Epidemiological and clinical correlates of chlamydial infection of the cervix

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SUMMARY Of 474 women studied to identify epidemiological and clinical correlates of chlamydial infection of the cervix, Chlamydia trachomatis was isolated from 158 (33·3%) of all women, from 48·3% of those infected with Neisseria gonorrhoeae, from 43% of the sexual consorts of men with nongonococcal urethritis, and from 74% of those whose consorts were also infected with C trachomatis. C trachomatis was the sole pathogen found in 58 women. Age, marital state, occupation, past history of gonorrhoea, menstrual state, and symptoms had no predictive value. The isolation of C trachomatis was significantly associated with N gonorrhoeae, the use of oral contraceptives, cervical ectopy, cervicitis, and last sexual exposure more than one week previously. Except for three patients, none of the criteria alone or in combination was reliable enough to predict with acceptable accuracy that the 30 chlamydia-positive women among the 191 who were not infected with N gonorrhoeae and whose consorts were not known to have urethritis harboured chlamydia.

Introduction

Gonorrhoea and non-specific genital infection are now the commonest sexually transmitted diseases. Both are easier to detect in men than in women; hence their incidence in men would probably be a fair reflection of the true situation. In the year ending 31 December 1978, 35 366 male cases of gonorrhoea and 78 298 male cases of non-specific genital infection were diagnosed at the hospital clinics in England.[1] If, as is now generally accepted, half of these non-specific genital infections were due to Chlamydia trachomatis then the latter organism assumes an importance at least equal to that of Neisseria gonorrhoeae. A similar situation probably exists in many countries.

There are several broad similarities between N gonorrhoeae and C trachomatis, such as their intracellular habitat and their preference for columnar epithelium. Some clinical aspects are similar also; C trachomatis may cause urethritis, epididymitis, cervicitis, salpingitis, conjunctivitis, and possibly prostatitis, endocarditis, peritonitis, and perihepatitis.[2] Despite a wide spectrum of clinicopathological effects a routine laboratory service for culturing C trachomatis is not yet generally available, which probably encourages some workers to continue to regard the "non-specific" genital infection as trivial and which consequently results in a lack of enthusiasm for contact tracing. This attitude is further strengthened by the convenience it affords in assuring and protecting patients from the assumption that their condition was related to sexual indiscretion.

In view of the unlikelihood of a routine laboratory service for C trachomatis being provided in many areas in the immediate future, we have made a search for clinical and epidemiological correlates which might help in the management of the condition in the absence of laboratory diagnosis. A further aim was to establish a baseline of the incidence of chlamydial infections, in a community where contact tracing for non-specific urethritis had not been practised, for future trends and comparisons.

As patients with chlamydia in the genital tract may harbour other sexually transmitted organisms, we considered carefully the influence of intercurrent infections when analysing the data. The clinical and epidemiological findings in women associated with C

trachomatis compared with *N gonorrhoeae* and "combined infection", and where relevant *Candida albicans* and *Trichomonas vaginalis,* are reported in this paper.

Patients and methods

The study population comprised 474 women, who had not received an antibiotic in the preceding four weeks, seen by one observer (OPA) at the Special Clinic, St James's Hospital, Birkenhead, between January 1976 and September 1978.

LABORATORY METHODS
N gonorrhoeae
Smears and cultures were made from samples from the urethra and endocervix in all cases and from the rectum in some. Gram-stained smears were examined in the clinic. Swabs sent to the Regional Public Health Laboratory in Stuart's transport medium were cultured on selective media and processed in the usual way.

C trachomatis
Swabs were taken from the endocervix in all cases and from the urethra in 104 cases. The swabs were transported to the laboratory in 2·5 ml of a transport medium consisting of Wellcome Medium 199 with 0·11% sodium bicarbonate supplemented with 10% fetal bovine serum, 10% sorbitol, 0·5% glucose, and 100 units/ml each of vancomycin, streptomycin, and mycostatin. The isolation technique was based on that described by Reeve *et al*[3] using idoxuridine-treated McCoy cells. Modifications to the method were the seeding of coverslips with only 6×10^4 McCoy cells: inoculation of only 0·5 ml of specimen together with 1·5 ml of fresh growth medium containing 0·5% glucose; and staining of infected coverslips after 72 hours' incubation by the Giemsa method of Johnson.[4]

Trichomonas vaginalis
Wet preparations of material from the posterior vaginal fornix were examined in the clinic by phase-contrast microscopy, and swabs were sent to the laboratory in Feinberg-Whittington medium.

