Mycoplasma pneumoniae – A Tale of 50 Years

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The 40-paged February issue (1966) of *Indian Pediatrics* comprised of four original articles, case records and a review of current literature. Among these, we decided to review the article entitled "*Mycoplasma pneumoniae* infection in an Indian child." At that time, *M. pneumoniae* had been recently discovered, and it had generated a lot of interest as a possible cause of pneumonia. Much has subsequently been learned about this organism, and in this article we review the gain in insight about this organism in last 50 years.

THE PAST

The reviewed paper [1] was a combined effort from John Hopkins University Center for Medical Research and Training and Institute of Child Health, Calcutta. It describes the clinical course of an Indian girl with pneumonitis which was subsequently diagnosed to be due to *M. pneumoniae*. It was only 4 years prior to this publication that *M. pneumoniae* was successfully cultivated in cell-free medium [2]. The investigations of this child were supported by United States Public health service research grant from National Institute of Health. The reported case was a 23-month old Bengali girl, eleventh in birth order! She presented with a seven day history of fever, coryza and progressive cough that was severe at night with a few instances of post-tussive vomiting. There was a past history of repeated upper respiratory tract infections. She was immunized only against small pox. Examination revealed fever, tachycardia and tachypnea. The child had cervical and axillary lymphadenopathy, hyperemia of nasal mucosa and tonsils, and an injected tympanic membrane. The chest was hyperinflated; the right lower chest was dull on percussion with diminished breath sound. Occasional inspiratory and expiratory rales were heard over the affected area. Cardiac examination was normal; the liver was palpable 5 cm and spleen 2 cm below costal margin. The chest *X*-ray demonstrated a confluent opacification of the right middle lobe consistent with alveolar pneumonitis and peribronchitis in the right upper and lower lobes. Blood counts showed a neutrophilic leukocytosis. Tuberculin skin test was negative and sweat chloride was normal.

The bacteriological study of the nasopharyngeal, throat, per nasal tracheal aspirate, and percutaneous lung aspirate was non-contributory, and virological examinations failed to yield a viral agent. Meanwhile the child received treatment with penicillin and streptomycin for 8 days. She became afebrile on 7th day of hospitalization, and her respiratory findings, adenopathy and splenomegaly disappeared while the liver receded to 2 cm below costal margin. The throat swab specimen after being stored at –20 ºC to –70 ºC for 27 days was examined for *M. pneumoniae* by inoculating on agar plates and diphasic medium, and its growth was confirmed on the 7th day of first agar subculture. On day 14 of incubation in diphasic medium, a drop in pH (glucose fermentation) and spherular colonies were observed floating in the media. The colonies demonstrated guinea pig erythrocyte hemolysis (within 48 hours) and sheep cell haemadsorption, properties peculiar to *M. pneumoniae*. Isolation of *M. pneumoniae* from throat and tracheal aspirates of this child with pneumonia – whose extensive bacterial and viral investigations were non-contributory – indicated etiological role of *M. pneumoniae*. The child was treated with demethyl-chlortetracycline. A repeat *X*-ray 3 month later showed persisting middle lobe infiltrates, although child was asymptomatic.

Historical background and past knowledge: In the late nineteenth century, Nocard and Roux isolated the first mycoplasma organism from cattle suffering from pleuropneumonia [3]. This was followed by the discovery of other mycoplasma from various animal species, which were termed as pleuropneumonia like organisms (PPLO). The isolation of first human mycoplasma (Mycoplasma hominis) is credited to Dienes and Edsall [4] in 1937 from a Bartholin gland abscess. In 1944, Eaton and colleagues

discovered an organism termed Eaton agent, as the cause of pneumonia [5]. This agent was considered a virus till 1961, when Marmion and Goodburn [6] described it to be morphologically similar to PPLO. Eventually in 1962, Chanock, et al. [2] described the Eaton agent to be a mycoplasma after its successful cultivation in cell-free media. Shortly thereafter, he proposed the name *Mycoplasma pneumoniae* as the organism inoculated in human volunteers produced pneumonia. Kingston, *et al.* [7] established the therapeutic efficacy of demethyl-tetracycline for *M. pneumoniae* pneumonia.

The organism was peculiarly known for causing cold agglutinin positive primary atypical pneumonia in military recruits and close population subgroups [8], but it was relatively uncommon in early childhood. The isolation of this organism was slow and difficult, serological tests were being developed and indistinctive clinical picture did not itself lead to the diagnosis.

THE PRESENT

A lot has been learnt about its biological properties since isolation of the organism in 1962. *M. pneumoniae* are now known to be the smallest free-living ubiquitous organisms. The pathogenicity in humans is attributed to the phenomena of cyto-adhesion at the mucosa of the respiratory epithelium, impaired muco-ciliary clearance and elaboration of a cytotoxin called as CARDS toxin that damages the respiratory ciliated epithelial cells. In addition, infection with *M. pneumoniae* induces production of pro-inflammatory mediators and cytokines, which further exacerbate the disease process. The ability of mycoplasma to internalize and replicate in alveolar macrophages can lead to chronicity of infection and resistance to antibiotics [9]. Recurrences are known to occur.

In contrast to the historic belief of occurrence of *M. pneumoniae* infection among closed populations groups like military bases and boarding schools, the infection and disease occurs throughout the year punctuated by cyclical epidemics which are specific to geographic locations. Pneumonia is the most important manifestation seen in children aged between 5 and 15 years, while wheezing occurs more commonly in under-five children. It is speculated to cause asthma exacerbations [10], and can lead to decreased pulmonary clearance and airway hyper-responsiveness even after the control of infection.

Even today, it is practically very difficult to diagnose *M. pneumoniae* infection. Culture techniques require specialized expertise, and are used only for research purposes for geno-typing and anti-microbial susceptibility testing. Serological tests require paired samples (acute and convalescent 2-3 weeks apart) for definitive diagnosis. The detection of mycoplasma using cold agglutination test is non-specific with limited diagnostic utility. The complement fixation test, which was considered standard diagnostic tool till 2 decades ago, has been replaced by rapid, single-specimen, specific antibody detection (IgM or IgA) against mycoplasma antigens using membrane-based enzyme-linked immunosorbent assays or immuno-flourescence. Specific IgM and IgA responses need to be combined with direct pathogen detection for establishing causation. Polymerase chain reaction (PCR) using numerous gene sequences of *M. pneumoniae* as primers is a highly sensitive and specific test to detect *M. pneumoniae* in various specimens like respiratory secretions, blood, CSF and urine, but lacks validation and standardization. In this setting, it is commendable that authors of this case record could make a diagnosis of *M. pneumonia* infection 50 years ago!

For treatment of *M. pneumoniae* infection, macrolides (azithromycin and clarithromycin) are now prefered in children. The children and adult contacts are advised to follow personal protective measures during period of infectivity till the index case continues to cough. The trials for the development of vaccines are underway, but still have a long way to go.

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