ANTIBODIES TO *CHLAMYDIA TRACHOMATIS* IN PATIENTS PRESENTING WITH ECTOPIC PREGNANCY AT GROOTE SCHUUR HOSPITAL

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Objectives. To determine the prevalence of antibodies to *Chlamydia trachomatis* in women presenting with ectopic pregnancies to Groote Schuur Hospital.

Methods. *C. trachomatis* antibody titres were measured using a modified micro-immunofluorescence test in women presenting with ectopic pregnancy. Control subjects were drawn from women with term pregnancies and an uneventful reproductive history.

Results. Seventy-four patients and controls were studied. Demographic variables were controlled for at time of entry into the study. A significant association between the number of lifetime sexual partners and exposure to *C. trachomatis* was noted ($P = 0.001$). Patients with ectopic pregnancies had significantly higher antibody titres than control subjects ($P = 0.001$), and in both groups the prevalence of background antichlamydial antibody was high (ectopic pregnancies 59%, pregnant controls 32%).

Conclusions. While the role of *C. trachomatis* infection in women who develop ectopic pregnancies needs to be explored further, it seems wise to treat them all with empirical antibiotics at the time of presentation.

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Chlamydia trachomatis and *Neisseria gonorrhoeae* are major causes of pelvic inflammatory disease (PID) and subsequent tubal damage in women. Patients with a history of documented PID have a sevenfold increased prevalence of tubal pregnancies when compared with women who have never been infected with these organisms.[1] Westrom *et al.*[1] demonstrated that salpingitis is the most important predisposing factor for subsequent ectopic pregnancy.

In recent years, several industrialised countries have reported an increasing incidence of ectopic pregnancy, presumably the legacy of the sexually transmitted disease epidemic of the 1960s and 1970s.[1-4]

Maternal mortality as a result of ectopic pregnancy has dropped dramatically since the early 1970s, but the occurrence of an ectopic pregnancy has major long-term effects on a woman's reproductive capacity. It has been estimated that there is a 50% infertility rate after an ectopic pregnancy and that in 10 - 20% of patients subsequent pregnancies will again be extra-uterine.[5]

C. trachomatis appears to be the most frequent microbial agent involved in the pathogenesis of pelvic inflammatory disease in industrialised societies,[6-9] while *N. gonorrhoeae* remains an important cause in developing countries.[10]

The mechanism by which *C. trachomatis* induces tubal damage is unclear. It may be that persistent *C. trachomatis* infection causes tubal damage directly, or that tubal occlusion results indirectly from post-inflammatory changes. Attempts to isolate *C. trachomatis* in cell culture from tubes resected during surgery for ectopic pregnancy have usually not been successful.[11-13] It is postulated that many women with *C. trachomatis*-associated salpingitis remain undiagnosed as the infection does not present with overt clinical symptoms. Because medical treatment is not sought, the tubal damage is often not recognised and these women may therefore present years later with an ectopic pregnancy as a consequence.[14] This hypothesis is further supported by the fact that an association between high titres of antichlamydial antibodies and tubal pathology in apparently previously asymptomatic individuals has frequently been documented.[15-17] However, it should be noted that uncomplicated asymptomatic chlamydial infection of the endocervix may also elicit a significant antibody response.[10]

This study was undertaken to determine the prevalence of antibodies to *C. trachomatis* in women presenting with ectopic pregnancy at Groote Schuur Hospital. These antibodies could serve as an indicator of the possible role of *C. trachomatis* in the pathogenesis of ectopic pregnancy in Cape Town and the need for possible review of antibiotic policy for treatment of PID.

SUBJECTS AND METHODS

Subjects

The study population comprised women presenting with an ectopic pregnancy confirmed at the time of laparotomy at Groote Schuur Hospital, Cape Town. A group of control subjects was drawn from women with healthy term

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pregnancies and uneventful reproductive histories (i.e. no history of miscarriages, infertility, tubal surgery or PID) seen at the Maternity Centre of the same hospital. Controls were matched with the subjects for age (± 2 years), race, parity (1 - 5 or 6+) and number of lifetime sexual partners (1, 2 - 5, > 5). Both cases and controls were drawn from a relatively homogeneous population, with a similar socio-economic background. Informed consent was obtained from all women participating in the study. Consent to perform this study was obtained from the Ethics Committee of the University of Cape Town Medical School.

Laboratory methods

Blood taken from both subjects and controls was allowed to clot and the serum was separated, frozen and stored at –70°C. Analysis for chlamydial antibodies was conducted at the South African Institute for Medical Research in Johannesburg using a modified micro-immunofluorescence (micro-IF) test according to the method described previously.[18]

Statistical methods

All comparisons employed the chi-square test with a Yates correction or the one-tailed Fisher's exact test. A paired *t*-test was used to assess the difference between means.

Results

Seventy-four matched patients and controls were enrolled into the study. There was no difference in the demographic data between the study population and the controls, since we controlled for these variables at the time of entry into the study. Of the 74 patients presenting with ectopic pregnancy only 18 (24%) gave a history of a single lifetime sexual partner.

Taking both groups (ectopic pregnancies and controls) together, a significant association was noted between the number of lifetime sexual partners and lifetime exposure to *C. trachomatis* infection reflected by antichlamydial immunoglobulin G (IgG) antibody titres ≥ 1:16 ($\chi_2^2 = 6.76$, $P = 0.034$) (Table I). Of the 5 patients in the study group who gave a history of previous symptomatic PID, all had elevated IgG antibody titres of ≥ 1:64, which is consistent with more invasive disease.

The distribution of antichlamydial antibody titres in both the study and control groups is shown in Table II. Patients with ectopic pregnancy were found to have significantly higher IgG antibody titres than the pregnant controls ($\chi_4^2 = 18.30$, $P = 0.001$).

