

Abstract

Inactivated viral particles of Enterovirus type 71 were prepared to generate neutralizing antibodies successfully in immunized mice. We produced this prototype vaccine from Vero cells, 20-40 liters virus culture fluid per batch, to develop pilot plant scale study. The neutralization test was performed to investigate a dominant virus strain that elicited high potency and wide range of neutralizing antibody to other EV71 stains. We compare with 6 stains of 1998, 3 stains of 2000 and 6 stains of 2002. These stains were subcultured in Vero cells for three times and identified as Mycoplasma-negative by a culture method. The virus stock was stored at -80 °C. The virus suspension was harvested from the roller bottle culture system after the cytopathic effect up to 80% at multiplicity of infection 0.1-0.01. After removing cell debris by centrifugation, virus fluids were inactivated by formalin 1/4000 at 4 °C for 60 days. The virus fluid adsorbed with AlPO₄ pH 7.5 as a adjuvant was immunized in mice twice at the period of two weeks. The immunized sera collected by heat puncture were pooled from 15 mice. The different antigenicities of EV71 strains were compared by the cross-neutralization test.

Keywords : EV71 stains ; neutralization ; VP1 protein