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Research Note

RESPIRATORY SYNCYTIAL VIRUS: OCCURRENCE OF SUBGROUPS A AND B STRAINS IN RIO DE JANEIRO

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Respiratory syncytial virus (RSV) is the most common cause of severe lower respiratory infections among infants and children, specially those between 2 and 6 months of age (R. B. Belshe et al., 1984, RSV, p. 361-384. In R. B. Belshe (ed.) *Textbook of Human Virology*. Littleton, PSG Publishing Co.). In Rio de Janeiro, RSV infections occur annually in the fall and early winter and are widespread among infants and children in the community (J. P. Nascimento et al., 1981, *Bol. Epidemiol., 13:* 131-132; 1982, *14:* 175-176; 1983, *15:* 105-107; 1984, *16:* 137-139). Until recently, RSV has been considered to be of a single broad serotype with minor antigenic variations, although cross-neutralization tests with animal hyperimmune sera showed a certain degree of difference between isolates of the virus (H. V. Coates et al., 1963, *Proc. Soc. Exper. Biol. Med., 112:* 958-964). More recently, studies with monoclonal antibodies used against specific RSV proteins revealed that RSV isolates can be separated into two major subgroups, designated either as subgroups A and B (M. A. Mufson et al., 1985, *J. Gen. Virol., 66:* 2111-2124) or 1 and 2 (L. J. Anderson et al., *J. Infect. Dis., 151:* 626-633). Investigators from different countries have reported that the two subgroups of RSV co-circulated within the same community (R. M. Hendry et al., 1986, *J. Infect. Dis., 153:* 291-297; H. Tsutsumi et al., 1988, *J. Clin. Microbiol., 26:* 1171-1174). To verify the occurrence of RSV subgroups A and B strains in community outbreaks, we employed immunofluorescence staining directly in cell smears from nasopharyngeal secretions (NPS) using monoclonal antibodies (MAbs) specific for RSV subgroups A and B.

Nasopharyngeal secretions had been collected since 1981 from children under 5 years of age and processed for isolation in tissue culture and rapid diagnosis by immunofluorescence (IFAT), as previously described (M. M. Siqueira et al., 1986, *Mem. Inst. Oswaldo Cruz, 81:* 225-232; 1988, *Rev. Bras. Pat. Clin., 24:* 60-61). Fixed slide preparations of cells from the nasopharyngeal secretions were routinely stored at $-56\,^{o}C$ in dry ice. These slides were subgrouped by indirect immunofluorescence staining with monoclonal antibodies specific for subgroup A (92-11c) and subgroup B (102-10b). Both monoclonal antibodies recognize epitopes on the fusion protein of the virus (L. J. Anderson et al., 1985, *J. Infect. Dis., 151:* 626-633; 1986, *J. Clin. Microbiol., 23:* 475-480). The MAbs and conjugated were previously standardized using cell cultures and NPS slides RSV positives, and used at a dilution described as following. Monoclonal antibodies were used at a dilution of 1/10. Fluorescein isothiocyanate-conjugated goat anti-mouse immunoglobulin IgG (Cappel) was employed at a dilution of 1/30. In order to improve the results, replicate slides were stained with the same MAbs labelled with biotin and used at a dilution of 1/10. Fluorescein-Streptavidin (Amersham International, UK) was used at a dilution of 1/40.

During the years 1985, 1987 and 1988, 18 RSV positive nasopharyngeal secretions were available and analyzed. Both subgroups A and B were co-circulating during all the years studied. Of the 13 specimens analyzed in 1988, 11 reacted with MAb specific for subgroup A and 2 with MAb specific for subgroup B. In 1987, 2 specimens reacted with MAb specific for subgroup B and 1 specimen reacted with MAb specific for subgroup A. In 1985, 1 specimen reacted with MAb 92-11c and 1 with MAb 102-10b.

To subgroup RSV we could use either fixed NPS cells or RSV isolates in cell cultures. The advantage of using the former is that it precludes the possibility of the host cells selecting a

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diversity of virus variation subpopulations from a single clinical specimen during isolation and subsequent cultivation procedures, as it is reported to occur with influenza viruses (R. Pyhälä et al., 1988, *Epidem. Inf., 100:* 511-522).

Our results, as well as previously published reports (C. E. Taylor et al., 1989, *Lancet, 1:* 777-778; J. C. Russi et al., 1989, *J. Clin. Microbiol., 27:* 1464-1466) have shown that group designation by IFAT in NPS cells is possible. Another advantage of grouping RSV directly in NPS cells by IFAT is that it overcomes differences in the recovery efficiency of RSV strains and constitutes a usefull tool for laboratories with limited resources for the handling of cell cultures and virus isolation.

Using RSV isolated in cell cultures, B. Akerlind & E. Norrby (1986, *J. Med. Virol., 19:* 241-247) reported that subgroups A and B strains occurred together in Stockholm, Sweden. R. M. Henry and co-workers (1989, *J. Infect. Dis., 160:* 185-190) reported that the prevalence of subgroups A and B varied both between and within yearly outbreaks. Among the 5 epidemics of RSV that M. A. Mufson et al. (1988, *J. Infect. Dis., 157:* 143-148) studied, subgroups A and B strains occurred together throughtout the duration of each epidemic, although subgroup A strains occurred, on the average, three times as often as subgroup B strains. Reasons for such differences in the epidemiology of RSV have not been elucidated.

Our findings underscore the importance of conducting further epidemiological studies as to the pattern of subgroups A and B strains to determine the relative epidemiological importance of the two subgroups in our community. Studies using an expanded panel of RSV strain specific monoclonal antibodies with RSV isolates are in progress.

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