INCIDENCE OF *MYCOPLASMA PNEUMONIAE, CHLAMYDIA TRACHOMATIS,* AND VIRAL INFECTIONS IN PNEUMONIA CASES UNDER SIX MONTHS OF AGE, BANGKOK, THAILAND

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Abstract. The incidence of infections by *Mycoplasma pneumoniae, Chlamydia trachomatis* and respiratory viruses was investigated in 76 pneumonic patients aged under 6 months who attended Ramathibodi and Siriraj Hospitals in Bangkok during two study periods. *M. pneumoniae* infection was not found in any case from either hospital by serological diagnosis. By the isolation method, *C. trachomatis* infection was found in 7(16.7%) of 42 patients from Ramathibodi Hospital and 5(21.7%) of 23 patients from Siriraj Hospital with the average male:female raio of 2.6:1; and 91.7% of the infected cases were under 3 months old. Laboratory diagnosis of respiratory virus infection was performed by indirect immunofluorescence (IIF), isolation, and by antibody detection. Data from Ramathibodi Hospital showed that 11 (24.4%), 4 (8.9%), 3 (6.7%), and 3 (6.7%) of the 45 patients were infected by respiratory syncytial virus (RSV), adenoviruses, parainfluenza virus type3, and some other viruses, respectively; infection rates of 10 (32.3%), 4 (12.9%), 1 (3.2%) and 1 (3.2%) by those viruses respectively, were observed in the 31 patients from Siriraj Hospital.

INTRODUCTION

Viruses, bacteria, parasites and fungi are pathogenic agents in acute respiratory infection (ARI). A single agent can elicit different clinical manifestations varying from mild upper respiratory infection (URI) to severe lower respiratory infection (LRI). Conversely, the same disease may be caused by several etiologic agents (Marks, 1985). In general, certain agents may predominate over the others depending on the clinical entity, age-group, immune status and seasonality (WHO Scientific Group, 1980; Marks, 1985). Nevertheless, the causative agent cannot be diagnosed on the basis of these features.

Pneumonia is a serious manifestation of ARI in young children worldwide. In Thailand, ARI is the second most common disease next to diarrhea in children under 5 years of age, but the death rate due to ARI is higher than that caused by diarrhea. During 1977 to 1986, the annual mortality rate of pneumonia varied from 7 to 23 deaths per 100,000 children aged 1-4 years, and was 61 to 147 deaths per 100,000 children younger than one year (Pongthong, 1989). However, data on the etiology of these reported cases has been limited by the difficulty and cost of laboratory investigation. The present study was conducted in two university hospitals in Bangkok, where the incidence of infections by *Mycoplasma pneumoniae, Chlamydia trachomatis* and respiratory viral agents in pediatric pneumonia under 6 months of age was investigated during two different periods of time. IIF was used for the detection of viral antigens in exfoliated cells from nasopharyngeal aspirate (NPA), and cultivation methods were used for isolation of viruses and *C. trachomatis.* Serological methods were used for detection of antibodies to viruses and *M. pneumoniae.*

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MATERIALS AND METHODS

Study population

The subjects included 76 children aged under 6 months with clinical diagnosis of pneumonia. Forty-

five cases were patients who had attended Ramathibodi Hospital during the period between January and December 1987, and 31 cases were in-patients who had attended Siriraj Hospital during the period between June 1989 to May 1990. Criteria for diagnosis of pneumonia were based on presence of the following signs and symptoms: fever with tachypnea with or without wall retraction, fine to medium crepitation (rales) on auscultation, or evidence of pulmonary infiltration or consolidation on chest x-ray; cases with congestive heart failure were excluded. The study population comprised more than 95% of the total pneumonia cases attending both hospitals during the two study periods.

