

Prevalence of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* infections in Greenland

A seroepidemiological study

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SUMMARY In a sero-epidemiological study of the prevalence of chlamydial and gonococcal infections in Greenland three groups of subjects were studied—262 patients attending an outpatient department in the town of Nuuk (of whom 12% harboured *Chlamydia trachomatis* and 54% *Neisseria gonorrhoeae*), 63 controls from the same town, and the entire population of 150 in the settlement of Uvkusigat. Using a microimmunofluorescence test evidence of exposure to *C trachomatis* was found in 79% of the female and 26% of the male patients, in 12% and 5% of the female and male controls respectively, and in 51% and 21% of the female and male population of Uvkusigat respectively. Using an indirect haemagglutination test antibodies to gonococcal pili were found in sera of 92% of the female and 70% of the male patients, in 30% of the male and 10% of the female controls, and in 41% of the women and 33% of the men in Uvkusigat. The study indicates that genital chlamydial and gonococcal infections are serious public health problems in Greenland and that such infections are acquired early in both sexes and often occur concomitantly.

Introduction

Gonorrhoea, syphilis, and (since 1977) chancroid are serious public health problems in Greenland; in 1977 the number of reported cases in a population of about 50 000 were 10 095, 674, and 975 respectively.¹ This means that morbidity for these diseases is 100-fold greater in Greenland than in Denmark. In industrialised countries, genital infections due to *Chlamydia trachomatis* probably occur at least as frequently as gonorrhoea.² Information about the prevalence of genital chlamydial infections in developing countries is however still scanty.

Patients attending an outpatient department in the town of Nuuk, Greenland, were examined for chlamydial and gonococcal infections by culture and serological tests. In addition, serological studies were performed both in a group of subjects from Nuuk who had no history of sexually transmitted diseases (STDs) and in the entire population of the settlement

of Uvkusigat in the district of Umanaq. On the basis of the results obtained, the value of seroepidemiological studies of STDs in developing countries is discussed.

Patients and methods

STUDY POPULATIONS

Group 1 (patients)

A total of 262 patients attending the outpatient clinic, Nuuk (Godthåb), Greenland, were studied (group 1, patients). The investigation was carried out during two periods—September to November 1977 and September to October 1978. During the first period, 97 male and 97 female patients were included in the study and during the second, 35 and 33 respectively. The age distribution of the patients, who were consecutive cases with symptoms and signs of genital infection attending the clinic during the two study periods, are given in fig 1. One-third of the men were of Danish origin compared with only one woman.

Previous gonococcal infection occurred in 92% of female and 94% of male patients. The number of earlier episodes of gonorrhoea varied from 0 to 89

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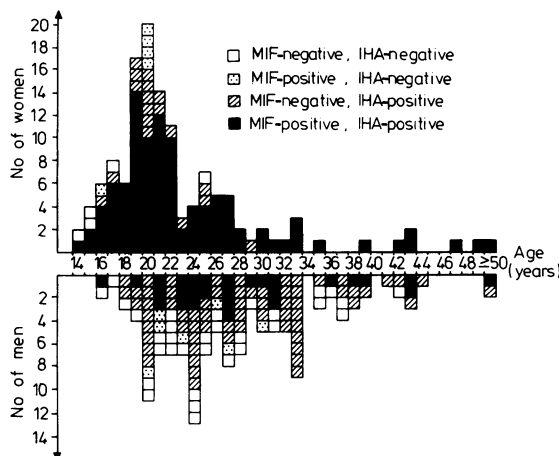


FIG 1 Results of microimmunofluorescence (MIF) tests for IgM and IgG chlamydial antibodies and of indirect haemagglutination (IHA) tests for gonococcal pili antibodies in relation to sex and age of 262 patients (group 1)

with a mean of eight episodes. The number of patients with current gonococcal infection is reported below. Non-gonococcal genital infections occurred in 28 (21%) of the 132 male and in 46 (35%) of the 130 female patients. Primary syphilis and chancroid were each diagnosed in eight patients whereas herpes genitalis and salpingitis of unknown aetiology each occurred in a further two patients. No evidence of current venereal or genital infection was shown in 24 (9.2%) of the 262 patients; all 24 patients were sexual contacts of patients in whom a STD had been diagnosed.

Group 2 (controls)

For comparison, 63 subjects (20 men and 43 women) from the Nuuk region, without any history of STD, were studied (group 2, controls). Eighteen of the 20 men and 16 of 27 women were of Danish origin.

