Imaging Viral Load in Chicken Embryos

Introduction

Newcastle Disease Virus (NDV) is a contagious disease, capable of transmission to humans, and with a high risk of a severe epizootic in birds, particularly domestic poultry which are typically reared at high density.

While the effects of NDV are mild in humans, its effect in avian hosts are more pronounced and vary based on the age and species of the host, as well as the strain of the virus. Three strains of NDV, with differing levels of virulence, have been well characterized in chicken eggs. Strains of intermediary virulence are known to infect primarily the umbilical tissue of the chicken embryo, whilst more virulent strains infect other tissues indiscriminately.

As such many studies have been performed to investigate the impacts of the varying strains along with the mechanisms of viral load in domestic chickens. However, such studies have often been limited in their ability to accurately and rapidly assess viral load in vivo. With the development of sensitive high-resolution imaging systems, such as the Azure c600, it is now possible to quickly and easily image fluorescently tagged NDV in chicken embryos, and, to some extent, quantify the viral load in various embryonic tissues.

Methods

Chicken embryos were infected at E10 and incubated for a further 72 hours. Embryos were infected with either 100-1000 PFU of virulent or non-virulent recombinant NDV tagged with eGFP, and compared with a non-infected control. The embryos were placed on petri dishes and imaged with the Azure c600 using Epi Blue imaging mode with an automated capture.

Results and Conclusions

Figure 1 shows an embryo infected with a non-virulent form of the virus next to an uninfected embryo (negative control). The infected embryo shows the non-virulent strain of the virus confined primarily in the umbilical tissue.

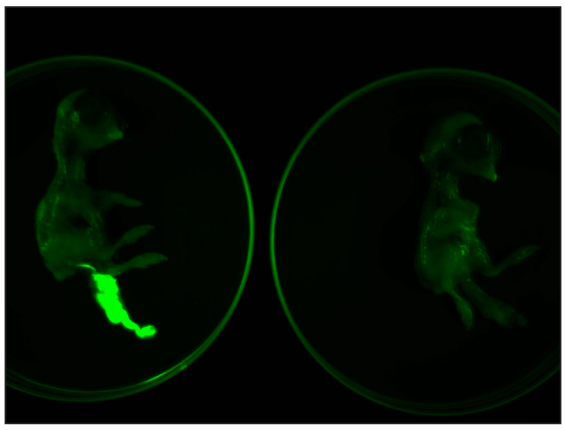

Figure 1. NDV eGFP No-VIR (left) and Negative Control (right).

Figure 2 shows the same non-virulent virus-infected embryo imaged alongside an embryo infected with a more virulent strain of NDV. The virulent form of the virus infects the embryo more indiscriminately, and the virus is detected throughout the embryo.

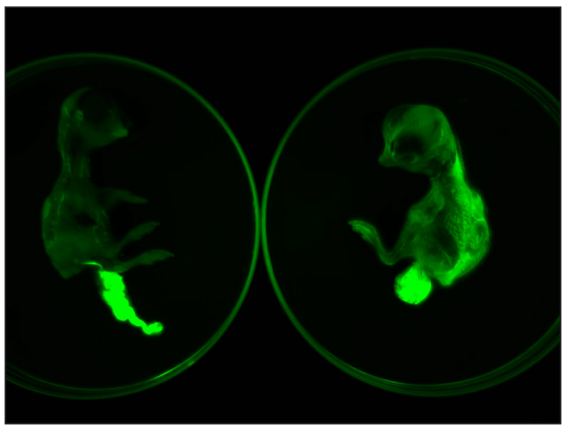

Figure 2. NDV eGFP No-VIR (left) and NDV eGFP VIR (right).

Figure 3 shows the extent of the virulent infection compared to the negative control, uninfected embryo.

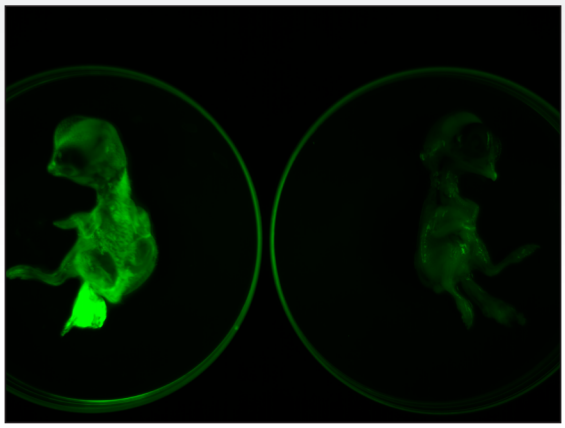

Figure 3. NDV eGFP VIR (left) and Negative Control (right).

Conclusions

The utility of fluorescent imagers like the Azure Biosystems c600 extends far beyond Western blotting, including a large number of applications that utilize fluorescence excitation. This application note showcases one such application – imaging a fluorescently tagged virus in animal models allowing for the generation of high-resolution quantifiable images.

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