Specimen collection and processing

Clinical specimens collected from each patient included NPA and paired blood samples. Nasopharyngeal secretion was aspirated with a suction catheter (Davol, Cranston, RI, USA) into a mucous trap (Nunc, Roskilde, Denmark) and then divided into two tubes containing different kinds of transport media. An NPA sample for chlamydial isolation was transported in 2 ml of 2-sucrose phosphate solution supplemented with 200 µg/ml streptomycin, 20 µg/ml gentamicin, and 2 µg/ml fungizone, and that for viral isolation was transported in 3 ml of Hanks' balanced salt solution (Gibco, Grand Island, NY, USA) supplemented with 200 U/ml penicillin, 20 µg/ml gentamicin and 2 µg/ml fungizone. Specimens from the two hospitals were transported in an ice chest and processed at the Virology Laboratory, Siriraj Hospital. Travel time between the two hospitals was about 1 hour. Usually, the specimens were processed within 24 hours after collection. The NPAs in transport media were mixed throughly before centrifugation at 1500 rpm for 10 minutes in a refrigerated centrifuge. The supernatant fraction was used for chlamydial or viral isolation, and the sediment was washed further with PBS two to three times to get rid of mucus. Finally, the cell pellet was smeared on microscope slides, air-dried and fixed in precooled acetone for 10 minutes at 4°C. The slides were air-dried again and stained immediately using IIF technique or kept at -70°C. Blood samples were collected twice, 3-5 ml each time. The first blood sample was obtained during the acute phase and convalescent blood was obtained 2 to 4 weeks afterward. Sera were separated from clotted blood and stored at-20°C. Paired sera were tested simultaneously in each serological test. Paired blood could not be obtained from every case, because some patients did not revisit the hospital after recovery.

Indirect immunofluorescence test (Gardner and McQuillin, 1980; Puthavathana *et al,* 1990).

Briefly, the fixed cell deposits on microscope slides were stained with antisera to influenza A and B, RSV, parainfluenza virus types 1 and 3 and adenovirus. Fluorescein-isothiocyanate conjugated antibody to immunoglobulin was used as the second antibody. These reagents were purchased from Wellcome Diagnostics (Dartford, England). The slides were counterstained with Evan's blue (Sigma) and examined under a fluorescence microscope (Nikon, Tokyo). The slides of cell culture infected by each specific virus were used as positive controls.

Isolation of *C. trachomatis*

Protocols for isolation of *C. trachomatis* followed Bird and Forrester (1981). McCoy cells were culti vated on a glass cover-slip in a culture vial under MEM (Eagles) (Gibco, NY, USA) supplemented with 10% fetal calf serum (FCS) (Gibco, NY, USA), 20 µg/ml gentamicin, 200 µg/ml streptomycin and 2 µg/ml fungizone. The cell monolayers aged one day were infected and maintained for 48-72 hours in growth medium supplemented with 2 µg/ml cycloheximide and 5 mg/ml glucose. The infection was recognized by staining for cytoplasmic inclusion bodies with iodine solution, and/or by direct immunofluorescence staining with monoclonal antibody specific to *C. trachomatis* (Microtrak, Syva, Palo Alto, CAL, USA).

Isolation and identification of virus

Isolation of respiratory viruses employed four kinds of cell culture systems: HEp-2, LLC-MK2, MDCK and MRC-5. Additionally, the chick embryo inoculation technique was also performed for isolation of influenza virus. The procedures for cell cultivation, recognition and identification of viral infections were as described elsewhere (Puthavathana *et al,* 1990).

Serodiagnosis

Serodiagnosis of an agent was based on the demonstration of at least a fourfold rise in antibody

titer in paired sera. Serological tests performed in this study were particle agglutination (PA) and/or complement fixation (CF) for *M. pneumoniae,* CF test for RSV, parainfluenza types 1 and 3 and adenovirus, and hemagglutination-inhibition (HI) test for influenza A and B. Paired sera which showed at least a fourfold rise in antibody titer were considered to be positive.

Passive agglutination test

Reagent kits for PA, "Serodia-Myco II", were purchased from Fujirebio (Tokyo, Japan). Gelatin particles coated with cell membrane component of *M. pneumoniae* were used as test antigen and the unsensitized gelatin particles were used as the control.

Complement fixation test

CF antigens were purchased from the Behring Institute (Marburg, Germany). The CF method was modified after Hawker (1979).

Hemagglutination-inhibition test

Panels of HI antigens and specific antisera were contributed by the Centers for Disease Control (CDC), Atlanta. The panel of antigens used in 1987 included A/Taiwan/1/86 (H1N1), A/Mississippi/1/85 (H3N2) and B/Ann Arbor/1/86; those used during 1989 to 1990 were A/Taiwan/1/86 (H1N1), A/Shanghai/1/87 (H3N2), B/Victoria/2/87 and B/Yamagata/16/88. The test sera were treated for eradication of nonspecific inhibitor by receptor destroying enzyme (Sigma) and of nonspecific agglutinator by chick red cells. The HI protocol as described by CDC, Atlanta, was followed (Palmer *et al,* 1980).