Candida albicans
Gram-stained vaginal smears were examined for yeast cells and hyphae, and cultures were sent to the laboratory on blood agar.

Herpes simplex virus
Swabs were taken from the endocervix and sent to the laboratory in virus transport medium to be cultured on human embryonic lung (MRC 5) cells.

A specimen of blood for tests for syphilis was taken in all cases.

STATISTICAL ANALYSIS
The data were recorded on precoded proformas and analysed with a computer using the Statistical Package for the Social Sciences. Tests of significance were performed using the χ^2 method with Yates's correction.

Results

A total of 474 women was included in the study.

MICROBIOLOGY
The organisms alone or in combination were isolated as follows: *C trachomatis,* the commonest organism, was found in 158 (33·3%) patients; *N gonorrhoeae* in 149 (31·4%); *C albicans* in 88 (18·6%); *T vaginalis* in 58 (12·2%); and herpes simplex virus in 14 (2·9%). In 150 (31·6%) patients none of these pathogens was isolated while 130 (27·4%) harboured more than one of these organisms.

EPIDEMIOLOGY
Interrelationship between pathogens
The chlamydial isolation rate in cervical specimens was significantly higher in patients with gonorrhoea but was not affected by the other concurrent infections studied (table I). This higher incidence of chlamydia in patients with gonorrhoea was also apparent (but did not reach significance) in the urethral specimens. Nine (39%) of 23 patients with gonococci in the urethra, but only 20 (25%) of 81 without, had chlamydia-positive urethral swabs ($\chi_1^2 = 1·21$; P>0·2). All of the 29 patients whose urethral specimens were chlamydia-positive harboured the organism in the cervix.

Although *T vaginalis* and *Candida* infections showed no correlation with chlamydial isolation rates, patients with gonorrhoea had significantly

TABLE I *Chlamydial isolation rates in the presence or absence of other organisms*

Concurrent organism	Total No of specimens	Chlamydial isolation rate	
		No	%
N gonorrhoeae	104	52	50·0*
C albicans	70	18	25·7
T vaginalis	22	6	27·3
Herpes simplex virus	5	1	20·0
Multiple with N gonorrhoeae	45	20	44·4
Multiple without N gonorrhoeae	20	3	15·0
None	208	58	27·9*
Total	474	158	33·3

*$\chi^2 = 13·90$; P<0·001

fewer candidal infections and significantly more *T vaginalis* infections than patients without gonorrhoea. Of the 149 patients with gonorrhoea, nine (6%) harboured *Candida* and 32 (22%) *T vaginalis*, whereas of 325 patients without gonorrhoea 79 (24%) harboured *Candida* ($P<0\cdot001$) and 26 (8%) *T vaginalis* ($P<0\cdot001$).

To eliminate the possibility of bias introduced by these associations (discussed later) the following categories were considered for making comparisons: (A) *N gonorrhoeae* as the sole pathogen (52 cases); (B) *C trachomatis* as the sole pathogen (58 cases); (C) combined infection (gonococci and chlamydia as the only pathogens) (52 cases); and (D) no pathogens, serving as a control group (150 cases).

Age
Of the groups A, B, and C, 57·7%, 63·7%, and 78·9% respectively were under the age of 25 compared with 51·9% of those belonging to group D. The difference between those with combined infections (group C) and no pathogens (group D) was significant ($\chi^2_1 = 10\cdot41$; $P<0\cdot01$).

Marital state
There was no significant difference in the proportions of single and married women in the four categories.

Occupation
Of those with combined infections (group C), 59·6% were unemployed or in unskilled occupations compared with 40·7% of those with no pathogens (group D); the difference was significant ($\chi^2_1 = 4\cdot85$; $P<0\cdot05$).

Past history of gonorrhoea
No significant differences were found among the four categories.

Last exposure
Significantly fewer (24·1%) of those with chlamydia (group B) had had sexual intercourse during the preceding week than those with gonorrhoea (group A, 48·1%) or those with no pathogens (group D, 44·7%) ($\chi^2 = 5\cdot86$ and $6\cdot57$; $P<0\cdot02$ and $<0\cdot02$ respectively). The rate among those with combined infections (group C) was 33%.

Previous exposure
Information about recent previous sexual exposure to another (other than the regular) partner was available for 277 patients. Of the 44 with gonorrhoea and of the 45 with combined infections 25% and 22·2% respectively had had sexual intercourse with another partner during the previous four weeks compared with only 8·1% of the 135 without pathogens ($\chi^2_1 = 7\cdot25$ and $5\cdot19$; $P<0\cdot01$ and $<0\cdot05$ respectively) and 9·4% of the 53 with chlamydia only.

