Thesis Title Respiratory Syncytial Virus Infection in Children and Its Antibody Response

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ABSTRACT

Acute lower respiratory tract infection (ALRI) in children is the most common cause of illness in children under 5 years of age throughout the world. Generally, acute respiratory infections in small children are caused by viruses, and bacteria, while fungi and parasites play role only under special circumstances. Previous reports have shown that respiratory syncytial virus (RSV) is the major cause of ALRI, followed by parainfluenza virus, and adenovirus, respectively. Laboratory investigation for diagnosis of respiratory viral infection can be achieved by several methods such as detection of viral antigens in infected cells from nasopharyngeal aspirate by indirect immunofluorescence (IF), isolation of respiratory viruses in cell culture and detection of specific antibodies in sera.

Diagnosis of respiratory viral infection by detection of specific antibody in serum was generally based on a four-fold rise in titer of specific antibody in paired sera. Complement fixation test (CF) mostly showed negative results, although the virus had been isolated or the viral antigen had been detected in those cases. The negative CF test is explained in such a way that small children have immature immunity and fail to produce antibody response. In addition, CF test itself is not sensitive enough to pick up small amount of specific antibody.

Nowadays, reagents for detection specific antibody by Enzyme-linked immunosorbent assay (ELISA) are commercially available. This method is highly sensitive and can specify class of immunoglubulin, e.g., IgG and IgM. According to high sensitivity of ELISA, specific IgM can be detected for several months after the infection. Thus, this may lead to misdiagnosis of the subsequent infections.

If ELISA is used to investigate for a four-fold rise in titer of any class of immunogloblin in paired sera, it would be possible to clarify that infants and small children can develop immune response to viral infection or not. For its importance, RSV was chosen as the model of our study.

Laboratory methods used to diagnose the viral infections in this study including immunofluorescence test (IF) for specific antigens of adenovirus, influenza virus types A and B, parainfluenza virus types 1 and 3, and RSV; virus isolation in cell cultures of HEp-2, MDCK and LLC-MK2; and serodiagnosis by complement fixation test (CF) for adenovirus, parainfluenza virus types 1 and 3, and RSV, hemagglutination inhibition test (HI) for influenza virus types A and B, and ELISA test for RSV.

Of 261 patients who were admitted to the Pediatric Ward, Siriraj Hospital from June 1990 to December 1991, males were more prevalent with the male to female sex ratio of 1.5:1. Incidence of ALRI was highest in patients of age-group 6-11 months old (37.9%). However, 91.2% (238 cases) were under 2 years of age. The most common clinical diagnosis was pneumonia (67.4%), followed by bronchiolitis (15.3%), croup (13.8%) and bronchitis (3.4%), respectively. RSV played major role in pneumonia, bronchiolitis and bronchitis. Parainfluenza virus type 3 played major role in croup.

Seasonal distribution of RSV was demonstrated in rainy season, and its peaks were seen in August or September. Parainfluenza virus type 3 had its peak in February. For other viruses, seasonal distribution could not be observed because number of the infected cases was too low. During the epidemic season, RSV subgroup B predominated in 1990, whereas RSV subgroup A predominated in 1991.

Of the 261 study patients, respiratory virus infecions were diagnosed in 49% (128 cases), and 135 respiratory virus agents were found. RSV was the most common virus observed (34.9%, 91 cases), followed by parainfluenza virus type 3 (8.8%, 23 cases), adenovirus (3.4%, 9 cases), influenza A (1.9%, 5 cases), influenza B (1.5%, 4 cases) and parainfluenza virus type 1 (1.1%, 3 cases). Mixed infections were found in 2.3% (6 cases).

Among a total of 261 cases, only 125 cases could be investigated completely i.e., there were adequte cells in nasopharyngeal aspirate and paired sera were obtained. It was demonstrated that serological methods was the most efficient diagnostic method (56.0%, 70 cases), followed by indirect IF (35.2%,

(35.2%, 44 cases) and isolation of virus in cell culture (24.8%, 31 cases). However, sensitivity of each method for each virus was different. The method of virus isolation in cell culture was the least sensitive for RSV, because it could diagnose only 17 of all 63 RSV infected cases. In contrast, the isolation method was the most sensitive test for parainfluenza virus type 3, since it could diagnose 7 of 10 cases of parainfluenza virus type 3 infection. Similarly, CF can diagnose RSV infection only in 11 of 63 cases, while it did so in 6 of 10 parainfluenza virus type 3 infected cases.

Our results indicated that if complete clinical specimens were obtained for laboratory investigation, viral infection will be found in 59.2% (74 of 125 subjects), while investigation on incomplete clinical specimens could diagnose viral infection in 49.0% (128 of 261 cases) of ALRI.

Among a total of 125 study cases with complete clinical specimens, RSV infection was diagnosed in 50.4% (63 cases) of whom investigation by ELISA and CF could diagnose 44.8% (56 cases). Seroconversion by ELISA was observed in all 56 cases while only 11 cases of them showed seroconversion by CF test.

Among 56 RSV infected cases who developed seroconversion, 31 cases (55.4%) produced one class of specific immunoglobulin, 23 cases (41.1%) produced two classes, and only 2 cases (3.6%) produced three classes. The most common immunoglobulin class detected was IgG (76.8%, 33 of 56 cases), followed by IgA (44.6%, 25 of 56 cases) and IgM (26.8% 15 of 56 cases), respectively. It also demonstrated that major CF activity in RSV infection was present in IgG, not in IgA and IgM classes.

The present study showed that most infants and small children could develop seroconversion to RSV infection. Nevertheless, their immune response was incomplete such that only some classes of immunoglobulin were produced. Among 7 RSV infected cases who could not develop seroconversion, 6 of them had pre-existing antibody in the acute sera. It was not known whether this antibody was derived from maternal origin or from previous infection. Nevertheless, pre-existing antibody may elicit suppression effect on development of seroconversion.

An attempt to determine the O.D. value at single serum dilution as the cut-off point for RSV infection has been made. Unfortunately, there were a large overlapping in the O.D. values from different study groups. And the O.D. values diagnostic of RSV infection could not be established.