Quality control of the methods

External and internal quality controls for the diagnosis of respiratory virus infection have been described in our previous paper (Puthavathana *et al,* 1990).

RESULTS

M. pneumoniae infection

Since the diagnosis of *M. pneumoniae* infection was based on serological tests of PA and CF, only cases with paired sera could be investigated; otherwise they were excluded from the data analysis. Thus, 31 out of 45 cases from Ramathibodi Hospital and 24 out of 31 cases from Siriraj Hospital were studied and it was shown that none of them developed *M. pneumoniae* infection.

C. trachomatis infection

Isolation of *C. trachomatis* was performed in 42 of 45 patients from Ramathibodi Hospital and 23 of 31 cases from Siriraj Hospital. Infection rates in the two hospitals were 16.7 and 21.7% of pneumonic patients, respectively. Males were more susceptible than females with the sex ratio between 2.6 and 2.7 : 1 (Table 1). The infected cases were clustered within the age-range of 0-3 months, except one case whose age was 5 months old.

Viral infections

Viral infection was diagnosed if any one of the three diagnostic methods, IIF, virus isolation or serology, gave a positive result. Nineteen (42.2%) of 45 cases from Ramathibodi Hospital, and 15 (48.4%) of 31 cases from Siriraj Hospital had contracted viral infections (Tables 2 and 3). Data from the two hospitals indicated that RSV was the most common virus found, followed by adenovirus, parainfluenza virus type 3 and other miscellaneous viruses. Totally, our investigation system could diagnose 51.1 and 61.3% of the study cases from both hospitals (Table 4).

Mixed infections

Mixed infections were observed in 4 (8.9%) of 45 patients from Ramathibodi Hospital. Three cases had mixed infections with two agents and one case was infected with three agents (Table 4). The combinations found in these mixed infections were RSV and influenza B virus, *C. trachomatis* and parainfluenza virus type 3, *C. trahomatis* and unidentified virus with positive hemadsorption test, and lastly *C. trachomatis* and RSV and parainfluenza virus type 3.

Two (6.5%) cases of mixed infection occurred among 31 patients from Siriraj Hospital. One case was infected with *C. trachomatis* and adenovirus, and another case was infected with parainfluenza virus type 3 and coxsackie B5 (Table 4).

Information from both hospitals indicated that *C. trachomatis* was the agent most frequently observed in mixed infections.

Table 1

Incidence of chlamydial pneumonia in children under 6 months of age.

Age-group	Sex	Ramathibodi Hospital		Siriraj Hospital	
		No. of patients	No. (%) of chlamydial infections	No. of patients	No. (%) of chlamydial infections
0-1		9	3 (33.3)	6	3 (50.0)
	M	6	2	3	2
	F	3	1	3	1
2-3		20	3 (15.0)	13	2 (15.4)
	M	15	3	8	2
	F	5	0	5	0
4-5		13	1 (7.7)	4	0 (0)
	M	8	1	3	0
	F	5	0	1	0
Total		42	7 (16.7)	23	5 (21.7)
	M	29	6 (20.7)	14	4 (28.6)
	F	13	1 (7.7)	9	1 (11.1)
M:F Ratio			2.7:1		2.6:1

Table 2

Incidence of viral pneumonia in children under 6 months of age: Ramathibodi Hospital (Jan-Dec 1987).

Age-group (months)	Sex	No.of patients	No.(%)of patients with virus	No. of viruses found	No.(%) of patients with indicated virus						
					FluA	FluB	RSV	PF1	PF3	Ad	Others
0-1		9	4 (44.4)	6	0	1	4	0	1	0	0
	M	6	3[a]	5	0	1	3	0	1	0	0
	F	3	1	1	0	0	1	0	0	0	0
2-3		21	9 (42.9)	9	0	0	4	0	2	2	1
	M	16	7[b]	7	0	0	2	0	2	2	1[c]
	F	5	2	2	0	0	2	0	0	0	0
4-5		15	6 (40.0)	6	1	0	3	0	0	2	0
	M	10	5	5	1	0	2	0	0	2	0
	F	5	1	1	0	0	1	0	0	0	0
Total		45	19 (42.2)	21	1 (2.2)	1(2.2)	11(24.4)	0(0)	3(6.7)	4(8.9)	1(2.2)
Subtotal	M	32	15(46.9)	17	1(3.1)	1(3.1)	7(21.9)	0(0)	3(9.4)	4(12.5)	1(3.1)
Subtotal	F	13	4(30.8)	4	0(0)	0(0)	4(30.8)	0(0)	0(0)	0(0)	0(0)

