# Some studies on toxoplasmosis in Ceylon using the Westphal Reaction (a complement fixation test)

BY

## CHARLES KULASIRI

(Department of Parasitology, Medical Research Institute, Colombo, Ceylon)

Toxoplasma gondii (Nicolle and Manceaux) was first described in Ceylon by Castellani (Castellani, 1949) in 1913, under the name Toxoplasma pyrogenes. The work was not persued subsequently.

Of the present methods available for the diagnosis of toxoplasmosis, the widely employed ones are the Sabin-Feldman dye test (1948) and complement fixation tests. The former requires the live organisms while complement fixation tests do not present this disadvantage. The complement fixation test after Westphal (1951) was selected for this study as the antigen was readily procurable.

## Materials and Methods

A survey of toxoplasmosis in healthy people was carried out first. Samples of blood from persons both males and females, of all age groups were taken from those calling at the Pasteur Institute, Colombo before they commenced their anti-rabies inoculation. The blood was allowed to clot in the refrigerator and centrifuged about four hours later. The serum was separated and inactivated at 56°C. overnight (for 16 hours or more), as required by the test. The serum was then diluted 1:5 with normal saline. The rest of the test was carried out as laid down by Westphal (1951). The sera were taken in batches of twenty-one. Every batch had a positive and a negative control which were subjected to identical conditions as the test sera. Each test serum had a saline control for the purpose of determining the presence of anticomplementary factors.

The second problem undertaken was to study the specificity of the reaction under local conditions. For this purpose, the bloods received at this Institute for the Standard Agglutination Test for the typhoid group of fevers, were used. The sera which has already been inactivated at 56°C. for the Standard Agglutination Test were diluted 1:5 with saline and inactivated once again at 56°C. overnight.

#### Results

These are summarized below:—

Group I. Sera from healthy people seeking treatment at the Pasteur Institute.

Number examined					 109
Number positive for t	oxoplas	smosis			 30
Number showing a doubtful reaction					 10
Number showing anti	factors		 nil		

	Percentage positive for toxoplasmosis				$27 \cdot 5$
	Percentage showing a doubtful reaction				$9 \cdot 1$
Group II	Sera from patients with a clinical history of to be typhoid infection.	fever a	nd susp	pected	
A.	Number examined  Number negative for agglutinins again or para 'A'	st Salm	 nonella	typhi	816 482
В.	Number positive for agglutinins of a titre S. typhi 'H' and/or 'O' or para 'A'	e of 1/50 , ' H '	0 or me	ore to	334
Α.	Sera that were negative for agglutinins $para$ ' $A$ '.	against	S. typ	hi or	
	Number positive for toxoplasmosis				153
	Number negative for toxoplasmosis				294
	Number showing a doubtful reaction				34
	Number showing anti-complementary fac-	tors			1
	Percentage of positives for toxoplasmosis				$27 \cdot 6$
	Percentage showing a doubtful reaction				$7 \cdot 0$
В.	Sera that were positive for agglutinins to more against S. typhi 'H' and/or 'O'				
	Number positive for toxoplasmosis			• •	166
	Number negative for toxoplasmosis				135
	Number showing a doubtful reaction				32
	Number showing anti-complementary fac	tors			1
	Percentage positive for toxoplasmosis				48.8
	Percentage showing a doubtful reaction				9.6
					0

### Discussion

According to the results of the survey in Group I, i.e. in those persons seeking treatment at the Pasteur Institute, endemic toxoplasmosis index for Ceylon is 27.5 per cent. The similar index for Germany is about 20 per cent (Westphal and Bauer 1952). The available data do not specify the method used in obtaining the latter igure and it appears to be likely that one of the other methods may have been used. Hence the two sets of results cannot be strictly compared. The percentage obtained for Ceylon from the sera giving a negative reaction for agglutinins in the S.A.T. (Group IIA) comes close to the above figure.

The perentage (48.8 per cent) of positives for toxoplasmosis in the Group IIB, i.e. sera that are positive for agglutinins in the Standard Agglutination Test to S. typhi and/or para 'A', is very much higher than the percentage in the Group IIA (sera of people without agglutinins to S.A.T.), the increase being roughly 21 per cent. Hein (1952) found that 59 out of 102 tuberculosis patients showed a positive toxoplasmosis reaction both by the Westphal Reaction and the Sabin-Feldman dye test. He interprets the higher percentage as a consequence of the coexistence of toxoplasmosis and tuberculosis as the latter disease paves the way for other maladies. In the case of this study such an interpretation is not justified because, unlike tuberculosis which is generally a chronic disease, typhoid fevers run an acute course.

The results of this study suggests that the Westphal Reaction is not specific.

## Summary

- 1. The endemic toxoplasmosis index, for Ceylon obtained employing the Westphal Reaction is 27.5 per cent. This figure was obtained from a survey carried out among people calling for anti-rabies treatment.
- 2. Evidence is adduced to show that the Westphal Reaction is not specific.

# Acknowledgements

I am grateful to Dr. P. C. C. Garnham of the London School of Hygiene and Tropical Medicine for the supply of a mouse-adapted strain of Toxoplasma gondii; to Dr. A. B. Sabin of the University of Cincinnati for much valuable advice and also for Ioxoplasma-infected mouse brains; to Dr. H. A. Feldman of the State University of New York for assistance received; and to Dr. I. A B. Cathie of the Hospital for Sick Children, Great Ormond Street, London for many sera and anti-sera. Messrs. Farbwerke Hoechst, Germany, supplied free of charge a stock of antigen used in this study.

My thanks are also due to Dr. E. E. Schmid for the bloods used in this experiment, to Drs. C. de S. Mutucumarana and L. B. T. Jayasundera and Mr. T. Velaudapillai for assistance and advice; and to Dr. V. Sivalingam for helpful criticism.

#### References

- Castellani, A., 1949. Little known Tropical diseases. Ann. Inst. Med. trop., Lisboa, 6:369. Hein, W., 1952. Das Verhalten des Sabin-Feldman Testes und der Westphal'schen Komplementbindungs Reaktion bei der Lungentuberkulose. Z. Tropenmed. u. Parasit., 3: (3): 339.
- Westphal, A., 1951. Eine neues Toxoplasmose-Komplementreaktion. Z. Tropen. u. Parasit., 3: (2): 191.
- Westphal, A. and Bauer, F., 1952. Weitere untersuchungen und Betrachtungen zur Toxoplasmose-bindungsreaktion nach Westphal. Z. Tropenmed. u. Parasit., 3: (3): 326.