Impact of Influenza A virus PA-X protein on host immune responses

Influenza A virus is a clinically important respiratory pathogen that annually infects millions of people worldwide and is, therefore, a serious threat to public health. Recurrent influenza infections occur throughout life because a relatively weak immune response is generated against viral infection. This weak immune response is partly due to an ability of influenza virus in suppressing general host protein synthesis, known as host shutoff. The host shutoff activity is believed to allow the virus to escape the host immune system by suppressing antigen presentation and limiting host immune recognition, allowing extended viral shedding. However, it is unclear how influenza virus induces host shutoff. In 2012, a novel influenza virus protein, named PA-X, was found to be expressed from viral PA mRNA via ribosomal frameshifting. Characterization of the PA-X of highly pathogenic 1918 pandemic influenza virus indicated that PA-X is a major factor for suppression of host protein synthesis.. However, whether PA-X protein of the currently circulating 2009 pandemic H1N1 virus (pH1N1) actually had any impact on viral growth, pathogenicity and host immune response is not known.

We rescued and characterized a mutant pH1N1 virus expressing reduced levels of PA-X due to mutations at the frameshift motif of the PA gene (PA-XFS). Host shutoff activity of the mutant PA-XFS virus was less efficient than that observed in wild type (wt) pH1N1-infected cells, proving a strong impact of PA-X in pH1N1 influenza-induced host shutoff. We detected host mRNA degradation in wt pH1N1-, but not in PA-XFS-infected cells, supporting that pH1N1 PA-X induces host shutoff by degrading host mRNA, as reported in 1918 pandemic virus. We also found that PA-XFS virus was attenuated in cultured human cells and infected mouse lungs. Interestingly, PA-XFS virus induced a stronger interferon response, which likely contributed to a reduction of viral growth in cultured cells and in mice. These data suggest that PA-X enhances viral growth by restricting the host innate antiviral response both *in vitro* and *in vivo*. Moreover, PA-XFS-infected mice cleared the virus and recovered significantly faster than wt-infected mice, implying that PA-X blocks proper antigen presentation necessary for viral clearance. Importantly, PA-XFS virus induced a stronger humoral response than wt virus in mice, despite its reduced viral replication in lungs. These data suggest that PA-X could also suppress the host adaptive immune response.

Vaccination is the primary approach for protection from influenza infection. Despite extensive efforts by many researchers, efficacy of the current influenza vaccine is 'sub-optimal' due to insufficient innate and adaptive immune response against the virus. Our study showed that PA-X expressed by influenza infection suppresses host innate and adaptive immune responses by inducing general host shutoff in infected cells, which likely contributes to annual epidemics of the virus. Our findings also suggest a potential approach to improve the efficacy of live attenuated vaccines through suppressing PA-X expression. Such PA-X-deficient live vaccine might have “optimal” efficacy for protection from annual influenza virus infections.

Tsuyoshi Hayashi, ***Chutikarn Chaimayo***, ***Toru Takimoto***
Department of Microbiology and Immunology, University of Rochester Medical Center
Rochester, New York, USA

Publication

Influenza A Virus Protein PA-X Contributes to Viral Growth and Suppression of the Host Antiviral and Immune Responses
Hayashi T, MacDonald LA, Takimoto T
J Virol. 2015 Jun