[a] One case was infected with RSV and influenza B, another case did with RSV and parainfluenza virus type 3 and *C. trachomatis*

[b] One case was infected with parainfluenza virus type 3 and *C. trachomatis*. another case did with unidentified virus with positive hemadsorption and *C. trachomatis*

[c] Unidentified virus with positive hemadsorption

Table 3

Incidence of viral pneumonia in children under 6 months of age: Siriraj Hospital (Jun 1989-May 1990).

Age-group (months)	Sex	No.of patients	No.(%)of patients with virus	No. of viruses found	No.(%) of patients with indicated virus						
					FluA	FluB	RSV	PF1	PF3	Ad	Others
0-1		6	3 (50.0)	3	0	0	1	0	0	2	0
	M	3	2[a]	2	0	0	0	0	0	2	0
	F	3	1	1	0	0	1	0	0	0	0
2-3		16	7(43.8)	8	0	0	4	0	1	2	1
	M	10	4[b]	5	0	0	3	0	1	0	1[c]
	F	6	3	3	0	0	1	0	0	2	0
4-5		9	5(55.6)	5	0	0	5	0	0	0	0
	M	6	3	3	0	0	3	0	0	0	0
	F	3	2	2	0	0	2	0	0	0	0
Total		31	15(48.4)	16	0(0)	0(0))	10(32.3)	0(0)	1(3.2)	4(12.9)	1(3.2)
Subtotal	M	19	9(49.0)	10	0(0)	0(0))	6(31.6)	0(0)	1(5.3)	2(10.5)	1(5.3)
Subtotal 0(0)	F	12	6(50.0)	6	0(0)	0(0)	4(33.3)	0(0)	0(0)	2(16.7)	0(0)

[a] One case was infected with adenovirus and *C. trachomatis*
[b] One case was infected with parainfluenza virus type 3 and Coxsackie B5
[c] Coxsackie B5

Table 4

Pathogens associated with pneumonia in children under 6 months of age.

Hospital	No. of *C. trachomatis* infected patients/ No.(%) of patients studied	No. of virus infected patients/ No.(%) of patients studied	No. (%) of mixed infections	Total patients diagnosed (%)
Ramathibodi	7/42 (16.7)	19/45 (42.2)	4/45[a] (8.9)	23/45 (51.1)
Siriraj	5/23 (21.7)	15/31 (48.4)	2/31[b] (6.5)	19/31 (61.3)

[a] One case was infected with two different viruses, two cases did with one kind of virus and *C. trachomatis*, and one did with two kinds of virus and *C. trachomatis*
[b] One case was infected with two different viruses, and one case did with one kind of virus and *C. trachomatis*

DISCUSSION

The present study was confined solely to mycoplasmal, chlamydial and viral infections which were associated with more than half of pneumonia cases under 6 months of age. It has been generally accepted that patients who attended Ramathibodi Hospital had higher socioeconomic status than those who attended Siriraj Hospital. Nevertheless, the incidence of infection by those agents in both hospitals were very similar, irrespective of differences in the study populations and study periods. *M. pneumoniae* infection was not detected in any study case from either hospital, eventhough PA, a more sensitive test, had been introduced in conjunction with CF in

the second study period. Our previous work conducted on 596 young children with acute lower respiratory infection (ALRI) during 1986-1987 also revealed no cases with *M. pneumoniae* infection (Suwanjutha *et al,* 1990). These results indicate that *M. pneumoniae* is not a common etiologic agent in pneumonia in young children. Our findings agree with some other reports (Forgie *et al,* 1992; Ray *et al,* (1993 a). However, this idea would be more strongly supported if no infection was found when a more sensitive test such as RNA-DNA hybridization technique for the detection of ribosomal RNA of *M. pneumoniae* was employed (Kleemola *et al,* 1993).

