Epidemiological Report

High Prevalence of Antibody to *Toxoplasma gondii* among Humans in Surabaya, Indonesia

Eiji Konishi*, Yumi Houki, Kazuyoshi Harano**, Retno S. Mibawani, Djoko Marsudi, Soetrisno Alibasah and Yoës P. Dachlan

Department of Health Sciences, Kobe University School of Medicine, Tomogaoka 7-10-2, Suma-ku, Kobe 654-0142, Japan,

*Emergency Medical Care System of Doctor Soetomo Hospital, Japan International Cooperation Agency and

**Tropical Disease Center, Airlangga University, Surabaya 60115, Indonesia

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SUMMARY: Infection with *Toxoplasma gondii* is of medical importance in relation to a recent increase in cases of acquired immunodeficiency syndrome (AIDS). In the present study, we surveyed antibody to *Toxoplasma* among 1,761 people in Surabaya, Indonesia. The overall prevalence was 58% with significant differences between males (63%) and females (52%; *P* < 0.001). Although antibody prevalences at 0-9 years in both genders were less than 10%, those at ages over 10 years were more than 50% in males or more than 40% in females, suggesting an extremely high transmission rate of the parasite to humans in this area. A bimodal pattern in the frequency distribution of *Toxoplasma* antibody levels suggested a persistent feature of *Toxoplasma* infection in humans.

INTRODUCTION

*Toxoplasma gondii* is an intracellular protozoan parasite that can infect a variety of hosts ranging from birds to mammals. Humans, once infected with *Toxoplasma*, persistently harbor this parasite throughout the life, since human defense mechanisms cannot eliminate the cyst form of *Toxoplasma*. Epidemiological studies have indicated that this parasite is ubiquitously distributed throughout the world (1). Although the majority of infections are found to be asymptomatic, this parasite can cause congenital infections in infants and acute infections in immunocompromised individuals. Therefore, the increase in patients with acquired immunodeficiency syndrome (AIDS) has contributed to the increase in *Toxoplasma* encephalitis (2) and pneumonia cases (3).

The region of South and Southeast Asia is reported to have an estimate of 6 million people infected with human immunodeficiency virus (HIV), the second largest such population in the world. Although the largest population with HIV/AIDS is distributed in sub-Saharan Africa, the recent rapid growth of HIV/AIDS populations in Southern and Eastern Asia is a serious concern (4). Therefore, the epidemiology of *Toxoplasma* infection in this region is important in relation to the possible acquisition of opportunistic *Toxoplasma* infection. We have previously surveyed *Toxoplasma* antibody among people in the Philippines and revealed an overall prevalence of 11% in an urban area (Metro Manila) and much higher prevalences in rural areas, such as Mindoro (61%), and Leyte (30%; Kawashima et al., in submission). These prevalences in rural areas were relatively high in comparison with our previous survey data (20%) from a rural area in Japan (5).

In this study, we surveyed the prevalence of *Toxoplasma* antibodies among people in Surabaya, Indonesia. Previous surveys carried out in this country have revealed a wide range from 2% to 63% (6-8) among a variety of human populations. Surabaya is the second largest city in Indonesia with a population of approximately 3 million. Two previous surveys of Surabaya populations revealed much different prevalences of 9% (9) and 63% (10). We revealed in the present survey of over 1,700 people that the overall prevalence was 58% with a significantly higher prevalence in males than females in some age groups.

MATERIALS AND METHODS

Study subjects: A total of 1,761 sera were collected from general patients at the Emergency Unit of Doctor Soetomo Hospital in Surabaya, Indonesia, from November 1999 through March 2000. The ages of the patients ranged from 0 to 100 years, and the patients were grouped at 10-year increments, except for those over 80 years which were grouped in one age group. Serum samples from babies less than 6 months old, which may have contained maternally transferred antibodies, were not used in this survey. Age and sex compositions of

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<td>Total</td>
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*Corresponding author: Tel/Fax: +81-78-796-4594, E-mail: ekon@ams.kobe-u.ac.jp

**Present address: St. Mary’s Hospital, Kurume 830-8543, Japan
the study subjects are shown in Table. Mean ages (± standard deviation (SD)) were 44.9 (±22.1) years in males, 45.2 (±19.5) years in females and 45.0 (±20.3) years in total with no significant differences between males and females (t = 0.303; P > 0.05). The location of Surabaya in Indonesia is shown in Fig. 1.

