PREVALENCE OF ANTIBODIES TO TOXOPLASMA GONDII AMONG URBAN AND RURAL RESIDENTS IN THE PHILIPPINES

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Abstract. A survey of antibodies to Toxoplasma gondii was carried out among residents in urban (Metro Manila) and rural (Mindoro and Leyte) areas in the Philippines. A total of 1,173 serum samples were examined for Toxoplasma antibody by an ELISA method. The overall seropositivity was 11.1% (n=904, 12.4% in males, 10.0% in females) in Metro Manila, 61.2% (n=152, 63.3% in males, 53.1% in females) in Mindoro, and 30.1% (n=113, 34.3% in males, 22.5% in females) in Leyte, indicating significantly higher (p<0.001) seropositivities in rural than urban settings. No significant differences in seropositivities were observed between males and females. In each group, seropositivity tended to increase with age of the subjects.

INTRODUCTION

Toxoplasma gondii is an intracellular protozoan parasite of medical importance. This parasite is ubiquitously distributed throughout the world, and antibodies specific for Toxoplasma are observed in humans at various rates in almost all nations (Remington and Desmonts, 1990). Especially, old surveys in some human populations reported extremely high prevalence rates of approximately 90%. Once infected, humans cannot eliminate the parasite, and specific antibody persists throughout life. Therefore, antibody prevalence rates indicate the percentage of humans who harbor Toxoplasma parasites (Bearman et al, 1995).

Toxoplasma gondii causes severe congenital infections in infants and acute infections in immunosuppressed patients, although almost all cases of Toxoplasma infections are asymptomatic. When a woman acquires the first infection during pregnancy, the parasite may cause intra-uterine infection and eventually lead to developmental abnormality including hydrocephalus and mental retardation in her offspring. On the other hand, the recent increase in populations of acquired immunodeficiency syndrome (AIDS) patients have defined a significant opportunistic feature of Toxoplasma infection, since cerebral infection caused by pre-existing Toxoplasma parasites has been a major cause of death in AIDS patients in Europe and the USA (Porter and Sande, 1992). The increasing number of AIDS patients in a population with a high Toxoplasma infection rate may raise serious public health concern. This is especially true in the Asian region, which has one of the fastest growing populations of HIV/AIDS patients in the world (WHO, 1996). However, there have been few reports on how many people harbor Toxoplasma parasites in Asian countries. In this study, we surveyed the prevalence of Toxoplasma antibodies among rural and urban residents in the Philippines, where no report has been published in these past 22 years (Cross et al, 1977).

MATERIALS AND METHODS

Study population

A total of 1,173 serum samples were used in the study (Table 1). These samples were collected from 908 urban residents in Metro Manila (Barzaga, 1996), and 152 and 113 local residents in Mindoro and Leyte, respectively. Mean ages (±standard
deviations) of these residents were 27.0 (±15.1) years in Metro Manila, 28.3 (±17.6) years in Mindoro, and 28.6 (±16.5) years in Leyte: there were no significant differences in age composition among three populations (p>0.05). Subjects of Metro Manila belonged to upper middle income families in which 40% are students and 20% were employees. These populations in Metro Manila were composed of residents from northeastern (39%), southwestern (39%), and central parts (22%). The locations of Metro Manila, Mindoro and Leyte are shown in Fig 1. These serum samples were stored at -20ºC until tested.

**ELISA**

*Toxoplasma* antibody levels in human sera were measured by ELISA as previously described (Konishi and Takahashi, 1983) with some modifications (Khin-Sane-Win et al, 1997). Briefly, 96-well, flat-bottom non-ELISA microplates were sensitized with *Toxoplasma* crude antigen prepared from tachyzoites of the RH strain. These plates were incubated with a 1:100 dilution of test sera, and then with alkaline phosphatase-labeled anti-human IgG. The enzyme activity in each well was measured with *p*-nitrophenyl bisodiumphosphate. Each absorbance value was adjusted with the value obtained with the constant positive control serum to minimize interplate variations. Sera showing the adjusted values (ELISA values) of more than 0.357 were determined to be positive for *Toxoplasma* antibody (Konishi and Takahashi, 1983).