Prevalence of chlamydial cervical infection among pregnant women in the USA and Europe was estimated to be in the range of 2-37%, whereas 13% of the pregnant women in Thailand had positive cultures for *C. trachomatis* during the third trimester of gestation (Limudomporn *et al,* 1989). Infants born to infected mothers acquire chlamydial infection through contact with infected cervical secretions at the time of birth. Anatomical sites affected can be the eye, nasopharynx, rectum and vagina (WHO Working Group, 1986). Chlamydial pneumonia was first described in 1975, and later found to be one of the most common causes of pneumonitis in infancy (Schachter *et al,* 1975). The disease usually presents in infants between 3 and 11 weeks of age (Rettig, 1986). It was reported that during the first month of life 22% of 112 infants born vaginally to mothers infected with *C. trachomatis* had become culture-positive in the conjunctiva and 25% in the pharynx. Initially positive rectal and vaginal cultures were delayed until the third or fourth month of life (Bell *et al,* 1987). Similarly, in our study 33-50% of the infected infants were less than 2 months of age, and the incidence decreased to 15% in the age-group 2-3 months (Table 1). Of all *Chlamydia* infected cases, only one child was as old as 5 months. It is not known whether this child acquired the infection from the birth canal or postnatally. Another study in Thailand reported that *C. trachomatis* accounted for 26% of afebrile pneumonia in infants under 6 months of age (Limudomporn *et al,* 1989). It is noteworthy that none of our cases with chlamydial pneumonia was preceded by manifestation of conjunctivitis (unpublished data). Interestingly, we found that *C. trachomatis* was more prevalent in male than female infants (male-to-female sex ratio = 2.6:1), as it was in the Thai study cited above (ratio = 2:1) (Limudomporn *et al,* 1989).

Regarding viral infections in our study population, RSV was the most common agent found followed by adenovirus and parainfluenza virus (Tables 2, 3). We previously reported that RSV was predominant in the rainy season, parainfluenza virus was so at the beginning of the hot season, whereas adenovirus was scattered all year round (Suwanjutha *et al,* 1990). However, seasonality of these viruses could not be seen in this report according to small size of the study population. High incidence of RSV in ALRI has been found not only in developed but also in developing countries throughout the world, *eg* Thailand (Suwanjutha *et al,* 1990), the Philippines (Tupasi *et al,* 1990), Pakistan (Ghafoor *et al,* 1990), Argentina (Wissenbacher *et al,* 1990) and Uruguay (Hortal *et al,* 1990). In contrast, the most common viral agents found in ALRI in the Gambia were adenovirus and influenza A virus, while RSV was occasionally observed (Forgie *et al,* 1992).

Our study found 6.5 to 8.9% prevalence rates of mixed infections in which *C.trachomatis* was the most common agent. Codetection of two or three agents has been observed in 4.9-28% of the study population reported by many other workers (Hortal *et al,* 1990; Nohynek *et al,* 1991; Ray *et al,* 1993a,b). In most of these cases, illnesses associated with more than one agent were not significantly different from those involving only single agent, with respect to month of onset, illness type and age or duration of illness (Ray *et al,* 1993b). These general findings also hold true in our study.

In conclusion, the most common agent causing pneumonia in children under 6 months of age was RSV, followed by *C. trachomatis* and adenovirus. Our investigation system could not diagnose 38.7 and 48.9% respectively of the two populations studied (Table 4). Etiologic agents responsible in those cases might be bacterial agents such as *Streptococcus pneumoniae, Haemophilus influenzae, Staphylococcus aureus* or some other viral agents which could not be detected by our methods (Nohynek *et al,* 1991). In addition, young infants usually developed poor antibody responses, which have led to missed diagnosis by means of the serological approach, as has been shown previously (Puthavathana *et al,* 1990; Klee-mola *et al,* 1993).

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REFERENCES

Bell TA, Stamm WE, Kuo C-C, Wang S-P, Holmes KK, Grayston JT. Delayed appearance of *Chlamydia trachomatis* infections acquired at birth. *Pediatr Infect Dis J* 1987; 6 : 928-31.

Bird BR, Forrester FT. Laboratory diagnosis of *Chlamydia trachomatis* infections. Centers for Disease Control, Atlanta; US Department of Health and Human Services, 1981.

