Short Communication

Seroprevalence of Coxsackievirus B3 in Yantai, China

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SUMMARY: Coxsackievirus B3 (CVB3) is an important human pathogen, which is frequently associated with aseptic meningitis and encephalitis in many parts of the world. However, the seroprevalence of this viral infection in China is not well understood; therefore, in the present study, we conducted a serological survey in which 373 serum samples obtained from healthy people belonging to 10 age groups in Yantai city, China were examined. The results revealed that CVB3 is widely distributed in the population with a seroprevalence of 52.3%. The seroprevalence of CVB3 was lowest (3.7%) in the group aged 8–12 months and >50% in all groups aged >15 years. A significant increase (P < 0.001) was observed among the successive four groups aged 1, 2–4, 5–7, and 8–10 years. Variation of geometric mean titer related to age showed similar tendencies. Our results indicated that CVB3 infections occurred mostly in preschool and early-elementary school settings, and children <5 years of age are the most susceptible populations due to their low CVB3 antibody levels.

Coxsackievirus (CV) B3 is a member of the genus Enterovirus, family Picornaviridae, and order Picornavirales. It is commonly associated with myocarditis, aseptic meningitis, neonatal systemic illness, meningoencephalitis in immunodeficient persons, herpangina, and hand, foot, and mouth disease (HFMD) (1–4). CVB3-associated aseptic meningitis is an emerging concern in mainland China (5,6). However, to date, there is no specialized enterovirus surveillance system in China; thus, the seroprevalence of CVB3 is not well understood.

Shandong Province is located in eastern China and covers an area of 156,700 km2, which is inhabited by a population of 94.7 million. In the summer of 2008, an outbreak of CVB3-associated aseptic meningitis occurred in southern Shandong Province (6), and most of the cases were in children <15 years of age. CVB3 has also been demonstrated to account for 7.7% of all non-polio enterovirus isolates from acute flaccid paralysis cases in Shandong Province from 1990 to 2010 (6), suggesting that it is one of the predominant serotypes in mainland China.

To investigate the seroprevalence of CVB3 infection and identify susceptible populations, we conducted a seroepidemiological study in a healthy population residing in Yantai city, Shandong Province, China. Yantai is a coastal city with a population of 2.23 million. A representative panel of 373 serum samples was obtained in August 2010, which were stored at –20°C and inactivated at 56°C for 30 min before use. To discern the age distribution of CVB3 infections, 10 age groups were chosen (3–7 months, 8–12 months, 1 year, 2–4, 5–7, 8–10, 11–14, 15–19, 20–39, and >40 years) with each group comprising 27–40 people. The study protocol was approved by the Ethics Review Committee of the Shandong Center for Disease Control and Prevention.

Neutralizing antibody against CVB3 isolate 2008TC012 (GenBank accession no. FJ919564), the reference strain responsible for the aseptic meningitis outbreak in Shandong Province in 2008, was examined by a microneutralization test as described previously (7). For the assay, samples were diluted 1:4 to 1:1024. Twenty-five microliters of 100 × the tissue culture infective dose (TCID50) virus were mixed with 25 μL of the appropriate serum dilution and incubated for 2 h at 37°C, followed by addition of 100 μL of HEp-2 cell suspension. Each dilution was tested in duplicate. The antibody titer for each sample was the highest dilution to prevent the development of cytopathic effects in both wells. An antibody titer of ≥8 was considered positive. The geometric mean titer (GMT) was also calculated. For statistical analysis of the seroprevalence and GMT, the chi-squared test was performed using SPSS 17.0 statistical software. Differences with an error probability (P) value of <0.05 were considered statistically significant.

CVB3 was widely distributed in the population with a seroprevalence of 52.3%. No significant gender-specific difference in seroprevalence was observed; 53.9% of the females and 51.5% of the males (P = 0.639) showed neutralizing antibodies to CVB3.

Figure 1 shows age-dependent positive rates and GMTs of neutralizing antibody. CVB3 seroprevalence decreased from 43.2% in the first age group (3–7 months) to 3.7% in the second group (8–12 months). Subsequently, a significant increase (P < 0.001) was observed in the next four age groups (1, 2–4, 5–7, and
8–10 years). Further, except for a decrease in the group aged 11–14 years, the seroprevalence reached a plateau and was > 50% in the groups aged > 15 years old. There was no significant difference ($P = 0.094$) in the seroprevalence among the five groups aged 8–10, 11–14, 15–19, 20–39, and > 40 years. The GMT variation with age showed similar tendencies, and significant differences were observed among the four groups aged 1, 2–4, 5–7, and 8–10 years and between the groups aged 8–10 and 11–14 years.

In mainland China, due to the concern of large-scale HFMD epidemics since 2007, the seroprevalence of enterovirus (EV) 71 and CVA16, the main causative agents of HFMD, has been monitored (8). However, seroepidemiological information regarding other EV serotypes is limited. In the summer of 2008, an aseptic meningitis outbreak occurred in southern Shandong Province, in which a number of hospitalized children received attention from public health officials, and CVB3 was demonstrated to be responsible for the outbreak. Hence, the present study was subsequently conducted to investigate the seroprevalence of CVB3.

The present study revealed that maternal anti-CVB3 antibody levels rapidly waned after birth, and only 3.7% of children aged 8–12 months were positive for neutralizing antibodies. The increase in seroprevalence among the group aged 1–10 years implied that most CVB3 infections occurred in preschool and early-elementary school children. Thus, it is likely that playing together and sharing toys and teaching tools contribute to the spread of CVB3 infection. For children < 10 years of age, younger the age, lower the titer of neutralizing antibody, and higher the possibility of being infected by CVB3. These findings are consistent with age-specific seroprevalence plots of EV71 and CVA16 constructed from the data acquired from main-land China, Singapore, and Germany (7–9). Therefore, methods to prevent and control CVB3 (or other human EVs) associated diseases should focus on these age groups of children. For the group aged 11–14 years, the seroprevalence was also high (> 50%), but a dramatic decrease in the GMT value was observed. A previous study revealed that CVB3 has an epidemic pattern of long-term trends of circulation characterized by substantial fluctuations in circulation levels over time, including large peaks when the CVB3 serotype was among the most prevalent EVs (1). Hence, the low GMT level in the group aged 11–14 years not only reflects lower incidence of CVB3 infection in this age group but also suggests that local CVB3 activity occurred during a period of quiescence several years ago when this group was highly susceptible to CVB3 infection.

This is the first report to detail the seroepidemiology of CVB3 in mainland China. The results indicated that spread of CVB3 in Shandong Province occurred mostly in preschool and early-elementary school settings.

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Conflict of interest None to declare.

REFERENCES