

Pattern of toxoplasma antibodies in Malaysian Pregnant Women

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Toxoplasmosis is a disease caused by the coccidian parasite, *Toxoplasma gondii*, whose oocysts are transmitted via the faeces of felines, the common one being the domestic cat. The disease has been known to be contacted by man either:-

- 1) by Acquired infections orally, or
- 2) by Congenital transmission through the placenta.

Work on the life cycle and various aspects of transmission have been stepped up tremendously during the past 10 to 15 years, (Jacob & Lunde 1957, Frenkel 1970, Singh et al. 1972). Likewise, work on life cycle and prevalence of the infection have been in progress in Singapore and Peninsular Malaysia during the past 5-10 years. (Singh et al. 1967, 1968, Zaman et al. 1967, 1969).

One of the first cases of congenital toxoplasmosis was isolated by Pinkerton and Weinman (1940). Recent studies on congenital toxoplasmosis include those reported by Desmots and co-workers (1965), Remington (1969), and Kimball et al. (1971). These projects involved a large population for study and a long period of follow-up studies.

In June 1972, Singh *et al.* using the Indirect Haemagglutination Test reported an incidence of 13.0% Singapore newborns with toxoplasma antibodies.

This infection can cause congenital chorioretinitis, microcephaly, hydrocephaly and fits in infants. Up to date, the problem of congenital toxoplasmosis has not been investigated in Malaysian population.

Prompted by the above reports, we aimed at surveying an antenatal population for the incidence of Toxoplasmosis, with a follow-up of the positive cases to detect any rise in titre and subsequently to follow-up the newborns for their antibody levels and any clinical manifestations of toxoplasmosis. This paper reports the distribution of toxoplasma antibodies in pregnant women.

MATERIALS AND METHODS

Study Population

The study population consisted of 1,459 pa-

tients who booked themselves for antenatal care and delivery at the University Hospital, in Petaling Jaya from July 1972 to March 1974. The patients came from the developing town of Petaling Jaya and its neighbouring suburban villages. Some of them came from the city of Kuala Lumpur, some five to ten miles away and a few foreigners from other countries.

When first seen, the patients were in all stages of pregnancy, with gestation periods ranging from 9 to 38 weeks.

Blood Collections

Blood was routinely taken from first-booking antenatal patients for various haematological and serological investigations. 2 ml. was separated for the indirect fluorescent anti-body test for toxoplasmosis.

The positive titres were tagged on to the front of the patients' case records. Should these patients have booked earlier than 3 months before they were due to deliver, a repeat specimen of blood would be taken during a follow-up clinic. Otherwise, at the time of delivery, two other 2 ml. blood collections were taken from the mother and from the cord or baby.

Follow-up blood collections of babies with positive IgM titres were taken at the ages of 4 months to detect any rise in titre.

The Fluorescent Antibody Test for Toxoplasmosis

A strain of *Toxoplasma gondii* is maintained in infected mice at the IMR. laboratories and antigen slides were freshly prepared and frozen until required for use, usually within one month.

The indirect fluorescent antibody technique (IFAT) for toxoplasmosis was performed using Cooke's microtitre apparatus and Burrough Wellcome's Anti-Human Immunoglobulin, fluorescein labelled. The counter-stain Evans Blue was used. The fluorescein labelled Anti-Human IgM was used in the case of blood from the cord or baby. The IFAT tests were performed according to Palmer (1970) and Remington (1968). A positive reaction was one which showed at least Grade I peripheral fluorescence (Fig.1) whilst a completely negative reaction showed the organism entirely stained red.

A serum was considered positive if a dilution of 1:256 or more showed at least Grade II peripheral fluorescence. However, positive reactions at dilutions of 1:16 and 1:64 were also noted for follow-up studies.

