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Infections with *Legionella pneumophila* in Children

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To learn the role of *Legionella pneumophila*, the agent of Legionnaires' disease, in childhood illness, a prospective study was conducted among 52 children younger than four years of age with acute disease of the lower respiratory tract. Viral, mycoplasmal, and bacterial cultures and acute- and convalescent-phase sera were obtained during 64 episodes of acute illness; additional sera were drawn annually for three to five years. On the basis of serologic evidence, none of the acute episodes appeared to be due to *L. pneumophila* serogroup 1 or 2. However, examination of annual serum specimens showed that 27 (52%) of the children had rises in titer of indirect immunofluorescent antibody (a fourfold or greater rise to a reciprocal titer of ≥128). Most rises in titer were in response to the serogroup 2 antigen. These results suggest that *L. pneumophila* is not a common cause of acute respiratory disease in early childhood in the study area but that children are frequently exposed to the organism. Alternatively, the serologic responses might be to unrelated cross-reacting microorganisms.

Much has been learned about the bacterium *Legionella pneumophila* since it was discovered to be the cause of an outbreak of pneumonia at a hotel in Philadelphia, Pa., in 1976 [1, 2]. The organism is now known to be responsible for about 1% of the cases of sporadic pneumonia in adults [3] and for opportunistic infections in compromised hosts [4]. However, very little is known about the importance of this agent in respiratory disease in children. Because most microorganisms that commonly cause pulmonary infection in adults cause similar illnesses in children, it seemed likely that *L. pneumophila* could be a significant pathogen for children. Therefore, we studied the role of *L. pneumophila* in acute disease of the lower respiratory tract and its seroepidemiology in children.

Patients and Methods

One hundred fifty children were enrolled between November 1972 and November 1974 in a prospective longitudinal study of etiologic agents in acute infections of the lower respiratory tract. The children were from two private pediatric practices in Denver, Colo. Signed, informed consent was obtained from the parents. The majority of children were entered into the study during their first known episode of pneumonia, bronchiolitis, asthmatic bronchitis, or tracheobronchitis. Chest roentgenograms were obtained in 63 of 64 illnesses, and infiltrates were noted in 29 (46%). Serum was drawn from the children during each acute respiratory illness, four to six weeks later, and approximately annually until their seventh birthday. Fifty-two of the children had one or more sets of acute- and convalescent-phase sera drawn and at least three annual follow-up specimens; data from those 52 children are the basis of this report.

Throat swabs and nasal washes obtained during acute illnesses of the lower respiratory tract were cultured for viruses in human diploid fibroblast, Hep 2, and rhesus monkey kidney tissue cultures and for *Bordetella pertussis* and *Mycoplasma pneumoniae* using conventional culture methods [5, 6]. Acute- and convalescent-phase sera were stored at −80 C and tested simultaneously. Antibodies to respiratory syncytial virus, influenza A and B virus, adenovirus, coronaviruses OC43 and 229E, and *M. pneumoniae* were measured by CF, and antibodies to parainfluenza vi-

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rus types 1, 2, and 3 were measured by HAI. Antibodies to strains of *L. pneumophila* serogroup 1 (Philadelphia 1) and serogroup 2 (Togus 1) were measured in duplicate using heat-killed organisms as antigens by the indirect immunofluorescence technique [7].

Results

The study group consisted of 33 boys and 19 girls. Upon entry into the study, they ranged in age from one month to 46 months (mean, 14 months); 39 (75%) were younger than two years old, and nearly half of the children in the study were enrolled during their first year of life. The average duration of follow-up was 3.8 years. A total of 64 pairs of sera from children with acute illnesses and 191 annual serum specimens, unassociated with acute respiratory illness, was examined. Fourteen (27%) of the 52 patients were initially diagnosed or subsequently found to be asthmatic.

The 64 acute respiratory illnesses were diagnosed clinically by the children's pediatricians as bronchiolitis (32.8%), asthmatic bronchitis (26.6%), pneumonia (25.0%), and tracheobronchitis (15.6%) and were due principally to viral infections (table 1). The high frequency of infections with respiratory syncytial virus reflects the importance of this agent in infancy. Viral or mycoplasmal infections were identified in 48 (75%) of the illnesses; no agent was identified in the remaining 16 (25%). No child had a fourfold or greater rise in titer of antibody to *L. pneumophila* of either serogroup in association with acute illness.