Oral contraceptives
Of those in groups A, B, and C, 49%, 81·5%, and 67·3% respectively were taking an oral contraceptive compared with 55·8% in group D. Only the difference between those with chlamydia (group B) and those with no pathogens (group D) was significant ($\chi^2_1 = 10\cdot07$; $P<0\cdot01$).

Menstruation
Chlamydial isolation was not significantly affected by the stage of the menstrual cycle.

FEMALE CONSORTS OF MEN WITH URETHRITIS
Gonococcal and chlamydial isolation
C trachomatis was isolated from 64 (49%) of 130 consorts of men with gonorrhoea (56 of these 64 chlamydia-positive women also harboured gonococci); from 52 (43%) of 121 consorts of men with nongonococcal urethritis (NGU); from seven (50%) of 14 consorts of men with presumptive urethritis (that is, men who were alleged to have attended a special clinic and had been treated with tablets or injection); from 27 (17%) of 158 consorts of men on whom no information about urethritis was available; and from eight (13%) of 61 consorts of men who had no urethritis. The difference between the chlamydial isolation rate in consorts of men with urethritis and in consorts whose male partners did not have urethritis was significant ($P<0\cdot001$).

Thirty-one (72%) female consorts of 43 chlamydia-positive men with NGU harboured *C trachomatis* in the cervix compared with eight (22%) consorts of 36 chlamydia-negative men with NGU: the difference was significant ($\chi^2_1 = 17\cdot55$; $P<0\cdot001$). Twenty-three (74%) female consorts of 31 men with combined infections harboured both chlamydia and gonococci.

The chlamydial and gonococcal isolation rates in female consorts of men with urethritis are given in table II according to the type of consort (primary, secondary, or unknown) and to the organisms associated with the man's urethritis. Isolation rates of 100% were found in women who were primary contacts, but the chlamydial isolation rate of 52% among the secondary contacts was lower than the gonococcal isolation rate of 81·2% among the corresponding secondary contacts.

CLINICAL FEATURES
Interrelationship between pathogens
Of 16 women infected with *T vaginalis* only and of 52 infected with *Candida* only, 62·5% and 69·2%

TABLE II *Isolation rates for C trachomatis and N gonorrhoeae in female partners of men with urethritis according to the type of sexual contact and infective agent in men*

	Infective agents (men) and No of female partners infected:			
Type of contact	N gonorrhoeae (28)	C trachomatis (43)	Both (31)	NGU/no chlamydia (36)
Primary contact				
Total	6	8	6	17
Gonococci	4	0	0	1
Chlamydia	0	7	0	3
Both	2	1	6	0
Isolation rate (%)	100 (G)	100 (C)	100 (G + C)	
Secondary contact				
Total	16	25	18	10
Gonococci	9	0	4	0
Chlamydia	0	12	1	1
Both	4	1	12	0
Isolation rate (%)	81·2 (G)	52·0 (C)	66·7 (G + C)	
Status unknown				
Total	6	10	7	9
Gonococci	3	0	2	0
Chlamydia	0	10	0	4
Both	1	0	5	0
Isolation rate (%)	66·7 (G)	100 (C)	71·4 (G + C)	
Grand total				
Total	28	43	31	36
Gonococci	16	0	6	1
Chlamydia	0	29	1	8
Both	7	2	23	0
Isolation rate (%)	82·1 (G)	72·1 (C)	74·2 (G + C)	

G = gonococci
C = chlamydia

respectively complained of vaginal discharge or irritation or both compared with 17 (32·6%) and 21 (36·2%) respectively of those harbouring gonococci and chlamydia. The differences between those with *Candida* and those with gonorrhoea, and between those with *Candida* and those with chlamydia, were significant ($\chi^2_1 = 12\cdot47$ and $10\cdot69$; $P<0\cdot001$ and $<0\cdot01$ respectively). When compared with the group with no pathogens (group D) *T vaginalis* infection was significantly associated with vaginal discharge (34/150 and 8/16 respectively; $\chi^2_1 = 4\cdot36$; $P<0\cdot05$) Hence, patients with *Candida* and *T vaginalis* must be excluded when the clinical features of chlamydial and gonococcal infections are studied. Hence the same four categories defined above are retained here.