Enzyme-linked immunosorbent assay (ELISA) system:
Crude soluble Toxoplasma antigen used for ELISA was prepared from tachyzoites of the RH strain of T. gondii as previously described (11). Briefly, tachyzoites harvested in the peritoneal cavity of mice were filtered through a polycarbonate membrane, homogenized and clarified. The supernatant was stored at -30°C until used for ELISA.

The ELISA for measuring antibodies to Toxoplasma was performed as previously described (12) with some modifications (13). In brief, microplate wells sensitized with Toxoplasma antigen were incubated serially with test sera, alkaline phosphatase-conjugated anti-human IgG, and p-nitrophenyl phosphate. Absorbance values obtained in duplicate at 410 nm were averaged and adjusted with the value for the positive control as 1.0 to minimize interplate variations. Sera showing adjusted values (ELISA values) of more than 0.357, which is the cutoff value established previously (12, 13), were determined to be positive for Toxoplasma antibody.

Statistical analysis: Significance of differences in prevalence rates was evaluated by the chi-square test with the Yates’ correction factor. Significance of differences in mean ELISA values was evaluated by the Student’s t test.

RESULTS

Prevalence of Toxoplasma antibody: Figure 2 shows age-dependent prevalence curves obtained with 914 males, 847 females, and a total of 1,761 serum samples. The overall prevalence was 63.2% in males, 52.4% in females and 58.0% in the total population, with a significant difference between males and females (χ² = 20.7; P < 0.001). The prevalence of antibody at 0-9 years was less than 10% in both males and females, which increased significantly to more than 50% in males (χ² = 19.4; P < 0.001) and more than 40% in females (χ² = 9.6; P < 0.01) at 10-19 years. These high antibody prevalences were also observed in age groups over 20 years for both males and females. Males showed significantly higher prevalences than females in age groups of 20-29 (χ² = 4.95; P < 0.05), 30-39 (χ² = 14.5; P < 0.001), and 70-79 (χ² = 8.86; P < 0.01) years.

Distribution of Toxoplasma antibody levels: Figure 3 shows frequency distributions of ELISA values determined for male, female and the total populations. The maximum value was 1.750 in males and 1.636 in females. The distribution pattern was bimodal in both genders with the negative group at a peak of 0.0-0.1 and the positive group at a peak of 0.5-0.8. The mean ELISA values (±SD) of antibody-positive specimens were 0.758 (±0.261) in males and 0.763 (±0.261) in females without significant differences (t = 0.346; P > 0.05). Comparison of age groups in frequency distribution of ELISA
body prevalence in 0-9 years, this transmission mechanism involves (i) congenital transmission, (ii) ingestion of the oocyst contained in infected raw meat, and (iii) accidental ingestion of the oocyst defecated by cats (1). Since the rate of congenital transmission is considered to be less than the antibody prevalence usually increases with age. For instance, our previous survey among a Japanese population revealed 1-4% in 0-39 years, 14% in 40-49 years, 18% in 50-59 years, and 30% in 60-69 years (13). Therefore, the age prevalence curve of the Surabaya population is very different from those of Japanese populations. One particularly distinct point is the difference between prevalences at 0-9 years and 10-19 years. Since the survey was a horizontal but not longitudinal one, this is not direct evidence that transmission is currently active. However, a dramatic increase in prevalence observed in the Surabaya population suggests that this population has a much higher transmission rate than Japanese populations. It is also important to note that the prevalence did not reach near 100% in spite of the possible high transmission rate of Toxoplasma infection.

A bimodal pattern in frequency distribution of Toxoplasma antibody levels was found in the Surabaya population. The bimodal patterns were also observed in our previous (5, 13) and other (33) studies using Japanese populations. Again, based on the persistence of Toxoplasma infection in humans, it is possible that an equilibrium could exist between host immunity and parasite activity (34). Equivalent mean antibody levels in seropositive specimens between males and females suggest that the equilibrium may be maintained in both genders at a similar level. Also, consistent mean antibody levels in seropositive specimens among age groups ranging from 0 to 100 years suggest that the equilibrium may be maintained throughout the life of infected individuals.

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REFERENCES