In contrast, when sera taken during annual follow-up visits were examined for evidence of infection with *L. pneumophila*, 27 (52%) of the 52 children had a fourfold or greater rise in reciprocal antibody titer to ⩾128 (table 2). Seventeen (63%) of these 27 children had greater rises in titer of antibody to the serogroup 2 antigen than to the serogroup 1 antigen. Rises in titer were often followed by a sustained elevation in titer (⩾128) in subsequent annual serum specimens. Some cross-reactivity was apparent between serogroups 1 and 2, and in these cases, the greater rise was considered diagnostic.

Antibody titers, by age of the child, are shown in table 3. Titers of ⩾64 to serogroup 1 were found in 7%–29% of the specimens; to serogroup 2, in 11%–64%. The titers tended to be higher after the first year of life.

Discussion

The recognition of the importance of *L. pneumophila* in both sporadic and epidemic pneumonia in adults inevitably leads to questions about its pathogenic potential during childhood. At present there is little information about this infection in children. Investigation of the 1976 epidemic in Philadelphia revealed a single case diagnosed sero-

Table 1. Agents identified in 64 acute infections of the lower respiratory tract in 52 children.

Agent	Positive culture and serology	Positive culture only	Positive serology only	Total (%)
Respiratory syncytial virus	17	4	0	21 (32.8)
Influenza A or B virus	2	1	5	8 (12.5)
Parainfluenza virus types 1, 2, or 3	3	3	0	6 (9.4)
Adenovirus	0	2	1	3 (4.7)
Mycoplasma pneumoniae	0	1	2	3 (4.7)
Coronavirus	0	0	1	1 (1.6)
Herpes simplex virus	0	1	0	1 (1.6)
Mixed infections*	5	0	0	5 (7.8)
Legionella pneumophila	0	0	0	0
No agent identified	0	0	0	16 (25.0)
Total	...	...	...	64 (100)

* Mixed infections were identified by culture or seroconversion as follows (one child each): influenza A virus (seroconversion) and parainfluenza virus type 3 (culture and seroconversion); respiratory syncytial virus (culture and seroconversion) and adenovirus (seroconversion); respiratory syncytial virus (culture and seroconversion) and influenza A virus (seroconversion); respiratory syncytial virus (culture and seroconversion) and herpes simplex virus (culture); respiratory syncytial virus (culture) and parainfluenza virus type 3 (seroconversion).

Table 2. Seroconversions to two serogroups of *Legionella pneumophila* in 52 children followed annually for three or more years.

Serogroup*	No. of children (%)
Serogroup 1 (Philadelphia 1)	2 (3.8)
Serogroup 2 (Togus 1)†	17 (32.7)
Both serogroups simultaneously‡	5 (9.6)
Both serogroups at different times	3 (5.8)
No titer rise	25 (48.1)
Total	52 (100)

* If antibody titers to both serogroups rose simultaneously, the greater rise was considered diagnostic.

† Two children had a fourfold or greater rise in antibody titer to serogroup 2 at different ages.

‡ Simultaneous and equivalent rises in antibody titer to both serogroups.

logically in a three-year-old child [1]. More recently it was reported that a 2.5-year-old boy with acute lymphocytic leukemia and a three-year-old boy with trisomy 21 had developed severe pneumonitis associated with a striking rise in titer of antibody to *L. pneumophila* [8, 9]. The latter case was confirmed by direct immunofluorescence of lung biopsy tissue.

Our results suggest that *L. pneumophila* is not a common cause of acute symptomatic infection of the lower respiratory tract in young children in the geographic area of our study. However, the high frequency of seroconversions seen with annual follow-up testing suggests that this organism is commonly encountered in the early years of life. Infection with *L. pneumophila* may resemble infection with *M. pneumoniae* in that preschool children frequently experience subclinical infection with seroconversion, but few experience significant clinical illness until later in life [10]. Alternatively, it may be that infection with *L. pneumophila* in children is more likely to cause an illness not characterized by symptoms in the lower respiratory tract, perhaps like the undifferentiated febrile illness Pontiac fever [11]. If the latter possibility was true, we would not have collected acute- and convalescent-phase sera at the appropriate times to detect acute infections.

