Association of Enterovirus 71 3C Protease Protein Activity with Clinical Feature

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Background/Objective
EV71 is the most important enterovirus, besides poliovirus, related to the fatal and morbid cases with hundreds of deaths, especially in Asia during large ourbreaks. To delineate pathogenesis of EV71 infection, we had studied EV71 viral genetics and correlated the results with their clinical outcome.

Method
We enrolled cases infected with EV71 from 1998 to 2003 in Taiwan. We tried to find the association between EV71 viral genetics and clinical outcome. EV71 3C is a protease known to involve in viral replication and RNA binding. We over-expressed wild EV71 3C and mutant EV71 3C, created by site-directed mutagenesis at 79th amino acid (T79A, T79I, and T79V), in SF268 cells and analyzed the protease activity by Western blot with antibodies of 3C substrates. We also produced EV71 infectious clone with wild 3C and mutant 3C, which were then transfected into RD cell. We accessed the viral replication rate by plaque assay. Finally, we analyzed host-virus protein interaction by immunoprecipitation of cell lysates.

Result
We found that the polymorphisms of EV71 3C 79th amino acid affect the clinical outcome. Concerning the protease activity, there were no difference of protease activity among the different variant EV71 3C. However, we found that EV71 with mutant 3C T79V had highest virus replication rate, followed by wild-type 3C. We also identified some important interacting proteins including modulation and degradation of mRNA, vesicle trafficking protein, and ATP synthase.

Conclusion
We found up-regulation of COX-2 by wild 3C but not mutant 3C, which might be a hint to inflammatory modulation through COX-2 by EV71. We also identified viral interaction with important host proteins for modulation and degradation of mRNA, vesicle trafficking, and ATP synthase. These interacting proteins might have important roles on EV71 infection and may have further clinical application in the future.