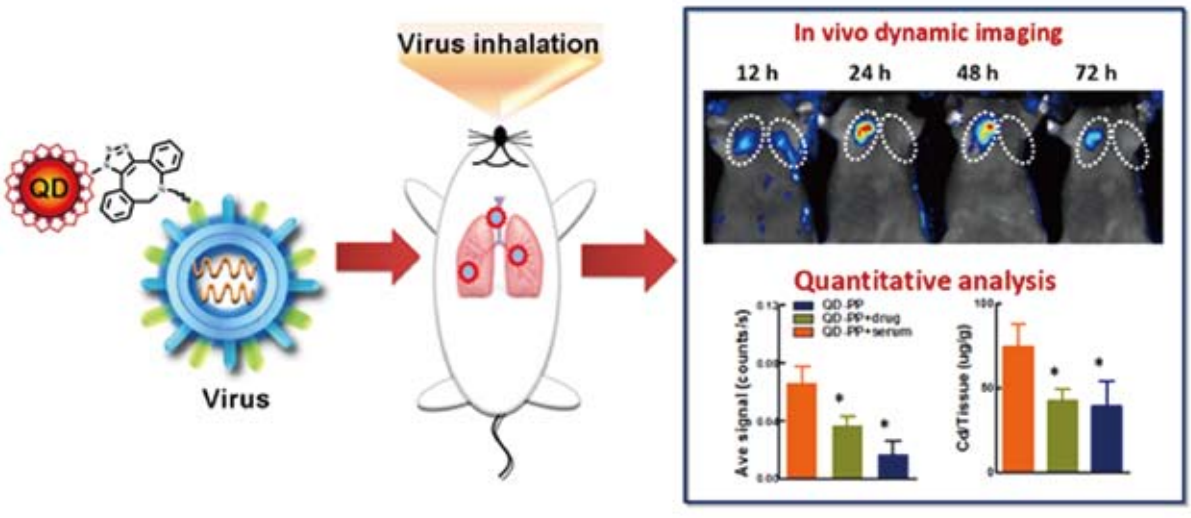
Monitor *In Vivo* Dynamics of Avian Influenza Viruses Using Bioorthogonal-conjugated Near-infrared Quantum Dots

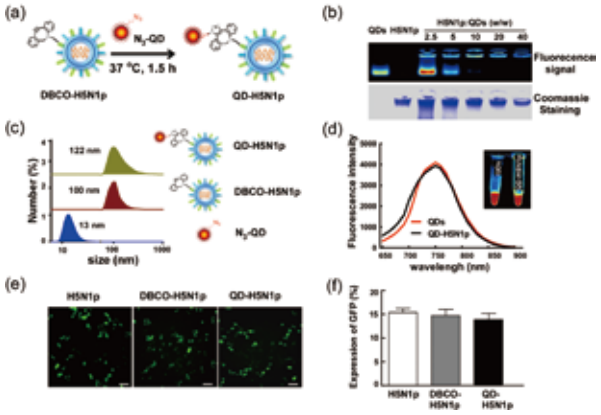
Highly pathogenic avian influenza A viruses are emerging pandemic threats that have caused several outbreaks with high mortality in human beings in the past decades. The infection of avian influenza viruses, such as subtype H5 and H7, could cause a series of severe respiratory and extra-respiratory complications, both of which are directly associated with viral loading and dissemination in tissues. Understanding the route and pathogenesis of avian influenza viral infection is extremely important for developing prophylactic and therapeutic strategies against viral infections. Although the studies using cell cultures have made significant contributions to exploring the pathogenesis of avian influenza viruses, it is highly desirable to track the in vivo dynamics of viral infection in order to further dissect the interactions between the virus and the host.

Viral particle labeling with quantum dots (QDs) has emerged as an effective strategy for virus tracking. Compared with conventional dyes, QDs demonstrate bright photoluminescence, broad size-tunable emission spectrum, and photochemical stability and have been successfully applied for single-virus tracking in vitro. Especially, QDs with near-infrared (NIR) spectrum from 700 to 900 nm have attracted intense attention due to the ability of deep-tissue imaging in this wavelength range. Bioorthogonal chemistry, a biocompatible chemical reaction that can occur in the presence of other rich chemical functionalities found in biological systems without interacting or interfering with native biochemical processes, allows a wide range of bioactive molecules to be specifically labeled in living organisms. Previous studies reported that the copper-free click chemistry is a simple, effective, and bioorthogonal strategy for live viral labeling and does not significantly affect viral infectivity.

