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Antichlamydial antibodies in pelvic inflammatory disease

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SUMMARY The role of *Chlamydia trachomatis* in pelvic inflammatory disease (PID) diagnosed without laparoscopy was assessed by measuring antichlamydial antibodies in the patient's serum and by comparing the results with those in patients with uncomplicated non-specific genital infection (NSGI) and gonorrhoea and in non-infected controls. A modified micro-immunofluorescence test was used.

Patients with severe PID had significantly more positive antichlamydial IgG and IgM results than did control subjects, patients with gonorrhoea, and patients with NSGI. Less severe PID was associated with significantly raised levels of antichlamydial IgG antibodies compared with NSGI and controls and with raised levels of IgM antibodies compared with controls. Two patients with PID had lower genital tract gonorrhoea, one of whom had raised antichlamydial antibody levels. These findings may indicate a mixed infection and therapy should be reviewed in such patients. A serological diagnosis of chlamydial infection is relatively easy and cheap and enables a rapid diagnosis of chlamydial infection to be made.

Introduction

Chlamydia trachomatis is associated with non-specific genital infection (NSGI) in women (Alani *et al.*, 1977), and causes acute salpingitis in some patients (Mårdh *et al.*, 1977). Chlamydial serum antibody levels correlate with *C. trachomatis* isolation from the cervix (Treharne *et al.*, 1978) and from the Fallopian tubes in patients with salpingitis diagnosed by laparoscopy (Treharne *et al.*, 1979). Unfortunately we, as well as others, do not have access to the type of laparoscopy service described by Treharne *et al.* (1979). However, when investigating causes of pelvic inflammatory disease (PID) we considered it useful to assess the serum chlamydial antibody levels in patients with PID diagnosed on clinical grounds. These were compared with the levels of antichlamydial antibodies in the serum of patients with uncomplicated genital infections and of controls.

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Patients and method

Patients attending the Departments of Genital Medicine at St Bartholomew's Hospital and the Eastern Hospital, London, who were thought to have PID were included in this study. Criteria for the diagnosis of PID were based on those of Forslin *et al.* (1978) as follows:

(A) Lower abdominal pain,
(B) Pelvic tenderness with or without pelvic mass,
(1) Vaginal discharge,
(2) Menstrual irregularities,
(3) Dysuria,
(4) Dyspareunia,
(5) Temperature $\geqslant$38°C,
(6) ESR $\geqslant$15/hour (Westergren),
(7) White cell count 10 000 cu/mm (10×10^9/l).

Patients were divided into two main groups: group 1 had strict admission criteria—that is, (A) + (B) + 2 of (1) − (7)—whereas group 2 had less strict criteria—that is, (A) + (B) + 1 of (1) − (7). Patients in group 1 were automatically included in group 2.

All patients were investigated for sexually transmitted diseases. Material from the lateral vaginal fornices was examined microscopically for

yeast cells and hyphae of *Candida* species after staining by Gram's method; secretion from the posterior fornix, mixed with saline, was examined microscopically for *Trichomonas vaginalis*. Vaginal material was inoculated on to Sabouraud's medium for culture for *Candida* species, and into Oxoid No. 2 trichomonas medium and examined after 48 hours' incubation. Specimens from the urethral meatus and cervical canal, stained by Gram's method, were examined for leucocytes and Gram-negative diplococci, and plain swabs from these sites were placed in Amies's modification of Stuart's transport medium. Within 24 hours they were plated on to lysed blood agar containing vancomycin, colistin, trimethoprim, and natamycin (Riddell and Buck, 1970). *Neisseria gonorrhoeae* was identified by sugar fermentation reactions using serum-free medium (Flynn and Waitkins, 1972). Specimens from the rectum, taken through a proctoscope, were also examined when gonorrhoea was particularly suspected. Cervical material was collected with an Ayre's spatula, wet-fixed in alcohol and ether, stained by Papanicolaou's method, and examined microscopically for malignant cells and inflammatory changes (Thin *et al.*, 1975). Blood was taken for syphilis antibody tests using the Venereal Disease Research Laboratory (VDRL) and *Treponema pallidum* haemagglutination (TPHA) tests. Cultures for *Chlamydia trachomatis* were not taken routinely.