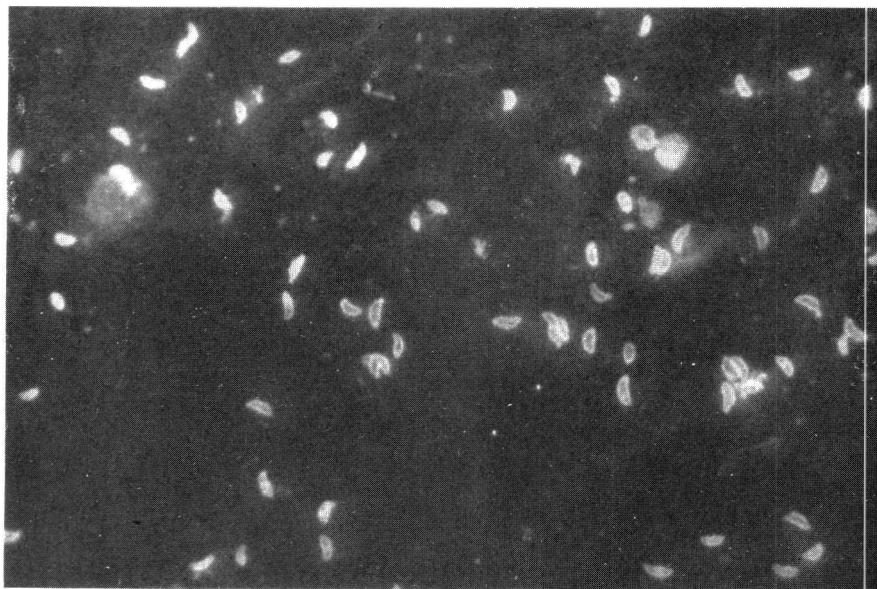


Fig. 1 - Peripheral fluorescence in toxoplasmosis.

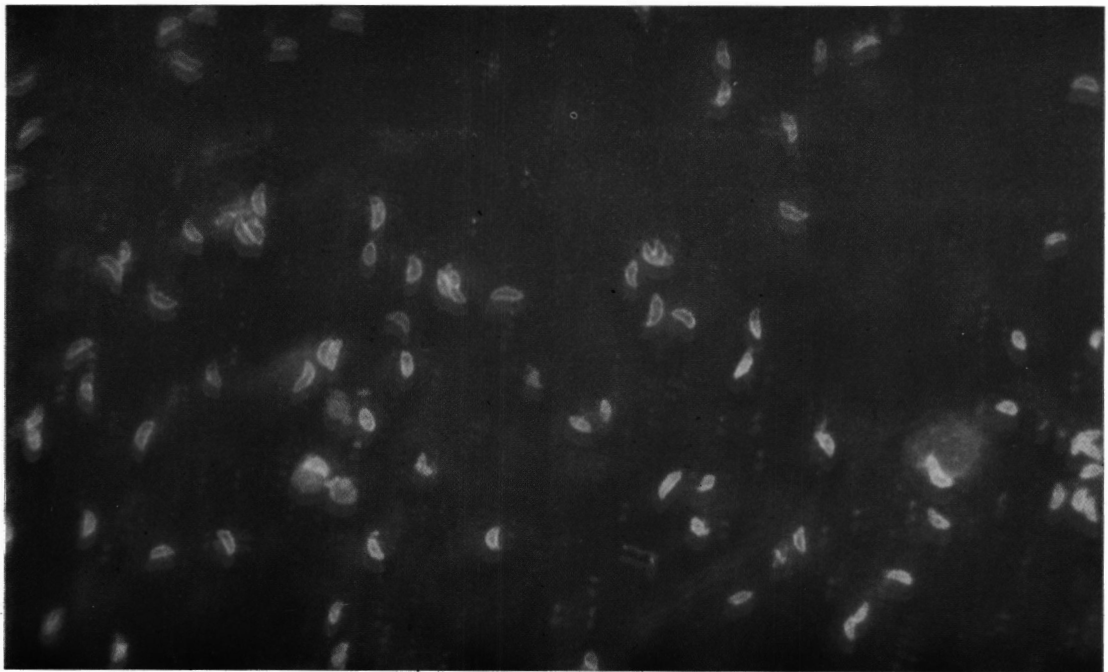


Fig. 1a. Grade IV Fluorescence.