Symptoms
Thirty-five (67·3%), 37 (63·8%), and 31 (59·6%) respectively of those with gonococci, chlamydia, and combined infections were asymptomatic compared with 78 (52%) of those with no pathogens; the differences were not significant. The complaint of vaginal discharge was also almost equally common among all the four groups (range 21·1-26·9%).

Fifty-two patients had urinary symptoms and there was no difference in this respect among the four groups.

Signs
Cervical ectopy. Chlamydia, whether in the presence or absence of gonococci, were found significantly more often in patients with cervical ectopy (table III). No such association with ectopy was found in those infected with gonorrhoea, *Candida*, or *T vaginalis*.

Interrelationship between oral contraception, cervical ectopy, and chlamydial isolation. Of the 274 women who were taking an oral contraceptive, 151 (55·1%) had cervical ectopy compared with 56 (30·4%) of 184 who were not; the difference was significant ($\chi^2_1 = 26\cdot07$; $P<0\cdot001$). We had noted earlier (see above) a significant association between the use of an oral contraceptive and the isolation of chlamydia. It was, therefore, necessary to examine the effect of oral contraceptives and cervical ectopy independently of each other on the chlamydial isolation rate. (Patients with concomitant infections were excluded.) The results were:

(a) If no oral contraceptive was used and cervical ectopy absent, *C trachomatis* was isolated in none of 53 patients;

(b) If a contraceptive was used and cervical ectopy absent, *C trachomatis* was isolated in 14 (23·7%) of 59 patients;

TABLE III *Presence or absence of cervical ectopy in relation to isolation of infective agents*

Ectopy	Total	Gonococci		Chlamydia		Both organisms		Candida		Trichomonads		No pathogen	
		No	%	No	%	No	%	No	%	No	%	No	%
Present	170	18	34·6	42	72·4	36	69·2	15	28·9	9	56·3	50	33·3
Absent	210	34	65·4	16	27·6	16	30·8	37	71·1	7	43·7	100	66·7
Total	380	52	100	58	100	52	100	52	100	16	100	150	100

Chlamydia *vs* no pathogen: $\chi^2 = 24\cdot34$; P<0·001
Gonococci and chlamydia *vs* no pathogen: $\chi^2 = 18\cdot91$; P<0·001

(c) If no oral contraceptive was used and cervical ectopy was present, *C trachomatis* was isolated in 10 (45·5%) of 22 patients; and

(d) If an oral contraceptive was used and cervical ectopy was present, *C trachomatis* was isolated in 30 (44·8%) of 67 patients.

Cervical discharge. Mucopurulent discharge in the endocervix was seen in 23·1% of those with gonococci, 22·4% of those with chlamydia, and 30·8% of those with combined infection compared with only 4% of those with no pathogens. The difference between each infected group and group D was significant ($\chi_1^2 = 15\cdot68$, 14·62, and 24·74; P<0·001 in each case). Of those infected with *T vaginalis*, 18·7% also had a mucopurulent discharge in the endocervix.

Cervicitis. Using Paavonen's criterion[5] (congested cervix and hypertrophic ectopy usually associated with mucopus) cervicitis was found in 23 (39·6%) of 58 cases infected with chlamydia and 19 (36·5%) of 52 with combined infections compared with only 20 (13·3%) of 150 in group D. The difference was significant in each case ($\chi_1^2 = 16\cdot10$ and 11·89; P<0·001 and <0·001). But the difference between those with gonococci (21·1% with cervicitis) and group D was not significant.

Salpingitis. Using diagnostic criteria based on those of Simmons *et al*[6] eight patients were considered to have salpingitis; two harboured chlamydia only, four gonococci only, and the remaining two both chlamydia and gonococci.

Discussion

The pattern of significant association between the isolation of *N gonorrhoeae* and *C trachomatis* but not between the presence of chlamydia, *Candida*, and *T vaginalis* was strikingly similar to that reported by Hilton *et al*.[7] This association seemed even more remarkable in the absence of cervical ectopy, with 32% of women being chlamydia-positive in the

presence of gonococci compared with only 13·8% without (table III; $\chi_1^2 = 6\cdot32$; P<0·02). We also found a significant negative association between *N gonorrhoeae* and *C albicans* and a positive association between *N gonorrhoeae* and *T vaginalis*. Whether these associations and others, such as a high coincidence of chlamydial infection with *T vaginalis* noted by Kinghorn and Waugh[8] and Tait *et al*,[9] act in an inhibitory or symbiotic way is not clear. In addition, *C albicans* and *T vaginalis*, although predominantly infecting the vagina, may cause erythema of the ectocervix and an ectocervicitis or endocervicitis respectively.[10-12] These associations are therefore, potentially capable of introducing bias; thus the study of epidemiological and clinical features must (as in this paper) be related separately to pathogens existing singly or in specified combinations. This fact must also be borne in mind when comparisons with other studies are made.