The interpretation of any seroepidemiologic study depends on the specificity of the serologic test used. Wilkinson et al. demonstrated that the indirect fluorescent antibody test for *L. pneumophila* measures not only serogroup-specific and species-specific antibody but also antibody to antigens shared by other gram-negative bacteria [7, 12]. Cross-reactions also have been reported with *Yersinia pestis, Francisella tularensis, Leptospira interrogans, Bacteroides fragilis, Chlamydia psittaci, M. pneumoniae,* and several newly described organisms associated with pulmonary disease [13–18]. These cross-reactions probably do not greatly diminish the value of the indirect fluorescent antibody test in diagnosing Legionnaires' disease in adults with pneumonia, but they may be important in analyzing the results of seroepidemiologic studies. We recognize the possibility that some of the rising titers of antibody to *L. pneumo-*

Table 3. Distribution as measured by indirect immunofluorescence of titers of serum antibody to *Legionella pneumophila* serogroups 1 and 2, by age group, in 52 children followed annually for three or more years.

	No. of children and cumulative percentage with titers by age in years						
Reciprocal titer	<1 (n = 26)	1 (n = 39)	2 (n = 45)	3 (n = 51)	4 (n = 46)	5 (n = 25)	6 (n = 11)
Serogroup 1							
≥1,024	0	0	1 (2.2)	0	0	0	0
512	0	0	1 (4.4)	2 (3.9)	1 (2.2)	0	0
256	0	1 (2.6)	2 (8.9)	2 (7.8)	0	0	0
128	1 (3.8)	0	2 (13.3)	3 (13.7)	4 (10.9)	2 (8.0)	2 (18.2)
64	1 (7.7)	4 (12.8)	3 (20.0)	8 (29.4)	6 (23.9)	3 (20.0)	1 (27.3)
<64	24 (100.0)	34 (100.0)	36 (100.0)	36 (100.0)	35 (100.0)	20 (100.0)	8 (100.0)
Serogroup 2							
≥1,024	0	0	1 (2.2)	1 (2.0)	1 (2.2)	0	0
512	0	1 (2.6)	1 (4.4)	1 (3.9)	1 (4.3)	0	0
256	0	3 (10.3)	3 (11.1)	2 (7.8)	2 (8.7)	1 (4.0)	3 (27.3)
128	2 (7.7)	3 (17.9)	7 (26.7)	13 (13.3)	8 (26.1)	6 (28.0)	0
64	1 (11.5)	6 (33.3)	4 (41.0)	9 (51.0)	11 (50.0)	6 (52.0)	4 (63.6)
<64	23 (100.0)	26 (100.0)	29 (100.0)	25 (100.0)	23 (100.0)	12 (100.0)	4 (100.0)

phila that we observed in young children may have resulted from infection with organisms with cross-reacting antigens, but we could not measure the importance of these cross-reactions because we did not perform surveillance cultures for such organisms.

A study by Storch et al. of the prevalence of antibody to *L. pneumophila* in four major American cities showed that only 1.7% of adults ≥46 years of age possessed reciprocal antibody titers of ≥64 to the serogroup 1 antigen [19]. The authors concluded that an indirect fluorescent antibody titer of ≥64 would have a specificity of 98.3% if a single titer was used as a diagnostic test in patients with an acute illness resembling Legionnaires' disease. A serologic study by Ryan et al. of 55 children with leukemia or other malignancies and pneumonia found only the aforementioned 2.5-year-old child with detectable antibody; the remaining children all had antibody titers of <64 to serogroup 1 antigen [8]. These two studies indicate that titers of antibody to *L. pneumophila* of ≥64 are unusual in adults and children in the United States. However, our data suggest that these observations in adults and immunocompromised children cannot be safely extrapolated to healthy children in the geographic area of our study. Seven (13%) of 52 children with acute disease of the lower respiratory tract in our study had antibody titers of ≥64 to *L. pneumophila* serogroup 1 in convalescent-phase sera without diagnostic rises in titer. Foy et al. [3] also found elevated titers in children; 25% of their children younger than five years with pneumonia had antibody titers of ≥128 to *L. pneumophila* serogroup 1. Similarly, none of their patients had a fourfold or greater rise in antibody titer [3].

In conclusion, if we assume that the indirect fluorescent antibody test for *L. pneumophila* is specific, our serologic data show that this organism was commonly encountered by the children enrolled in our study. It is likely that the majority of exposures resulted in either subclinical infection or atypical illness. The definitive diagnosis of Legionnaires' disease is possible with use of culture or direct immunofluorescence examination of lung tissue. Greater effort to identify *L. pneumophila* in children by these methods and additional seroepidemiologic studies will help establish its true importance as a pathogen in childhood.

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