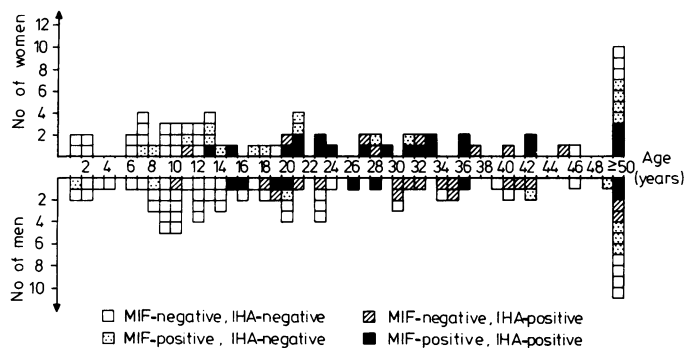


FIG 2 Results of microimmunofluorescence (MIF) tests for IgM and IgG antibodies to *C trachomatis* and of the indirect haemagglutination (IHA) tests for gonococcal pili antibodies in relation to sex and age of 150 controls (group 3)

Group 3 (controls)

In addition, the entire population (80 men and 70 women) of Uvkusigsat, a settlement located far north of Nuuk on the west coast of Greenland, was studied (group 3, controls). In October 1977, blood samples had been collected from this population during a seroepidemiological survey because of a case of typhoid fever. The age distribution of the population is given in fig 2.

CULTURAL TECHNIQUES

Specimens from 194 of the 262 patients (group 1, first period) attending the outpatient clinic in Nuuk were cultured for *C trachomatis* and *N gonorrhoeae* while those from the remaining 68 patients (group 1, second period) were cultured solely for gonococci. No specimens from either group 2 or group 3 were cultured.

C trachomatis

Specimens for the isolation of chlamydia were collected from the urethra of men and from the cervix of women using a calcium-alginate swab (Medical Wire and Equipment Co, UK). The specimens were transported frozen (see also Discussion) to the laboratory in Sweden in a sucrose-phosphate buffer (2-SP) containing gentamicin 10 µg (Schering) and amphotericin B 2.5 µg (Squibb) per ml. Cycloheximide-treated McCoy cells were used for the isolation of chlamydia according to a method described elsewhere,³ except that RPMI 1640 was used as cell culture medium. The cells were cultured on coverslips 13 mm in diameter.

N gonorrhoeae

Specimens for the culture of gonococci were collected from the urethra of men and from the urethra, cervix, and rectum of women. All samples were collected using charcoal-impregnated swabs and immediately inoculated on to a selective chocolate agar medium.⁴ *N gonorrhoeae* was identified by

colonial morphology, oxidase reaction, and Gram staining.

SEROLOGICAL TESTS

Sera from all three groups were tested for antibodies to *C trachomatis* and *N gonorrhoeae*.

C trachomatis

Serum IgG and IgM antibodies to *C trachomatis* were demonstrated by the microimmunofluorescence (MIF) test using pooled antigens according to the method described by Treharne *et al.*⁵ The presence of IgM antibodies at a titre of ≥ 8 or IgG antibodies at a titre of ≥ 32 or both was considered a positive result.

N gonorrhoeae

Antibodies to gonococcal pili were determined by an indirect haemagglutination (IHA) test performed as previously described.⁶ The antigen was the same as that used in two earlier studies.^{7,8} Titres ≥ 40 were considered a positive result.

Results

CULTURE (GROUP 1)

Chlamydia were recovered from 12 (12%) of the 97 male and 11 (11%) of the 97 female patients examined in 1977. Of these 23 positive specimens, 20 showed less than 20 inclusions and three 25-40 inclusions per coverslip.

Gonococci were recovered from 84 (63%) of the 132 male and from 58 (45%) of the 130 female patients during 1977 and 1978. Of the 23 patients harbouring *chlamydia*, 10 (43%) also had gonorrhoea.

SEROLOGY

C trachomatis

Group 1. IgM antibodies to this organism, at a titre of ≥ 8 , occurred as infrequently in the 132 (7%) male as in the 130 (6%) female patients. IgG antibodies at a titre of ≥ 32 were detectable more often in the women (79%) than in the men (26%) (table I).

