INDIAN INSTITUTE OF TECHNOLOGY GUWAHATI
SHORT ABSTRACT OF THESIS

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Thesis Title: **"Understanding the role of Newcastle disease virus-mediated cellular stress and altered proteostasis: Outsmarting the smart virus"**

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SHORT ABSTRACT

Viruses are known to instigate a variety of stresses in their host cells, including endoplasmic reticulum (ER) and oxidative stress, to affect normal cell functioning and their replication cycle. The virus-induced stress subsequently triggers cellular stress responses by activating numerous stress-responsive proteins (SRPs). Afterward, all the viral proteins and SRPs substantially exacerbate ER protein flux, eliciting the unfolded protein response (UPR). This can be characterized by the modulation of Glucose-Regulated Protein 78 (GRP78) as an ER stress sensor. Moreover, viruses have also been shown to induce oxidative stress by disrupting the balance of reactive oxygen species (ROS) inside the cells. However, the purpose of virus-induced stresses and the precise role of SRPs in infection remain elusive. The present work utilizes the Newcastle disease virus (NDV) to investigate the role of cellular stress and the function of specific SRPs in pathogenesis.

Firstly, the thesis deals with the instigation of ER and oxidative stress following NDV infection. Here, we show that NDV induces ER stress, confirmed by the GRP78 upregulation, and oxidative stress, established by the accumulation of intracellular ROS levels. Additionally, the results demonstrate that NDV infection modulates the expression of key oxidative stress-responsive genes, including Nrf2, HO-1, and SOD-1. The members of the

sirtuin (SIRT) family were also found to be differentially modulated due to NDV infection. Secondly, the thesis specifically focuses on establishing the function of SIRT7 protein in NDV pathogenesis. SIRT7 primarily functions as a NAD+-dependent histone deacetylase enzyme, thus regulating multiple biological processes, including genome stability, metabolic pathways, and stress responses. However, its enzymatic

activity, physiological function, and target proteins in virus-infected cells remain unexplored. Here, the detailed mechanistic studies reveal that elevated expression of NDV-mediated SIRT7 protein metabolizes the NAD+ to deacetylate the host proteins, thus contributing to high virus replication. Considering the results, we establish the constructive role of SIRT7 in NDV replication.

The heightened production of viral proteins and SRPs can eminently disrupt protein homeostasis, prompting the cells to activate specific proteolytic pathways. These pathways employ post-translational modifications (PTMs) to direct ubiquitin-mediated proteasomal degradation of aberrantly accumulated proteins. Among various PTMs, the addition of Arg to the N-terminus of target proteins (Arginylation) by arginyltransferase 1 (ATE1) enzyme remains unexplored mainly in virus pathogenesis. Lastly, the thesis delves into the mechanism of arginylation on host proteins and the engagement of proteasomal degradation machinery. The results show that NDV alters proteostasis, prompting the cells to activate the ATE1 enzyme for arginylation. Here, we also explored the HN protein of NDV as a presumable target for arginylation-mediated degradation.

The thesis findings collectively indicate that NDV infection induces stress within cells, resulting in the upregulation of numerous SRPs. Among these SRPs, the SIRT7 protein was observed to facilitate NDV infection by deacetylating the host proteins, thereby impeding the cellular antiviral response. Subsequently, excessive protein production leads to proteostasis imbalance, which is further mitigated by the activation of proteolytic pathways. We also confirmed the active participation of the arginylation process in infected cells. Finally, this process can be employed to degrade the HN-like viral proteins, thereby establishing a unique host defense mechanism.

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