the minimum sufficient model. Many other studies have also linked the presence of toxoplasmosis with environmental and socioeconomic conditions namely; eating habits, health related practices, hygiene levels, host susceptibility, geographical location (geolatitude) and humidity of the soil [7, 8, 39]. However, when our analysis focused on the significant effects from the first univariate stage analyses, age and ethnicity turned out to be the two strongest and independent risk factor for IgG seroprevalence.

Therefore, our results call for the implementation of a health education program not only for the urban poor communities, but also for the general public that incorporates promotion of knowledge on parasite transmission. This includes awareness of toxoplasmosis and risk of infection particularly in relation to host age, ethnicity and of the need for care in handling cat faeces as well as the health risks associated with consumption of contaminated meat and water. Efforts should be made to encourage improvements in personal hygiene prior to consumption of food and fluids and thorough cooking of meats [32]. Overall improvements are also necessary for better provision of facilities and services particularly among residents in PPR flats.

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Compliance with ethical standards

Conflict of interest On behalf of all authors, the corresponding author states that there is no conflict of interest.

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