Of the 23 *chlamydia*-positive patients, five had IgG antibodies at a titre of < 32 whereas 3, 5, 2, 2, 3, and 1 had antibodies at a titre of 32, 64, 128, 256, 512, and 2048 respectively. One patient had a IgG titre of 64 and a IgM titre of 32 whereas the remaining patient had only IgM antibodies (titre 32).

Group 2. Of the 63 controls in this group none had IgM antibodies to *C trachomatis*. IgG antibodies to this organism at a titre of ≥ 32 occurred in one man and five women (table I).

Group 3. Of the 80 men and 70 women from Uvkusigsat 17 (21%) and 36 (51%) respectively had IgG antibodies to *C trachomatis*.

N gonorrhoeae

The results of the IHA tests for antibodies to gonococcal pili in the three groups are shown in table II.

Group 1. Positive results were found for 70% of the male and 92% of the female patients. The results of the IHA test in relation to previous or current gonococcal infections or both in the male and female patients are given in table III. Sixty-one (92%) of 66 female patients in whom gonococci could not be isolated but who had a previous history of gonorrhoea had a positive result. The corresponding figure for the male patients was 61%.

Group 2. Sera from two (10%) of the 20 male and 13 (30%) of the 43 female controls gave positive results to the IHA test.

Group 3. Positive IHA test results were found in 26 (33%) of the 80 men and in 29 (41%) of the 70 women.

TEST RESULTS IN RELATION TO AGE

The results of the MIF and IHA tests in relation to sex and age of groups 1 and 3 are given in figs 1 and 2 respectively.

Discussion

The prevalence of sexually transmitted diseases in Greenland is similar to that estimated for several

TABLE I Titres of IgG antibodies to *C trachomatis* in sera from patients (group 1), controls from Nuuk (group 2), and the population of Uvkusigsat (group 3)

Group	Sex	No	With antibody titre										With positive result (titre ≥ 32)
			≤ 16	32	64	128	256	512	1024	2048	4096	8192	
1	M	132	98	18	11	3	0	2	0	0	0	0	34
	F	130	27	19	19	21	21	11	4	3	4	1	103
2	M	20	19	1	0	0	0	0	0	0	0	0	1
	F	43	38	1	3	1	0	0	0	0	0	0	5
3	M	80	63	5	7	3	2	0	0	0	0	0	17
	F	70	34	6	10	9	3	7	1	0	0	0	36

TABLE II *Titres of antibodies to gonococcal pili in sera from patients (group 1), controls from Nuuk (group 2), and the population of Uvkusigsat (group 3)*

Group	Sex	No	With antibody titre									With positive result (titre ≥ 40)
			≤ 20	40	80	160	320	640	1280	2560	5120	
1	M	132	39	20	25	21	13	11	1	1	1	93
	F	130	11	13	20	15	20	27	14	10	0	119
2	M	20	18	0	1	1	0	0	0	0	0	2
	F	43	30	7	4	1	0	1	0	0	0	13
3	M	80	54	6	9	6	5	0	0	0	0	26
	F	70	41	8	9	6	2	3	1	0	0	29

TABLE III *Results of indirect haemagglutination tests for gonococcal pili antibodies in 132 male and 130 female patients (group 1) from the outpatient clinic, Nuuk, in relation to history of gonorrhoea*

History of gonorrhoea		No of patients	Test result	
Current	Previous		Positive*	Negative
Male patients:				
Yes	Yes	78	62	16
	No	5	2	3
No	Yes	46	28	18
	No	3	1	2
Total		132	93	39
Female patients:				
Yes	Yes	53	49	4
	No	5	4	1
No	Yes	66	61	5
	No	6	5	1
Total		130	119	11

*A titre of ≥ 40 is considered a positive test result

developing countries. In contrast to most of these countries, Greenland has a medical infrastructure which allows a comparatively precise review of the problem.

A number of socioeconomic factors contribute to the high prevalence of STDs in Greenland, such as rapid urbanisation of the society, excessive alcohol consumption, and liberal sexual habits. Immigration of young single male guest workers from Denmark certainly influences the epidemiological pattern. The pattern for gonorrhoea in Greenland differs from that in many developed countries—that is, people over 30 years of age, and also young children between 7 and 12 years, not infrequently acquire gonococcal infection.