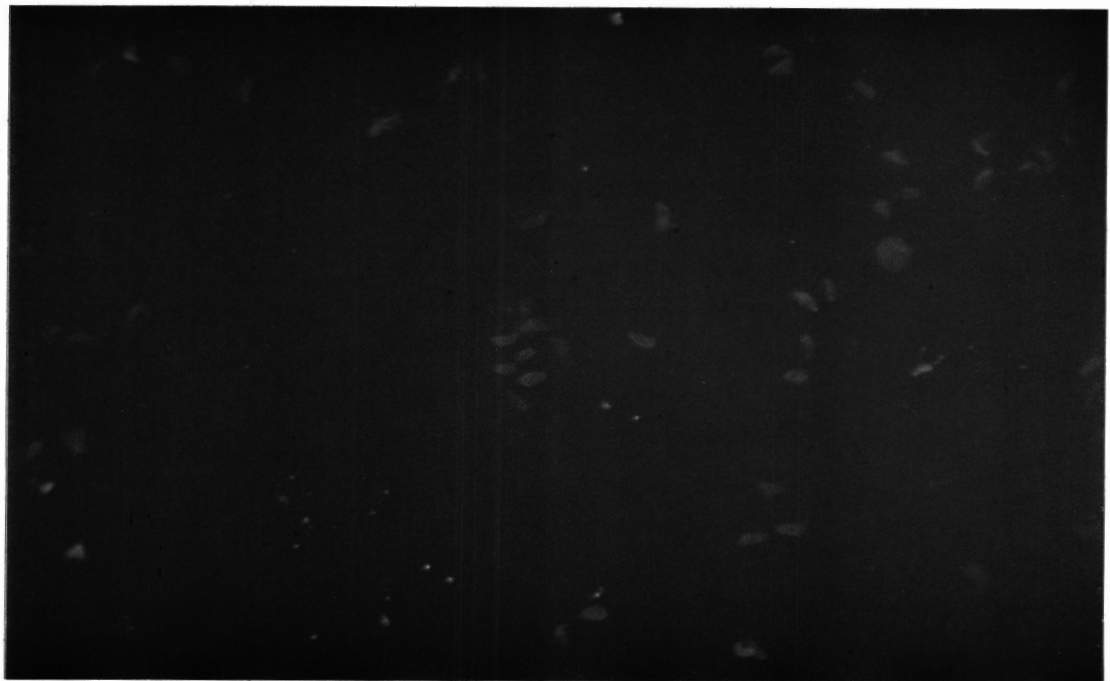


Fig. 1b. Grade I Fluorescence.

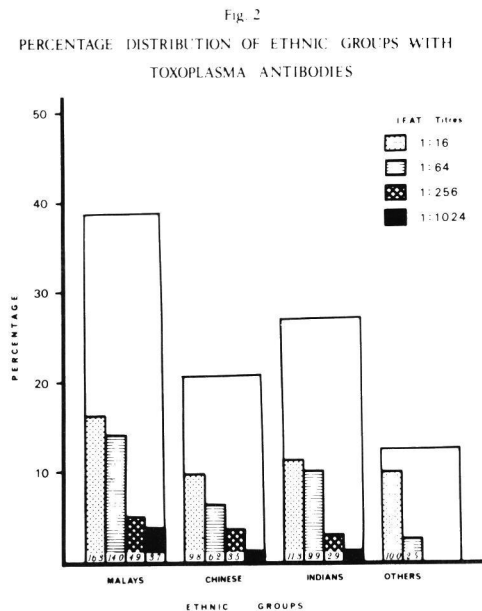
Fig.1. Fluorescence in the Indirect Fluorescent Antibody Test for Toxoplasmosis.

RESULTS

Of the 1,459 women examined, 394 (27.0%) showed positive titres of toxoplasma antibodies; 313 (21.5%) with titres below 1:256, 54 (3.7%) with titre of 1:256 and 27 (1.8%) with titre of 1:1024. (Table I).

