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RESEARCH ARTICLE

Age-Dependent Pathogenesis of Influenza A Virus H7N9 Mediated Through PB1-F2-Induced Mitochondrial DNA Release and Activation of cGAS-STING-NF-κB Signaling

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ABSTRACT

Exactly why human infection of avian influenza A virus H7N9 causes more severe disease in the elderly remains elusive. In this study, we found that H7N9 PB1-F2 is a pathogenic factor in 15–18-month-old BALB/C mice (aged mice) but not in 6–8-week-old young adult mice (young mice). Recombinant influenza A virus with H7N9 PB1-F2-knockout was less pathogenic in aged mice as indicated with delayed weight loss. In contrast, survival of young mice infected with this virus was diminished. Furthermore, tissue damage, inflammation, proinflammatory cytokine and 2′3′-cGAMP production in the lung were less pronounced in infected aged mice despite no change in viral titer. cGAS is known to produce 2′3′-cGAMP to boost proinflammatory cytokine expression through STING-NF-κB signaling. We found that H7N9 PB1-F2 promoted interferon β (IFNβ) and chemokine gene expression in cultured cells through the mitochondrial DNA-cGAS-STING-NF-κB pathway. H7N9 PB1-F2 formed protein aggregate and caused mitochondrial cristae collapse, complex V-dependent electron transport dysfunction, reverse electron transfer-dependent oxidized mitochondrial DNA release to the cytoplasm and activation of cGAS-STING-NF-κB signaling. PB1-F2 N57 truncation, which is frequently observed in human circulating strains, mitigated H7N9 PB1-F2-mediated mitochondrial dysfunction and cGAS activation. In addition, we found that PB1-F2 of pathogenic avian influenza viruses triggered more robust cGAS activation than their human-adapted descendants. Our findings provide one explanation to age-dependent pathogenesis of H7N9 infection.

1 | Introduction

Uncontrolled pulmonary inflammation is a major cause of acute respiratory distress syndrome (ARDS) induced by influenza A virus (IAV) infection [1, 2]. Overproduction of proinflammatory cytokines results in the recruitment of leukocytes such as monocytes and neutrophils to the infected respiratory sites and their consequent excessive activation [3]. Dense and activated leukocytes generate damaging molecules such as nitric oxide, superoxide, neutrophil extracellular traps and more proinflammatory cytokines that amplify the unwanted immune responses. Ultimately, the epithelial–endothelial barrier is damaged leading to acute lung injury, accumulation of fluid, downregulation of gas exchange, and pulmonary dysfunction.

In 2013–2019, a case fatality of 39.3% was documented for human infection of H7N9 virus [4]. Severe H7N9 infection with pulmonary

Pak-Hin Hinson Cheung and Tin-Long Yuen contributed equally to this article.