Until now beta-lactam antibiotics, with a few exceptions, have been prescribed to patients in Greenland in whom an STD had been diagnosed; this is a treatment which is not effective against chlamydial infections. Therefore, it should be possible to study the natural spread of chlamydial infections in this community. Of conditions associated with chlamydial infection, non-gonococcal urethritis is the most frequently diagnosed whereas lymphogranuloma venereum has not yet been encountered in Greenland.

Our attempts to diagnose chlamydial infections by culture were only partly successful. The low number

of chlamydial inclusions found in the McCoy cell cultures may suggest that the specimens from the patients in Nuuk had been thawed during transportation to the laboratory in Sweden. Thus, the percentage of patients in whom a current chlamydial infection was found must be regarded as the minimum figure for the true frequency of such infections in the group of subjects studied. The low chlamydial isolation rate compared with the frequent finding of antibodies to the organism might also be due to an immune response that had eradicated the organisms or to the fact that chronic genital infections with only a few chlamydia and specific antibodies in urethral and cervical secretions were common in these patients. In experimental studies, antibodies to chlamydia were found to interfere with the formation of chlamydial inclusions in cell cultures.¹⁰

Culture studies have shown that double infections with *C trachomatis* and *N gonorrhoeae* commonly occur in STD clinic patients.² In Lund, Sweden, we have found 1/4-1/5 of such patients harbouring both these organisms. *N gonorrhoeae* was recovered from approximately half of the 23 patients in the present series with positive culture results for *C trachomatis*.

The presence of serum IgM antibody to *C trachomatis* is associated with an early immune response to experimental genital infections in

monkeys. After inoculation of *C trachomatis* into the uterine cavity or the Fallopian tubes of grivet monkeys, we found that IgM antibodies could be demonstrated for 5-8 weeks after the infection whereas IgG antibodies were still present after 2-3 months—that is, when the experiments were completed.¹¹ However, in STD clinic patients, the search for serum IgM antibodies to *C trachomatis* seems to have limited value in the diagnosis of current genital infections due to this organism. In the present study, all except two of the 23 chlamydia-positive patients had no IgM antibodies to *C trachomatis*. The low frequency of IgM antibodies to *C trachomatis* in the patients from Nuuk—that is, in 7% of male and 6% of female patients—might suggest that most of these subjects had been previously exposed to chlamydia. In reinfections or in long-standing infections, IgG antibodies are more likely to occur.

It may be argued which titre of IgG antibodies to *C trachomatis* should be considered "positive" in MIF tests. In the present study, which is a sero-epidemiological survey, and in which we did not intend to establish the incidence of current chlamydial infections, we considered a titre of 32 to be a positive result.

Serum antibodies to *C trachomatis* occur more often in female than in male clinic patients.⁹ This was also true for the outpatients in Nuuk. Thus such IgG antibodies occurred in 79% of the female but in only 26% of the male patients. Similar figures of seropositive men and women have been reported in STD clinic patients from the United Kingdom and United States.^{2,12} The observed sex difference agrees with the result of serological studies of other agents causing STDs and seems to be due to a more efficient antigen stimulation in women than in men.

In the population of Uvkusigsat, serological evidence of exposure to chlamydia was demonstrated in 79% of the women over 12 years of age; the corresponding percentage in the men was 24. These findings suggest that chlamydial infection is a common occurrence in communities in Greenland.

The finding that six of the 10 women of Uvkusigsat, who were over 50 years of age, had high titres of IgG antibodies to *C trachomatis* might suggest that such antibodies may persist for many years or that women at this age are also exposed to the possibility of acquiring STDs.

The high prevalence of previous as well as current gonococcal infection in the patients in Nuuk was associated with the presence of antibodies to gonococcal pili in most of the sera in both the male (70%) and female (92%) patients.

In the controls, with one exception, such antibodies only occurred in women of local origin. These women had probably been previously exposed to

gonococci. The correlation between age and the results of the IHA tests suggests that antibodies to gonococcal pili only occur in the age groups exposed to gonococcal infections and that the antibodies persist for years. These results agree with those obtained in previous studies.^{7,8}

In a community with a high prevalence of STDs, the demonstration of serum antibodies to *C trachomatis* and *N gonorrhoeae* (at least with the tests used in the present study) is not a suitable diagnostic procedure for the establishment of current infections with these agents. However, the MIF and IHA tests used seem to be valuable for sero-epidemiological surveys in areas where, for instance, attempts are made to establish the impact of sexually transmitted diseases on fertility.

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