Distribution of antibodies in the various race groups

Table I shows the distribution of toxoplasma antibodies in the various ethnic groups of antenatal patients mentioned (Fig.2).



It was interesting to observe that the antibody rate was significantly higher in the Malays than those in the other groups. Though with a smaller population of 431 Malays, which was less than the number of 615 Chinese, the overall incidence in Malays was 38.8% as compared to the 20.65% in Chinese. The 373 Indians showed an in-between incidence of 25.2%.

Table I also shows the distribution of the various titres of toxoplasma antibodies in the 4 ethnic groups. Each of the 4 four-fold antibody titres were found to be significantly the highest in the Malays, being about 30.3% with dilutions

1:16—1:64, 4.9% with the significant dilutions 1:256 and about 3.7% with 1:1024, and lowest in the Chinese, 16.0% with titres of 1:16—1:64, 3.5% with 1:256 and 1.1% with 1:1024.

Distribution in economic groups

Table II shows the distribution of toxoplasma antibodies in the two social-economic groups of income below and above \$200/.

75% of the patients were housewives, Of the remaining 25% who worked, 10.5% come from the higher income group while the other 14.5% were from the lower income group and they worked as in-door or outdoor labourers.

71.8% of the husbands of the patients with positive titres were from the lower-income group of Grades IV and V manual workers who were factory-workers and out-door labourers. The other 28.2% were executive, businessmen, salesmen, teachers and clerks in Grades I, II and III.

Geographical distribution

Table III shows the geographical distribution of the patients with positive titres. It was found that 56.7% of the patients with antibodies came from the rural and suburban areas, namely estates, and villages with or without electricity and water supply. The other 43.3% was from various parts of Petaling Jaya and Kuala Lumpur.

There was no real difference in the distribution of low antibody levels in the village or town folks. However, of those with titres of 1:256, the percentage of positive village folks was three times that of the townsmen, and of those with 1:1024, it was 2½ times more in the villagers than in the townsmen.

DISCUSSION

The toxoplasma antibody incidence of 20.65% in our current group of Chinese agrees with the incidence of 17.2% (29/169), that was found by Singh *et al.* (1968) in their apparently healthy individuals from Singapore where the population was mainly Chinese. Tan and Zaman (1973) found a very low incidence of 5% or 11/220 in their Chinese group of outdoor workers in Peninsular Malaysia.

Singh *et al.* (1967) reported incidences of 27.7% or 56/202 and 12.5% or 6/48 in their survey of pig sera from Singapore and Malaysia respectively. Zaman *et al.* (1972) found a 26% incidence in 131 pigs from Singapore (Zaman *et al.* 1967).

Peterson *et al.* (1972) in assessing the role of cats in the transmission of toxoplasmosis, found that 20.9% of their 235 subjects had possessed a pet cat sometime during their life.

Here in our study a wide significant difference of the incidences of toxoplasma antibodies in the Malays (38.8%) and Chinese (20.6%) was also recognised to be due to two main factors. Firstly, the Chinese have their pork, cut up into small slices or shreds and thoroughly cooked, thus killing off the organism before the meat is consumed. Secondly most of the Malays are very fond of possessing pet cats while the Chinese usually keep dogs as watch-dogs and very few of them are fond of cats. Hence it is the Malays who are likely to have the toxoplasma oocysts transmitted to them from the faeces of their cats.

Hence it is realised that toxoplasmosis is a public health problem of the lower-income group. A careful consideration of their daily life and habits lead us to the many facets in their lives when oocysts transmissions are very likely. Firstly people of lower economic status look for daily past-times and hobbies in and around their homes and the domestic cat is one of the favourite pets especially amongst the Malays. Other favourite past-times of the rural people include gardening and sitting outdoors in the evening.

Cat lovers are often very lenient with their cats that are allowed to roam the entire household. Cats can be seen lurking around their owners;

Following this preliminary paper where the pattern of toxoplasma antibodies has been traced in our Malaysian pregnant women, we hope to report the congenital pattern of toxoplasmosis from our follow-up studies at a later date. A study into the association of toxoplasmosis and cats is now in progress.

