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British Journal of Obstetrics and Gynaecology
August 1991, Vol. 98, pp. 832–834

ADONIS 030654569100206G

Difficulties in the diagnosis of congenital toxoplasmosis by cordocentesis. Case report

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Case report

A 28-year-old woman who booked at 17 weeks gestation had read about toxoplasmosis in a newspaper and wished to know her status with regard to this infection. After discussion of the limitations of serological testing and consideration of the potential sequelae, a sample of maternal blood was investigated for toxoplasma. Her serology indicated acute toxoplasmosis. Further questioning revealed a history of foreign travel 8 weeks before conception and a mild influenza-like illness at 6 weeks gestation. Ultrasound did not show any fetal abnormality and she chose to continue the pregnancy.

Amniocentesis at 20 weeks for isolation of *Toxoplasma gondii* by animal inoculation did not confirm fetal infection. The woman was given prophylactic spiramycin 3 g daily. At 23 weeks amniocentesis was repeated and cordocentesis was undertaken. The purity of fetal blood was confirmed by a Kleihauer test. In view of the positive fetal serology the woman was given alternating courses of spiramycin 3 g daily for 21 days and a combination of pyrimethamine 25 mg/day, sulphadiazine 2 g/day and folinic acid 5 mg twice weekly for 21 days. This alternating antiparasitic therapy was continued until 38 weeks gestation when spontaneous labour ensued. A female infant was born by ventouse and there were no postnatal maternal complications.

The baby was born in good condition with Apgar scores of 9 at 1 min and 10 at 5 min. Her birthweight was 2800 g (10th centile), length 50 cm (50th centile) and head circumference 33 cm (10th centile). There were no clinical stigmata of congenital infection. A skull X-ray did not show intracranial calcification and liver function tests were normal. When the infant was 3 days old, a loud pan-systolic heart murmur was heard. Investigations revealed a ventricular septal defect of minimum clinical significance.

Because of the serological results the baby was treated for 3 weeks with spiramycin 100 mg/kg/day in two doses, and then for 6 weeks with a combination of pyrimethamine 1 mg/kg/day in two doses, sulphadiazine 200 mg twice daily and folinic acid, 5 mg twice weekly. Alternating courses of these drugs were given until the baby was 7 months old. During treatment with pyrimethamine white blood cell counts were checked weekly to detect bone marrow suppression. Mild leukopenia was noted, with the lowest neutrophil count being 0.88×10^9/l.

Regular clinical and ophthalmological follow-up of the child revealed no abnormality. When last seen, at the age of 7 months, her growth, developmental and ophthalmic assessments were normal.

Laboratory investigations

Toxoplasma-specific IgG was measured using the dye test, direct agglutination test and latex

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Table 1. Toxoplasma specific antibody findings in mother and infant

Serum sample	Dye test (IU)	Latex agglutination test (titre)	Direct agglutination test (units)	IgM ELISA (EIU)	IgM ISAGA (Index)
Maternal					
At 23 weeks	250	256	512	39 (Pos)	12 (Pos)
At delivery	125	256	–	69 (Pos)	12 (Pos)
Fetal					
At 23 weeks	31	16	128	5 (Neg)	11 (Pos)
At delivery	250	–	–	0 (Neg)	0 (Neg)
Infant (weeks)					
3	62	64	–	0 (Neg)	0 (Neg)
28	2	2	–	0 (Neg)	0 (Neg)
30	<2	<2	–	0 (Neg)	0 (Neg)

– = not performed.

agglutination test (Johnson *et al.* 1989). Specific IgM estimation was by a double sandwich enzyme linked immunosorbent assay (DS-ELISA) and immunosorbent agglutination assay (ISAGA) (Duffy *et al.* 1989). Parasite isolation was attempted by intraperitoneal inoculation of laboratory mice. In addition, a recently developed DNA based assay was employed (Savva *et al.* 1990). Serological results are shown in Table 1 and parasite isolation and DNA probe findings in Table 2. Serological studies confirmed acute toxoplasma infection of the mother and specific IgM was detected in fetal blood obtained by cordocentesis. Toxoplasma-specific nucleic acid was detected in cord blood and placental tissue samples taken at delivery. However, DNA probe investigation of neonatal blood samples showed no evidence of infection of the child and serial assessment showed complete loss of passively acquired specific IgG by the age of 7 months.

Table 2. *Toxoplasma gondii* isolation results

Sample	Time of sampling (weeks gestation)	Animal inoculation	DNA probe
Amniotic fluid	20	Neg	Neg
Fetal blood	23	Neg	Not performed
Amniotic fluid	23	Neg	Neg
Placenta	At birth	Neg	Pos
Cord blood	At birth	Neg	Pos
Neonatal blood	At birth	Neg	Neg

Discussion

The management of *T. gondii* infection pregnancy includes maternal and fetal investigation, prophylactic and therapeutic antiparasitic treatment and also consideration of legal termination of pregnancy (Daffos *et al.* 1988). Calls for the introduction of routine antenatal screening for this infection and media interest have recently led to an increased demand for fetal assessment in cases of acute maternal toxoplasmosis (Joynson & Payne 1988).

If antenatal therapy is to be of benefit it is vital that fetal toxoplasmosis is diagnosed specifically. Congenital toxoplasmosis affects not only the newborn but can also result in severe visual impairment presenting late in childhood or during adult life.