Table I. Association between number of lifetime sexual partners and antichlamydial antibody titre ($N = 146$)

Micro-IF titres	Lifetime sexual partners (*N*)		
	1 partner	2 - 5 partners	> 5 partners
Positive (≥ 1:16)	15	50	3
Negative (< 1:16)	32	45	1

$\chi_2^2 = 6.76$; $P = 0.034$.

Table II. Distribution of antichlamydial antibody titres in patients with ectopic pregnancy and pregnant controls

Patient group	Antichlamydial antibody titre				
	≤ 1:8	1:16	1:32	1:64	≥ 1:128
Ectopic pregnancy	30	6	13	13	12
Pregnant controls	50	8	4	2	10

$\chi_4^2 = 18.30$, $P = 0.001$.

Discussion

In this study the group of 74 women presenting with ectopic pregnancy were found to have significantly higher IgG antibody titres to *C. trachomatis* than the 74 women with normal pregnancies. Although we did not test for the prevalence of other pathogens that could cause tubal damage, such as *N. gonorrhoeae* and *Mycoplasma hominis*, it would appear that serological evidence consistent with previous exposure to *C. trachomatis* could be associated with ectopic pregnancy in many cases. The 32% prevalence of antichlamydial IgG antibodies at titres ≥ 1:16 detected in the control group is higher than the usual background figure of 10% quoted for random Western populations.[19-22] These higher rates are, however, in keeping with other data presented in local studies. We have previously detected antichlamydial antibodies in 35% of a random black population in Johannesburg, using micro-immunofluorescence serology,[23] while Jennings *et al.*[24] found a rate of 39% in their control group of pregnant patients admitted to Groote Schuur Hospital. The seroprevalence rates support the notion that chlamydial infection is common and usually remains undetected — given that none of the control subjects, despite careful questioning, had any past history of genital tract disease.

What is not known from our study is the prevalence of active tubal chlamydial infection at the time of ectopic pregnancy, since culture for *C. trachomatis* from the tubes was not performed. Studies conducted in industrialised settings appear to indicate that the tubal damage is the result of previous active disease, since most authors have failed to culture *C. trachomatis* from tubal specimens.[11,15,24,25]

However, Ville *et al.*[26] in Gabon cultured the organism from 71% of tubal specimens obtained from women with ectopic pregnancies at the time of laparotomy. Bentsi *et al.*[27] reported a prevalence of 31% active infection in a Ghanaian population. It therefore appears that the African scenario may be different

from industrialised countries; these studies suggest a higher prevalence of active chlamydial infection in Africa than elsewhere.

The association of antichlamydial antibodies with active PID is well documented, with seropositivity rates of 39.5% detected in the study by Kinghorn *et al.*[28] in Britain, 73% detected in a study conducted by Burchell and Welgemoed in Bloemfontein,[29] and 87.5% in a study conducted in Cape Town.[24]

A previous history of PID was noted in only 5 patients in our study, all of whom had elevated antichlamydial IgG titres, but this is not an unusual finding and has been noted by other authors,[11,15,30,31] demonstrating how frequently chlamydial PID may be asymptomatic and therefore untreated. Mehanna *et al.*[30] reported that only 25.5% of all women with laparoscopically diagnosed tubal damage resulting from prior PID reported a clinical history of PID. A study by Cumining *et al.*[32] showed that among 27 women attending an infertility clinic who subsequently developed an ectopic pregnancy, previous laparoscopy had indicated 'normal' tubal morphology in 12 instances (44%).

Our study supports the notion that chlamydial infections are common and remain undetected. Ectopic pregnancy is a major public health problem, and the rising incidence of this complication sustains this view. There is a strong need to identify the most effective means of reducing the ectopic pregnancy rate. Since there is concurrence among most studies that prior chlamydial infection is strongly associated with ectopic pregnancy, reduction of the chlamydial reservoir could result in a reduction in both the incidence of PID and its subsequent sequelae.

The high prevalence of antichlamydial IgG antibodies noted in pregnant women is of concern, as active infection could have occurred during the course of the index pregnancy or it could be current. Several studies have demonstrated pregnancy-related complications attributable to *C. trachomatis*, including premature rupture of membranes, preterm labour, low birth weight (< 2 500 g), impaired fetal growth, stillbirth and intrapartum and postpartum maternal and neonatal infectious morbidity.[33,34] To prevent neonatal morbidity, identification and treatment of maternal chlamydial infection during pregnancy is required. Clearly, if the background prevalence is as high, as we (and others) have demonstrated it to be, then in the appropriate clinical setting empirical treatment may be indicated.

Ideally, control of chlamydial infection should take place before development of complications and their sequelae, which would entail screening for uncomplicated cervical infection and provision of appropriate therapy. Unfortunately this approach requires the application of expensive laboratory testing that is not affordable in a developing country setting. An alternative is to provide adequate therapeutic cover for both gonococcal and chlamydial infection in women presenting with signs and/or symptoms consistent with sexually transmitted infections (syndromic management), as well as for their sexual partners with urethritis.

The potential benefits of antimicrobial therapy at the time of ectopic pregnancy remain controversial and further studies to determine whether such treatment is beneficial need to be conducted. Women with ectopic pregnancies have continuing high rates of infertility and recurrent ectopic pregnancy,[5] so if active infection underlies their disease, antibiotic therapy may be beneficial. Until this controversy is resolved, it would appear prudent in the African setting to treat empirically all patients presenting with either PID or ectopic pregnancy using appropriate antimicrobial therapy that includes antibiotics active against *C. trachomatis*.

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