We found that cervicitis was more frequently associated with chlamydial than with gonococcal infection. As also observed by Rees,[13] cervicitis in several cases with combined infections (chlamydia and gonococci) persisted after treatment of gonorrhoea with spectinomycin or single-dose regimens of penicillin (postgonococcal cervicitis) (Arya *et al*, unpublished data).

Apart from the association with *N gonorrhoeae*, the most outstanding correlates of chlamydial infection were the use of an oral contraceptive and cervical ectopy—acting independently of each other—but cervical ectopy had the more dominating influence since in its presence the addition of an oral contraceptive did not make any difference to the chlamydial isolation rate. These findings were at variance with those of Tait *et al*[9] in their study of female contacts of men with NGU. Oral contraceptives may produce microscopic and macroscopic changes in the cervix[14]—namely, hyperplasia and hypersecretion of the cervical glands, enlargement of columnar villi, hyperaemia and oedema of the columnar epithelium, and eversion of the endocervix. Some of these changes, especially in the presence of ectopy (which is simply an extension of the

endocervical epithelium), may provide a favourable environment for infection by *C trachomatis*.

Our finding that *N gonorrhoeae* was not significantly associated with cervical ectopy may possibly suggest that columnar epithelium is not an important requirement for infection by this organism. Indeed, Evans[15] in his ultrastructural study of cervical gonorrhoea found gonococci to be firmly attached to stratified squamous epithelium and not to the mucus-secreting columnar epithelium.

The 100% isolation rates of *N gonorrhoeae, C trachomatis,* and both organisms in patients with combined infections among the primary consorts of men infected with the corresponding organism(s) was to be expected and showed our accurate identification of contacts, which is not always possible, however. The lower isolation rate of *C trachomatis* than of *N gonorrhoeae* among the secondary contacts would be expected if *N gonorrhoeae* was more infectious than *C trachomatis* (suggested recently by Lycke *et al*[16]). However, transmission of infection can be influenced by the organism and by the host, and an important consideration here, among other determinants, must be the presence of cervical ectopy in the potential recipients.

Clinical conclusions

In this study 158 cases of chlamydial infection were identified, and one of the aims was to find correlates which might help in the management of these cases in the absence of laboratory facilities. As gonococcal infection is often accompanied by non-specific or chlamydial infection it has been proposed that gonorrhoea should be treated with a regimen which will be effective against both conditions.[17] In our study of 474 women this suggestion would have led to the treatment of 72 of the 158 chlamydia-positive patients but also to treating unnecessarily 77 patients who had gonorrhoea but no chlamydial infection. A further eight chlamydia-positive patients would have been recognised and similarly treated if 20 women who were consorts of men with gonorrhoea, but themselves without gonorrhoea, had been treated epidemiologically.

It has also been proposed that all female consorts of men with NGU should be treated on epidemiological grounds,[18] which would have included 48 further cases of chlamydial infection in our population by treating a further 114 women. We would then have been left with 30 cases of chlamydial infection out of the remaining total of 191 women who were not known to be contacts of men with urethritis. How could these cases (and their contacts) be identified? As shown earlier, age, marital state, occupation, symptoms, and past history of gonorrhoea by

themselves were unhelpful indices. However, among the remaining 191 women were 69 who did not have cervical ectopy and were not taking an oral contraceptive, and only one of these was chlamydia-positive. In contrast there were 15 chlamydia-positive patients among the 46 who had both cervical ectopy and were using an oral contraceptive. Only three of these 15 chlamydia-positive patients had cervicitis. Unfortunately, therefore, the predictive value of all the clinical and epidemiological indices studied, even when used in combination, remains unacceptably low. Thus the 30 cases (mentioned above), constituting 15·7% of the 191 women, might not have been treated (a figure very similar to 12% reported recently by Richmond *et al*).[19]

Even if laboratory facilities for chlamydial isolation become available, there will still be about the same number of cases, as those infected with *C trachomatis*, of non-specific genital infection which will remain unidentified and untreated (compare: half of the cases of NGU in men whose aetiology remains unknown). For female patients the answer lies in the provision of a laboratory service for chlamydial isolation as well as treatment of consorts of men with urethritis; otherwise these infections may continue unchecked.

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