**Statistical analysis**

Significance of differences was evaluated by the chi-square test for seropositivities and the Student’s *t*-test for mean ages. Probability levels (p) of less than 0.05 were considered significant.

**RESULTS**

**Comparison among Metro Manila, Mindoro and Leyte**

The seropositivity in the total population of each location was 11.0% in Metro Manila, 61.2% in Mindoro and 30.1% in Leyte (Table 2). There were significant differences between each of two rural areas (Mindoro and Leyte) and the urban area (Metro Manila) in age-matched populations (p<0.001).

Both in males and females, seropositivity in Mindoro was the highest (63.3% in males and
Table 1

Number of subjects used in this survey.

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Metro Manila</th>
<th>Mindoro</th>
<th>Leyte</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>Female</td>
<td>Male</td>
<td>Female</td>
</tr>
<tr>
<td>1-9</td>
<td>55</td>
<td>72</td>
<td>4</td>
</tr>
<tr>
<td>10-29</td>
<td>197</td>
<td>203</td>
<td>61</td>
</tr>
<tr>
<td>30-49</td>
<td>122</td>
<td>193</td>
<td>35</td>
</tr>
<tr>
<td>50-</td>
<td>21</td>
<td>41</td>
<td>20</td>
</tr>
<tr>
<td>Total</td>
<td>395</td>
<td>509</td>
<td>120</td>
</tr>
</tbody>
</table>

Table 2

Prevalence of *Toxoplasma* antibody among residents in Metro Manila, Mindoro and Leyte.

<table>
<thead>
<tr>
<th>Location</th>
<th>Seropositivity (No. positive/ no. tested)</th>
<th>Total (No. positive/ no. tested)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metro Manila</td>
<td>12.4% (49/398) a,b</td>
<td>11.1% (100/904) b,h</td>
</tr>
<tr>
<td>Mindoro</td>
<td>63.3% (76/120) a,c</td>
<td>61.2% (93/152) f,i</td>
</tr>
<tr>
<td>Leyte</td>
<td>34.2% (25/73) b,c</td>
<td>30.1% (34/113) h,i</td>
</tr>
</tbody>
</table>

Table 3

Prevalence of *Toxoplasma* antibody among residents in subareas of Metro Manila.

<table>
<thead>
<tr>
<th>Location</th>
<th>Seropositivity (No. positive/ no. tested)</th>
<th>Mean age±SD (years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Northeastern a</td>
<td>9.3% (32/341) d,f</td>
<td>27.6±15.0 d</td>
</tr>
<tr>
<td>Southwestern b</td>
<td>11.5% (42/350) e,c</td>
<td>24.5±15.4 c,b</td>
</tr>
<tr>
<td>Central c</td>
<td>13.0% (26/213) e</td>
<td>29.8±14.0 c</td>
</tr>
</tbody>
</table>

53.1% in females) followed by Leyte (34.2% in males and 22.5% in females) and Metro Manila (12.4% in males and 10.0% in females). No significant differences were observed between males and females in each area (Table 2) (p>0.05).

Comparison among three subareas in Metro Manila

Table 3 shows the prevalence of *Toxoplasma* antibodies among urban residents in three subareas of Metro Manila (northeastern, southwestern and central parts). No significant differences in seropositivity were observed among populations in these subareas. Although the southwestern population had a significantly lower mean age than other populations, age-adjusted seropositivities of the northeaster (8.3% at 24.5 years) and Central (11.3% at 24.5 years) populations were not significantly different from the seropositivity of the southwestern population (11.5% at 24.5 years; p>0.05), assuming a yearly increase of seropositivity in Manila population to be 0.32% based on the data shown in Fig 2 (see below: 5% at 5 years and 21% at 55 years).

Age-dependent prevalence

In Metro Manila, prevalence of *Toxoplasma* antibody increased with age (Fig 2). Statistically significant differences were observed among all age groups (p<0.05) except between groups of 1-9 years and 10-29 years, and between groups of 30-49 years and 50 years or more. In each age group, the differences between males and females were not significant.

