Abstract

Background: Enteroviruses are human pathogens that, along with other 4 genera of picornavirudae, contribute to a significant morbidity and mortality of human and livestock. The 69 serotypes of enteroviruses are etiological agents of a wide spectrum of clinical diseases depending on the combination factors related to tissue tropism, virulence of each virus, and immunological responses of the host. Several recent outbreaks of neurological diseases in children throughout the world have singled out enterovirus 71 (EV71) to be the most important neurotropic enterovirus during the post poliomyelitis eradication era. The recent recurrence of EV71 outbreaks (1998, 2000, and 2001) in Taiwan has highlighted the urgency in developing antiviral and/or vaccines against EV71. Recent advances in human and microbial genome studies have created ample opportunities for rationale antiviral drug design by delineating the virus-host interactions during viral infection. The conduct of this proposed study will require expertise in virology, protein chemistry, molecular biology, veterinarian science, and immunology.

Aims: To identify, clone, express, and characterize mammalian cell receptor or coreceptors to EV71 for use in delineating host-virus interaction, and transgenic mouse model for vaccine testing and antiviral testing.

Results: By virus overlay protein binding assay (VOPBA), three cell-membrane proteins, sized 70KD, 60KD and 34KD, have been identified. It is also found that the striated muscle cells of newborn mice express EV71 binding proteins while the muscle cells of adult mice do not, a phenomenon compatible with in vivo infection pattern of the newborn and adult mice. The recombinant viral protein ?C VP1, that is fused with a calmodulin binding protein, is able to bind to the 60KD cellular protein. We have been able to obtain from RD cell membrane preps approximately 10 ug of the purified 60KD-cellular protein, which is now undergoing MOLDI-TOF analysis.

Experimental design: The study will be undertaken by reverse engineering methodology. Mainly, the 60KD membrane protein will be captured by VP1 and affinity calmodulin-resin column and analyzed by MALDI-TOF. Cloning and expression of this protein will be followed by in vitro and in vivo functional study. The hypothesis of a recepoter/coreceptor complex will be tested.
Key Word：Enterovirus71、Animal model、Vaccine