



# ***Chlamydia pneumoniae* and *Mycoplasma pneumoniae*: Opportunistic Infectious Agents in HIV/AIDS**

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## **Abstract**

**T**he objective of this study was to investigate the prevalence of *Chlamydia pneumoniae* and *Mycoplasma pneumoniae*, correlated with AIDS patients. Ninety-seven cases of HIV infection were described. *C. pneumoniae* and *M. pneumoniae* infections were serologically diagnosed by micro-immunofluorescence test using standard diagnostic criteria and an agglutination test (SERODIA-MYCOII), respectively. The prevalence of antibodies to *C. pneumoniae* and *M. pneumoniae* was 87.6% (85/97) and 45.4% (44/97), respectively. By age, the group aged 21-30 years had the highest rates of infection with *C. pneumoniae* [44 (45.4%)] and *M. pneumoniae* [24 (24.7%)]. Injecting drug users had the highest rates of infection with *C. pneumoniae* [68 (70.1%)] and *M. pneumoniae* [39 (40.2%)]. *C. pneumoniae* subtype E was the most prevalent infectious agent [48 (49.5%)], followed by *M. pneumoniae* [22 (22.7%)]. The results of this study indicated that *C. pneumoniae* may play a role in the etiology of respiratory-tract infections among HIV-positive patients.

**Keywords:** *Chlamydia pneumoniae*, *Mycoplasma pneumoniae*, HIV/AIDS

## **Introduction**

During the last decade, *Chlamydia pneumoniae* and *Mycoplasma pneumoniae* have emerged as important causes of respiratory infections in human immunodeficiency virus (HIV) infected patients [1-4], where community-acquired pneumonia accounts for approximately 10% of infections among these patients [5,6]. Moreover, an association has been suggested between *C. pneumoniae* infection and coronary heart disease [7]. In Thailand, HIV is one of the major causes of morbidity and mortality; yet little is documented about the prevalence of *C. pneumoniae* or *M. pneumoniae* infections among HIV patients. Thus, the aims of this study were to determine the prevalence of *C. pneumoniae* and *M.*

*pneumoniae* among HIV patients, and to investigate any potential relationship between *C. pneumoniae* and *M. pneumoniae* infections and HIV patients of different age groups, injecting drug users, and HIV subtypes.

## **Materials and methods**

### **Clinical specimens**

Ninety-seven patients were assessed by the laboratory of Thanyarak Hospital (hospital for treatment of drug addicts) and the HIV/AIDS laboratory of the Thai National Institute of Health (NIH) to determine HIV-1 subtype distribution among injecting drug users seeking medical care at the hospital. The hospital agreed to send

blood specimens from injecting drug users for the years 1994, 1997, 1999, and 2000 to the HIV/AIDS laboratory. Confidential counseling and testing services are provided on a voluntary basis with written consent from patients [8]. The specimens were obtained from every patient seeking medical care for a period of 6 consecutive months in each of the specified years. They were initially tested for HIV-1 serostatus (HIV-1/HIV-2 Third Generation. Abbott GmbH Diagnostika, Germany), with confirmation of HIV-1 subtyping (V3-loop peptide enzyme immunoassay (PEIA)) using the modified method of Pau [9].

## Serology

### *Chlamydia pneumoniae* MIF

All sera were investigated for the presence of immunoglobulins (Ig), namely, IgA, IgM, and IgG antibodies to *Chlamydia* species (*C. pneumoniae*, *C. trachomatis*, and *C. psittaci*) using the microimmunofluorescence (MIF) test [10]. Antigen was provided by the National Institute of Health (NIH), Japan. Serum antibodies against *C. pneumoniae* elementary bodies were detected with fluorescein-conjugated monoclonal goat antihuman Ig antibodies. Based on the criteria of Verkooyen *et al* [11] and Niedzwiadek *et al* [12], IgM titers  $\geq 1:8$ , IgG titers  $\geq 1:512$  (single serum) and fourfold rising titers (paired sera) were considered to be acute infections. Meanwhile, IgG titers  $\geq 1:128$  and IgA  $\geq 1:32$  were considered chronic infections or re-infections, and IgG titers of 1:64-1:256 and IgA  $\leq 1:8$ , as past infections.

### *Mycoplasma pneumoniae* agglutination

The in-vitro diagnostic test for detecting antibodies to *M. pneumoniae* (Fujirebio Inc, Japan) uses artificial gelatin particles sensitized with cell membrane components of *M. pneumoniae* (Mac strain); the test is based on the principle that sensitized particles are agglutinated by the presence of antibodies to *M. pneumoniae* in human serum. The criterion for a positive infection is a titer  $\geq 1:40$  [13].