Forgie IM, Campbell H, Lloyd-Evans N, *et al.* Etiology of acute lower respiratory tract infections in children in a rural community in the Gambia. *Pediatr Infect Dis J* 1992; 11: 466-73.

Gardner PS, McQuillin J. Rapid virus diagnosis: Application of immunofluorescence. London: Butterworths, 1980.

Ghafoor A, Nomani NK, Ishag Z , *et al.* Diagnoses of acute lower respiratory tract infections in children in Rawalpindi and Islamabad, Pakistan. *Rev Infect Dis* 1990; 12:S907-14.

Hawker RA. General principles underlying laboratory diagnosis of viral infections. In: Lennette H, Schmidt NJ, eds. Diagnostic procedures for viral, rickettsial and chlamydial infections. 5th ed. Washington, DC: American Public Health Association, 1979; 1-48.

Hortal M, Mogdasy C, Russi JC, Deleon C, Suarez A. Microbial agents associated with pneumonia in children from Uruguay. *Rev Infect Dis* 1990; 12: S915-22.

Kleemola M, Heiskanen-Kosma T, Nohynek H, Jokinen S, Korppi M, Eskola J. Diagnostic efficacy of a *Mycoplasma pneumoniae* hybridization test in nasopharyngeal aspirate of children. *Pediatr Infect Dis J* 1993; 12:344-5.

Limudomporn S, Prapphal N, Nanthapisud P, Chomdej S. Afebrile pneumonia associated with chlamydial infection in infants less than 6 months of age: initial results of a three year prospective study. *Southeast Asian J Trop Med Public Health* 1989; 20:285-90.

Marks MI. Lower respiratory infections. In: Katz M, Stiehm ER, series eds. Pediatric infectious diseases for the practitioner. New York: Springer-Verlag, 1985; 292-395.

Nohynek H, Eskola S, Laine E, *et al.* The causes of hospital treated acute lower respiratory tract infection in children. *Am J Dis Child* 1991; 145; 618-22.

Palmer DF, Coleman MT, Dowdle WR, Schild GC. Advanced laboratory techniques for influenza diagnosis. Centers for Disease Control, Atlanta; US Department of Health and Human Services, 1980.

Pongthong B. Present status of ARIC in Thailand. Proceedings of the 2nd National Seminar on Prevention and Conrol of Acute Respiratory Infections in Children. Department of Communicable Diseases, Ministry of Public Health, Bangkok, Thailand. July 3-15, 1989; 141-9 (Thai).

Puthavathana P, Wasi C, Kositanont U, *et al.* A hospital-based study of acute viral infections of the respiratory tract in Thai children, with emphasis on laboratory diagnosis. *Rev Infect Dis* 1990; 12 : S988-94.

Ray CG, Holberg CJ, Minnich LL, *et al.* Acute lower respiratory illnesses during the first three years of life: potential roles for various etiologic agents. *Pediatr Infect Dis J* 1993a; 12 : 10-4.

Ray CG, Minnich LL, Holberg CJ, *et al.* Respiratory syncytial virus associated lower respiratory illnesses: possible influence of other agents. *Pediatr Infect Dis J* 1993b; 12 : 15-9.

Rettig PJ. Infections due to *Chlamydia trachomatis* from infancy to adolescence. *Pediatr Infect Dis* 1986; 5 : 449-57.

Schachter J, Lum L, Gooding CA, *et al.* Pneumonitis following inclusion blennorrhea. *J Pediatr* 1975; 87 : 779-80.

Suwanjutha S, Chantarojanasiri T, Watthana-kasetr S, *et al.* A study of nonbacterial agents of acute lower respiratory tract infection in Thai children. *Rev Infect Dis* 1990; 12 : S923-8.

Tupasi TE, Lucero MG, Magdangal DM, *et al.* Etiology of acute lower respiratory tract infection in children from Alabang, Metro Manila. *Rev Infect Dis* 1990; 12 : S929-39.

WHO Scientific Group. Viral respiratory diseases. *WHO Tech Rep Ser* 1980; 642.

WHO Working Group. Extra-ocular chlamydial infection. *Bull WHO* 1986; 66 : 481-92.

Wissenbacher M, Carballal G, Avila M, *et al.* Etiologic and clinical evaluation of acute lower respiratory tract infections in young children. *Rev Infect Dis* 1990; 12 : S889-98.