Fetal blood sampling by cordocentesis is advocated for fetal assessment (Grose *et al.* 1989). Measurement of specific IgM in of fetal blood requires a highly sensitive and specific assay. Currently, the recommended technique is ISAGA which has shown to be significantly more sensitive than DS-ELISA with no difference in specificity. Using ISAGA, Daffos *et al.* (1988) detected specific IgM in fetal blood samples from 9 of 39 pregnancies where the infant was subsequently found to have congenital toxoplasmosis. The low sensitivity of IgM assessment was thought to be a result of the relative immaturity of the fetal immune system at the time of sampling and inhibition of IgM synthesis due to the suppressive effect of IgG passively transmitted from the mother. However, the specificity of ISAGA was reported to be absolute: no positive reactions in fetal samples

were ever associated with uninfected children (Daffos *et al.* 1988). Our finding of an ISAGA reactive blood sample taken from a uninfected infant may have been a false positive finding. This might have been the result of contamination of the fetal sample with maternal blood but Kleihauer testing did not reveal such contamination. The use of multiple tests to exclude maternal contamination of fetal blood samples has been suggested but this reduces the proportion of the sample available for diagnostic purposes (Forestier *et al.* 1988). A further explanation for our observation could be the transplacental passage of non-viable *T. gondii* leading to stimulation of the fetal immune system without an active disease process. Our experience indicates a need for caution when interpreting toxoplasma-specific IgM assays on fetal blood as ISAGA reactivity cannot be assumed to be an absolute indication of fetal infection. In our patient, the child received potentially toxic treatment and underwent extensive ophthalmic and paediatric follow up. In retrospect this management was unnecessary.

Of the many fetal blood investigations Daffos *et al.* (1988) claimed that parasite isolation had the greatest predictive value; they isolated *T. gondii* from 34 of 42 fetuses (64%) with congenital infection by inoculation of fetal blood and amniotic fluid in laboratory mice. This investigation must be continued for up to 45 days to achieve optimum results. As cordocentesis is performed at 20–24 weeks gestation, isolation of the parasite would involve consideration of a late termination. A reduction in the time required to generate diagnostic results would be advantageous. Tissue culture techniques could speed up diagnosis but they are less sensitive (Hughes *et al.* 1986). Preliminary studies have suggested that detection of toxoplasma-specific DNA in clinical samples may represent a rapid method of investigation with comparable or superior sensitivity to established methods (Savva *et al.* 1990). Cordocentesis yielded insufficient fetal blood to permit DNA investigation in our patient. However, neonatal blood contained no detectable toxoplasma DNA. The detection of toxoplasma DNA in concurrent samples of placenta and cord blood may represent maternal parasitaemia and contamination of these samples with maternal blood. Failure to isolate the parasite following animal inoculation of cord blood and placental tissue may represent lower sensitivity of this technique compared with DNA detection. False positive reactions using the DNA probe method have not been reported (Savva *et al.* 1990). Isolated infection of the placenta without passage of the parasite to the infant has been noted, particularly after spiramycin treatment of the mother (Couvreur *et al.* 1988) and could account for our observations.

The case described illustrates the complexity of the diagnosis and management of toxoplasmosis in pregnancy and the need for an integrated multi-disciplinary approach to achieve optimum care of the patient.

Acknowledgments

Part of this work was funded by the Medical Research Council (R.E.H., D.S.). We are most grateful to Mrs I. Crossman for the preparation of this manuscript.

References

Couvreur J., Desmonts G. & Thulliez Ph. (1988) Prophylaxis of congenital toxoplasmosis. Effects of spiramycin on placental infection. *J Antimicrob Chemother* **22** Supp. B, 193-200.

Daffos F., Forestier F., Capella-Paulovsky M. *et al.* (1988) Prenatal management of 746 pregnancies at risk for congenital toxoplasmosis. *N Engl J Med* **318,** 271-275.

Duffy K. T., Wharton P. J., Johnson J. D., New L. & Holliman R. E. (1989) Assessment of an immunoglobulin-M immunosorbent agglutination assay (ISAGA) for the detection of toxoplasma specific IgM. *J Clin Pathol* **42,** 1291-1295.

Forestier F., Cox W. L., Daffos F., Rainaut M. (1988) The assessment of fetal blood samples. *Am J Obstet Gynecol* **158,** 1184-1188.

Grose C., Itani O. & Weiner C. P. (1989) Prenatal diagnosis of fetal infection: advances from amniocentesis to cordocentesis—congenital toxoplasmosis, rubella, cytomegalovirus, varicella virus, parvovirus and human immunodeficiency virus. *Paediatr Infect Dis* **8,** 459-468.

Hughes H. P., Hudson L. & Fleck D. G. (1986) In vitro culture of *Toxoplasma gondii* in primary and established cell lines. *Int J Parasitol* **16,** 317-322.

Johnson J., Duffy K., New L., Holliman R. E., Chessum B. S. & Fleck D. G. (1989) Direct agglutination test and other assays for measuring antibodies to *Toxoplasma gondii. J Clin Pathol* **42,** 536-541.

Joynson D. H. & Payne R. (1988) Screening for toxoplasma in pregnancy. *Lancet* **ii,** 795-796.

Savva D., Morris J. C., Johnson J. D. & Holliman R. E. (1990) Polymerase chain reaction for detection of *Toxoplasma gondii. J Med Microbiol* **32,** 25-31.

Received 3 December 1990
Accepted 25 March 1991

14710528, 1991, 8, Downloaded from https://obgyn.onlinelibrary.wiley.com/doi/10.1111/j.1471-0528.1991.tb13492.x by <Shibboleth>-member@kcl.ac.uk, Wiley Online Library on [01/11/2023]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License