

Abstract

Enterovirus (EV) is a RNA virus. Mutation was often found in enterovirus isolates due to the lack of RNA proof reading. The EVs that can not be typed by FA test had increased since 1999. The NT test is laborious and time consuming. Besides, the NT antibodies are expensive and are running out worldwide. To address the typing of EVs in recent years and to study the molecular epidemiology of EVs in the last two decades, the sequences of the VP4 region were analyzed. The results of pan EV typing were as follows. (1) Six pan EV isolates obtained in 2003 were: 3 strains of CA16 and CB3, respectively. (2) Seventeen pan EV isolates from 2002 were: 11 strains of E6, 2 strains of polio 3 and one each of polio2, E3, EV71 and CA, (3) Forty pan EV isolates obtained in 2001 were 18 strains of E30, 5 strains of CA16, 6 strains of E6, 4 strains of polio 1 and two each of polio 3, CA24, CB4, and one each of polio 2, E3. (4) Twenty-eight pan EV isolates from 2000 were: 16 strains of CA16, 8 strains of EV71 and one each of CA4, CA5, CA9, E30. (5) Three pan EV isolates obtained in 1999 were: 2 strains of CA16 and one strains of EV71. The typing by VP4 nucleotide sequence analysis is precise and rapid. The results of molecular epidemiology were as follow. (1) The CA16 isolates obtained in 2000 clustered in genotype B while the isolates from 2001 and 2003 clustered in genotype C. (2) The E30 isolates from 1993 and 2001 clustered in genotype 5. The nucleotide differences among 1993 and 2001 isolates were 7-13% while 0-7% differences were found among 2000 and 2001 isolates. (3) The E6 isolates from 2001-2002 outbreak had a variation of 22-27% in nucleotide sequence comparing to that of the prototype while only 3% variation was found among the isolates in the same outbreak . (4) The CA9 isolates in 1997 from Taiwan were similar to that of Rome isolates in 2002 (only 2-3% in nucleotide difference). However, 17-27% nucleotide differences were found among 1997 isolates from Taiwan and the isolates from US and Japan in 1998-2002. (5) The CB3 isolates obtained in 1995-1996 and 1999-2000 showed little nucleotide differences (0-6%) while 24-26% nucleotide differences were found comparing to that of the prototype. (6) There are two genotypes of CB5 in Taiwan. They showed 8-25% nucleotide difference with that of the Faulkner strain, the prototype. CB5 viruses were isolated since 1995 and turned out to be the major isolates in 2002-2003. (7) There are three EV71 genotypes worldwide. In Taiwan, most of the EV71 isolates in 1998 outbreak belonged to genotype C, only a few isolates belonged to genotype B. Intriguingly, all EV71 isolates during 1999-2003 belonged to genotype B. The results of this study revealed that typing and molecular epidemiological study of EV based on the VP4 gene analysis is simple, rapid, precise and cost effective. Information from this study will help the monitoring of the EV gene evolution of either endemic strains or imported strains. In addition, our information will help the Taiwan CDC to announce a precise disease alert and launch the effective prevention and control measures.

Keywords : enterovirus ; RT-PCR ; monoclonal antibody