Blood was taken for a modified micro-immunofluorescence test for chlamydial antibodies (Treharne *et al.*, 1977) on the day of diagnosis; the serum was separated and stored at −20°C until tested. Patients thought to have PID were matched for age, marital state, and country of origin with patients who had NSGI and with control women who had no evidence of genital infection or known contact with any sexually transmitted disease (STD). The diagnosis in patients with NSGI had been made on the basis of their contact with a man with non-specific urethritis or of the presence of excess urethral and cervical leucocytes in the absence of any pathogen (Treharne *et al.*, 1978). A group of patients with uncomplicated genitorectal gonorrhoea was also included, but unfortunately these could not be matched with the study patients.

Statistical comparisons were made using the χ^2 test with Yates's corrections.

Results

A total of 109 patients were included: 30 with PID (17 in group 1 and 30 in group 2), 30 with NSGI, 19 with gonorrhoea, and 30 as control subjects. The presence and levels of antichlamydial IgG and IgM antibodies are shown in Tables 1 and 2. If antichlamydial IgG antibody at a level of 1/64 or higher and antichlamydial IgM at a level of 1/8 or higher in the blood is considered to suggest active chlamydial infection (Treharne *et al.*, 1979), patients with PID in group 1 had significantly more positive results than control subjects ($\chi^2_1 = 4{\cdot}48$, $P<0{\cdot}05$) and patients with NSGI ($\chi^2_1 = 8{\cdot}60$, $P<0{\cdot}01$) and gonorrhoea ($\chi^2_1 = 3{\cdot}91$, $P<0{\cdot}05$) (Table 2). Patients in group 2 (including those with less strict criteria for PID) had significantly raised levels of antichlamydial IgG compared with control subjects ($\chi^2_1 = 5{\cdot}69$, $P<0{\cdot}02$) and those with NSGI ($\chi^2_1 = 4{\cdot}18$, $P<0{\cdot}05$), and significantly raised levels of antichlamydial IgM compared with control subjects ($\chi^2_1 = 4{\cdot}01$, $P<0{\cdot}05$). Antichlamydial IgM antibody at a level of 1/8 or higher was present in 29-30% of patients with PID in groups 1 and 2. One patient in group 1 had genital gonorrhoea with raised levels of antichlamydial IgG and IgM, and two patients in group 2 had genital gonorrhoea, one with raised antichlamydial IgG and IgM levels and the other with no detectable antichlamydial antibodies. All the other patients with PID had negative results for gonococcal investigations and so were considered to have NSGI. In group 1 three patients had *T. vaginalis* and three yeasts; in group 2 five patients had yeasts, three *T. vaginalis*, and one cervical herpes virus type 2.

Table 1 *Levels of antichlamydial IgG and IgM antibodies*

	Antichlamydial antibodies (titre)					
	IgG (≥1/64)		*IgM (≥1/8)*		*Total**	
Patient group	*No.*	*%*	*No.*	*%*	*No.*	*%*
With PID						
Group 1	10/17	59	5/17	29	10/17	59
Group 2	12/30	40	9/30	30	14/30	47
With NSGI	4/30	13	14/30	47	15/30	50
With gonorrhoea	4/19	21	5/19	26	7/19	38
Control	3/30	10	2/30	7	4/30	13

*With positive antichlamydial antibodies (IgG ≥1/64 or IgM ≥1/8)

Table 2 *Levels of antichlamydial IgG antibody*

		Levels of antichlamydial IgG antibody					
Patient group	*No. of patients*	*1/16*	*1/32*	*1/64*	*1/128*	*1/256*	*1/512*
With PID							
Group 1	17	2	2	3	2	3	2
Group 2	30	7	3	3	2	4	3
With NSGI	30	12	1	3	0	1	0
With gonorrhoea	19	8	0	3	0	1	0
Control	30	5	0	3	0	0	0