In both Mindoro and Leyte, the pattern of increasing seropositivity (Fig 3) was similar to that observed in the population of Manila. However, the differences among age groups were not sig-
significant due to the relatively high seropositivity of the age group of 1-9 years and lower seropositivity at 50 years or more than 30-49 years.

DISCUSSION

This study revealed a high prevalence of Toxoplasma antibody in Filipino populations, especially those in rural settings. Toxoplasma infection usually occurs asymptptomatically in immunocompetent hosts, and the diagnosis depends mainly on serological tests. We have surveyed Toxoplasma antibody in Japanese populations by the ELISA method, and revealed overall prevalence of 10-20% in rural populations (Konishi and Takahashi, 1987). Since the same diagnostic method was used in this study, Toxoplasma antibody prevalence in the Philippines can be compared with that obtained in Japan.

Serological surveys of Toxoplasma antibody in Asian countries have been performed mainly by indirect hemagglutination assays (IHA). The previous survey carried out in the Philippines 22 years ago (Cross et al, 1977) was done in North Samar, which is located in the center of Philippine Archipelago, close to one of our study areas, Leyte. This survey revealed an overall seropositivity of 2.5% (32 positive individuals out of 1,274 samples). In Taiwan, where the location is close to the Philippines, seropositivity varied from 1.9% to 18% (Cross and Hsu, 1989). In other Asian countries, the prevalence of Toxoplasma antibody varied from 4.6% to 38.8% in Malaysia (Yahtaya, 1991), from 2% to 63% in Indonesia, (Gandahusada, 1991), from 2.3% to 13.4% in Thailand. In Nepal, a survey done by latex agglutination and ELISA revealed seropositivities from 38% to 68% (Rai et al, 1996).

Toxoplasma is one of the pathogens which show the highest prevalence among humans in the temperate zone where most other parasites were eradicated. Recently, we compared age-dependent prevalence between two surveys at the same location with an interval of approximately 10 years in Japan (Khin-Sane-Win et al, 1997). The result indicated that the prevalence rate obtained in each age group at the first survey did not significantly increase at the second survey in the corresponding age group, suggesting a relatively low transmission rate during recent years. On the other hand, the present survey in rural areas in the Philippines revealed high seropositivities in young age groups of 1-9 years, suggesting high transmission rates. Surveys among humans in some other tropical and subtropical countries also indicated high seropositivities in infant groups (Remington et al, 1970). In tropical and subtropical regions, it appears that toxoplasmosis has not been a major concern because of a high incidence with other parasitic diseases. However, toxoplasmosis is now considered as one of the most important tropical and subtropical diseases.

Mechanisms of transmission of Toxoplasma to humans other than congenital transmission are (1) accidental ingestion of the oocyst defecated by cats and (2) ingestion of the cyst contained in infected raw meat. Variations of seropositivities among previous surveys are based on several factors: geographic, climatic, hygienic and socioeconomic factors, as well as lifestyles of the human population. These factors are closely related to the above two transmission mechanisms. In the present study, seropositivities in the rural area (61.2% in Mindoro and 30.2% in Leyte) were higher than that in the urban area (11.0% in Manila). Since people in the Philippines do not have the habit of consuming raw meat, it is highly probable that accidental ingestion of oocysts is the primary route of transmission in Filipinos. Therefore, the difference in seropositivity between rural and urban areas may be due to the hygienic and socioeconomic status that relates to oocyst shedding by cats and peoples' contact with the soil. Specifically, the pavement widely seen in the urban area is considered to contribute to the reduced surviving period of oocysts shed by cats. The relation of hygienic conditions to the prevalence of infectious diseases is also observed in epidemiological studies on hepatitis A in the Philippines (Barzaga et al, 1996). The persistent exposure to the contaminated environment could be a risk factor in acquisition of such infectious diseases.

In conclusion, a survey of antibodies to Toxoplasma gondii in the Philippines has shown that higher seropositivities were observed in the rural population, suggesting hygienic conditions and exposure to a contaminated environment is associated to the prevalence of Toxoplasma gondii.

REFERENCES