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REFERENCES

DESMONTS, G., *et al.*: Etude epidemiologique sur la toxoplasmose: de l'influence de la cuisson

des viandes de boucherie sur la fréquence de l'infection humaine. *Rev. Fr. Etude. Clin. Biol.* 10: 952-958, 1965.

FRENKEL, J.K.: Pursuing Toxoplasmosis. *J. Infect. Disease* 122: 553-559, 1970.

JACOBS, L. and LUNDE, M.N.: Haemagglutination Test for Toxoplasmosis. *Science* 125: 1035, 1957.

JACOBS, L. *et al.* A Haemagglutination test for toxoplasmosis. *J. Parasit.* 43: 308-314, 1957.

KIMBALL, ANNE C. *et al.*: Congenital toxoplasmosis: A prospective study of 4,048 obst. patients. *Am. J. Obstet. Gynaecol.* 111: 211-218, 1971.

PALMER, D.F. and LINCOLN, R.E.: A Procedural Guide to the performance of the Indirect Fluorescent Antibody Test for Toxoplasmosis Immunology Series No.1. U.S. Department of Health, Education and Welfare, Georgia, 1970.

PETERSON, D.R. *et al.*: Human toxoplasmosis prevalence and exposure to cats. *Am. J. Epidemiol.* 96: 215-218, 1972.

REMYINGTON, J.S. *et al.*: IgM antibodies in acute toxoplasmosis I. Diagnostic significance in acquired cases. *J. Lab. Clin. Med.* 71: 855-866, 1968.

REMYINGTON, J.S.: The present status of the IgM fluorescent antibody technique in the diagnosis of congenital toxoplasmosis. *J. Paediatrics.* 75: 1116-1124, 1969.

SINGH, M. *et al.*: A survey on the prevalence of Toxoplasmic antibodies in animal sera. *Med. J. Malaya.* 12: 115-117, 1967.

SINGH, M. *et al.*: A report on the prevalence of toxoplasmic antibodies in Singapore, Singapore *Med. J.* 9: 108-110, 1968.

SINGH, M. *et al.*: Toxoplasmic antibodies in Newborn Infants in Singapore. *Asian Fed. Obst. Gynae.* 3: 58-61, 1972.

TAN, DORA and ZAMAN, V.: Toxoplasma antibody survey in West Malaysia *Med. J. Malaysia.* 27: 188-191, 1973.

ZAMAN, V. *et al.*: Porcine toxoplasmosis in Singapore. *Singapore Med. J.* 8: 246-247, 1967.

ZAMAN, V. *et al.* Toxoplasmic antibodies in various ethnic groups in Singapore (Correspondence). *Trans. Roy. Soc. trop. Med. Hyg.* 65:884, 1969.

	Malays	Chinese	Indians	Others	Total
Total Exam.	431	615	373	40	1459
Positive	168	127	94	5	394
%	38.8	20.65	25.2	12.5	27.0
1:16	70(16.3%)	60(9.8%)	42(11.3%)	4(10.0%)	176(12.1%)
1:64	61(14.0%)	38(6.2%)	37(9.9%)	1(2.5%)	137(9.4%)
1:256	21(4.9%)	22(3.5%)	11(2.9%)	—	54(3.7%)
1:1024	16(3.7%)	7(1.1%)	4(1.1%)	—	27(1.8%)

Table I. Distribution of Toxoplasma antibodies in antenatals by race.

Titres	Percentage of Population with toxoplasma antibodies	
	Higher income (Grades I-III)	Lower income (Grades IV-V)
1:16 & 1:64	21.9	56.3
1:256	3.2	14.5
1:1024	3.1	1.0
	28.2	71.8

Table II. Distribution of toxoplasma antibodies by social economic groups. (Husbands' status).

Titres	Percentage of Population with toxoplasma antibodies	
	Villages	Towns
1:16 & 1:64	40.8	37.3
1:256	13.2	4.9
1:1024	2.7	1.1
	56.7	43.3

Table III. Geographical distribution of toxoplasma antibodies.