## Results

In ninety-seven sera samples, the prevalence of antibodies to *C. pneumoniae* and *M. pneumoniae* in HIV patients was 87.6% (80.6-94.6, 95%CI) and 45.4% (36.4-54.4, 95%CI), respectively. *C. pneumoniae* and *M. pneumoniae* infections among HIV patients classified by age group, are shown in Table 1. Age groups 11-20, 21-30, 31-40, and 51-60 years were *C. pneumoniae* and *M. pneumoniae* -infection-positive, as follows: [8 (8.2%), 4 (4.1%)], [44 (45.4%), 24 (24.7%)], [27 (27.8%), 12 (12.4%)] and [6 (6.2%), 4 (4.1%)], respectively. *C. pneumoniae* and *M. pneumoniae* infections among HIV patients by drug type, are shown in Table 2. IDU and Met were *C. pneumoniae* and *M. pneumoniae* -infection-positive, as follows: [68 (70.1%), 39 (40.2%)] and [17 (17.5%), 5 (5.1%)]. *C. pneumoniae* and *M. pneumoniae* infections among HIV patients classified by HIV subtype are shown in Table 3. Type B', type E, type dual, and negative were *C. pneumoniae* and *M. pneumoniae* -infection-positive, as follows: [21

**Table 1** *C. pneumoniae* and *M. pneumoniae* infections among HIV patients, by age group.

Age group	<i>C. pneumoniae</i> result					<i>M. pneumoniae</i> result		
	Acute infection (%)	Chronic or re-infection (%)	Past infection (%)	Negative (%)	Total (%)	Positive (%)	Negative (%)	Total (%)
11-20	6 (6.2)	-	2 (2.1)	2 (2.1)	10 (10.3)	4 (4.1)	5 (51.5)	9 (9.3)
21-30	30 (30.9)	3 (3.1)	11 (11.3)	3 (3.1)	47 (48.5)	24 (24.7)	24 (24.7)	48 (49.5)
31-40	18 (18.6)	1 (1.0)	8 (8.2)	3 (3.1)	30 (30.9)	12 (12.4)	18 (18.6)	30 (30.9)
41-50	6 (6.2)	-	-	3 (3.1)	9 (9.3)	4 (4.1)	5 (51.5)	9 (9.3)
51-60	-	-	-	1 (1.0)	1 (1.0)	-	1 (1.0)	1 (1.0)
Total	60 (61.9)	4 (4.1)	21 (21.6)	12 (12.4)	97 (100)	44 (45.4)	53 (54.6)	97 (100)

**Table 2** *C. pneumoniae* and *M. pneumoniae* infections among HIV patients, by drug type.

Drug type	<i>C. pneumoniae</i> result					<i>M. pneumoniae</i> result		
	Acute	Chronic or	Past	Negative	Total	Positive	Negative	Total
	infection	re-infection	infection	(%)	(%)	(%)	(%)	(%)
IDUs	49 (50.5)	4 (4.1)	15 (15.5)	12 (12.4)	80 (82.5)	39 (40.2)	41 (77.3)	80 (82.5)
Met	11 (11.3)	-	6 (6.2)	-	17 (17.5)	5 (5.1)	12 (12.4)	17 (17.9)
Total	60 (61.8)	4 (4.1)	21(21.7)	12 (12.4)	97 (100)	44 (45.3)	53 (54.7)	97 (100)

IDUs = Injecting drug users

Met = Metamphethamine

**Table 3** *C. pneumoniae* and *M. pneumoniae* infections among HIV patients, by HIV subtype.

HIV subtype	<i>C. pneumoniae</i> result					<i>M. pneumoniae</i> result		
	Acute	Chronic or	Past	Negative	Total	Positive	Negative	Total
	infection	re-infection	infection	(%)	(%)	(%)	(%)	(%)
Type B'	18 (18.5)	-	3 (3.2)	6 (6.2)	27 (27.9)	14 (14.4)	13 (13.4)	27 (27.8)
Type E	30 (30.9)	4 (4.1)	14 (14.4)	5 (5.1)	53 (54.5)	22 (22.7)	31 (32.0)	53 (54.7)
Type Dual	-	-	1 (1.0)	-	1 (1.0)	1 (1.0)	0	1 (1.0)
Negative	12 (12.4)	-	3 (3.2)	1 (1.0)	16 (16.6)	7 (7.2)	9 (9.3)	16 (16.5)
Total	60 (61.8)	4 (4.1)	21 (21.8)	12(12.3)	97(100)	44 (45.3)	53 (54.7)	97 (100)

(21.6%), 14 (14.4%)], [48 (49.5%), 22 (22.7%)], [1 (1.0%), 1 (1.0%)], [15 (15.5%), and 7 (7.2%)], respectively.

## Discussion

Miller [14] reported that bacterial infections, which occur more frequently among HIV-infected persons than the general population, and tuberculosis, are increasingly causes of morbidity and mortality. This study found a high prevalence of antibodies to *C. pneumoniae* in HIV-positive individuals, as reported by Blasi *et al* [15]. The study also found that the co-infection rate for *C. pneumoniae* and *M. pneumoniae* was 38/97 (39.1%). The prevalence of atypical agents causing pneumonia among HIV-infected patients is unknown, as no evaluated routine method for diagnosing these agents among HIV-positive patients currently exists.

The study of Koulla-Shiro [4] found that the primary etiological agent of pneumoniae in 50-52.4% of HIV-infected patients was *Streptococcus pneumoniae*; the second was *M. pneumoniae* (8.8-12%). *C. pneumoniae* appears to be involved in respiratory tract infections in HIV-1-infected subjects. Our data suggested that *C. pneumoniae* and *M. pneumoniae* should be included in the diagnostic approach for respiratory infections in HIV-infected subjects.

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