Discussion

Patients who attend departments of genitourinary medicine with evidence of PID, and in whom no evidence of gonorrhoea is found, are usually treated with tetracyclines for non-specific salpingitis. Our results suggest that this form of antibacterial treatment is reasonable as 59% of patients with PID in group 1 had raised serum antichlamydial IgG levels and 30% raised IgM antibody levels. Only one patient had gonorrhoea, and she also had raised antichlamydial IgG and IgM levels. When PID was, however, more loosely defined, only 40% of patients had raised antichlamydial IgG levels but still 30% had raised IgM levels. In this group, two patients had gonorrhoea, but only one had raised antichlamydial IgG and IgM antibody levels. Although some patients had *T. vaginalis* infection in the lower genital tract, it rarely appears to cause PID (Mårdh and Westrom, 1970). We found higher levels of antichlamydial IgM in patients diagnosed as having uncomplicated NSGI than in other categories, supporting our previous findings (Treharne *et al.*, 1978).

Since in our practice the presentation of PID is usually acute and the patients need antibacterial treatment on the day of consultation, rapid diagnosis is essential. Gonorrhoea can be quickly excluded by Gram-staining and culture, but at present *C. trachomatis* cultures may take time, are not widely available, and are expensive. Serodiagnosis of chlamydial infection, which can be performed readily, rapidly, and relatively cheaply (Treharne *et al.*, 1978) therefore has great advantages. Furthermore, it is useful in clinics which have no nearby facilities for *C. trachomatis* culture as serum is easily transported; pilot studies have shown that even small amounts of blood (0·1 ml) soaked on to cellulose sponges give reliable antibody results (Darougar *et al.*, 1978).

If antichlamydial antibody levels are raised in patients with PID and lower genital tract gonorrhoea, the therapeutic regimen should be reviewed, as these results may indicate a mixed infection.

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References

Alani, M. D., Darougar, S., Burns, D. C. MacD., Thin, R. N., and Dunn, H. (1977). Isolation of *Chlamydia trachomatis* from the male urethra. *British Journal of Venereal Diseases*, **53**, 88-92.

Darougar, S., Treharne, J. D., Minassian, D., El-Sheikh, H., Dines, R. J., and Jones, B. R. (1978). Rapid serological test for diagnosis of chlamydial ocular infections. *British Journal of Ophthalmology*, **62**, 503-508.

Flynn, J. and Waitkins, S. A. (1972). Serum-free medium for testing fermentation reactions in *Neisseria gonorrhoeae*. *Journal of Clinical Pathology*, **25**, 525-527.

Forslin, L., Falk, V., and Danielsson, D. (1978). Changes in the incidence of acute gonococcal and non-gonococcal salpingitis. *British Journal of Venereal Diseases*, **54**, 247-250.

Mårdh, P-A., Ripa, T., Svensson, L., and Westrom, L. (1977). *Chlamydia trachomatis* infection in patients with acute salpingitis. *New England Journal of Medicine*, **296**, 1377-1379.

Mårdh, P-A. and Westrom, L. (1970). Tubal and cervical culture in acute salpingitis with special reference to *Mycoplasma hominiss* and T-strain mycoplasmas. *British Journal of Venereal Diseases*, **46**, 179-186.

Riddell, R. H. and Buck, A. C. (1970). Trimethoprim as an additional selective agent in media for the isolation of *N. gonorrhoeae*. *Journal of Clinical Pathology*, **23**, 481-488.

Thin, R. N., Atia, W., Parker, J. D. J., Nicol, C. S., and Canti, G. (1975). Value of Papanicolaou-stained smears in diagnosis of trichomoniasis, candidiasis, and cervical herpes simplex virus infection of women. *British Journal of Venereal Diseases*, **51**, 116-118.

Treharne, J. D., Darougar, S., and Jones, B. R. (1977). Modification of the micro-immunofluorescence test to provide a routine serodiagnostic test for chlamydial infection. *Journal of Clinical Pathology*, **30**, 510-517.

Treharne, J. D., Darougar, S., Simmons, P. D., and Thin, R. N. (1978). Rapid diagnosis of chlamydial infection of the cervix. *British Journal of Venereal Diseases*, **54**, 403-408.

Treharne, J. D., Ripa, K. T., Mårdh, P-A., Svensson, L., Westrom, L., and Darougar, S. (1979). Antibodies to *Chlamydia trachomatis* in acute salpingitis. *British Journal of Venereal Diseases*, **55**, 26-29.