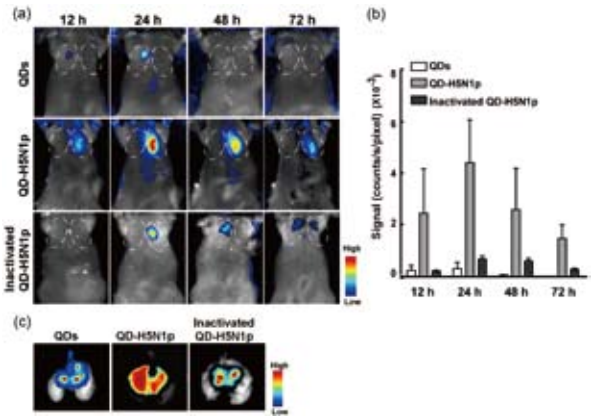
Dr. CAI Lintao, Dr. MA Yifan and their coworkers at the Shenzhen Institutes of Advanced Technology (SIAT)

Schematic illustration of the bioorthogonal labeling and in vivo imaging of viruses using conjugated near-infrared quantum dots.





Bioorthogonal Labeling of H5N1p with NIR QDs.



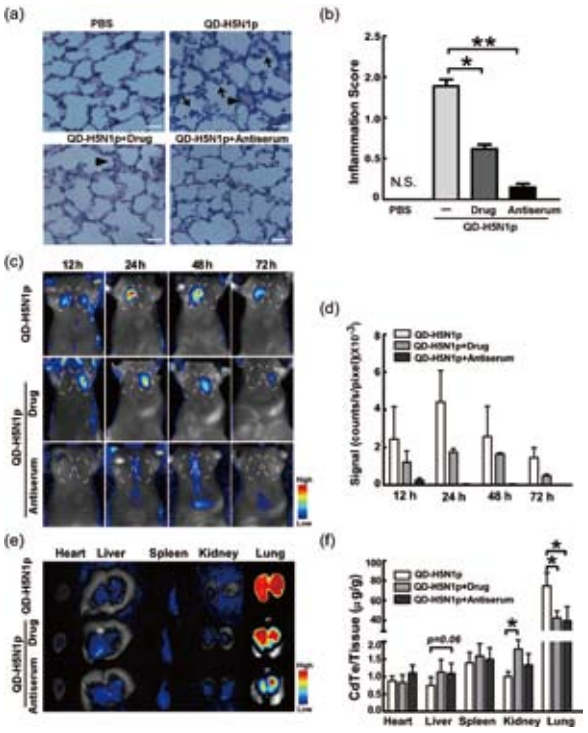
In vivo imaging of respiratory QD-H5N1p infection in mice.

worked together to label avian influenza H5N1 pseudotype virus (H5N1p) with NIR QDs by bioorthogonal reaction, and dynamically monitored the respiratory infection of QD-labeled H5N1p (QD-H5N1p) in mice using an in vivo imaging system. QD-H5N1p demonstrated bright and sustained fluorescent signals in mouse lung tissues, which were strongly correlated with the severity of viral infection that was verified by lung histology. The scientists also explored the potential application of QD-H5N1p for antiviral drug evaluation. Results showed that the administration of the antiviral agents oseltamivir carboxylate and mouse antiserum significantly affected the in vivo dynamics of QD-H5N1p infection, which could be directly quantified by measuring fluorescent signals and cadmium (Cd) concentration of virus-conjugated QDs using Inductively coupled plasma/optical emission spectrometry (ICP-OES).

Their paper, entitled "Noninvasive Visualization of Respiratory Viral Infection Using Bioorthogonal Conjugated Near-Infrared-Emitting Quantum Dots", was recently published online by *ACS NANO*.

The research was supported by the National Basic Research Program of China, the National Natural Science Foundation of China, the "Hundred Talents Program" of the Chinese Academy of Sciences, the Science and Technology Foundation of Guangdong Province, Guangdong Innovation Research Team of Low-cost Healthcare and the SIAT Innovation Program for Excellent Young Researchers.

Dr. CAI Lintao's group at SIAT has been carrying out world-leading research on a wide range of biomedical nanotechnologies. They study multifunctional and nanostructured composite materials, provide highly sensitive and bioorthogonal labeling methods for in vivo imaging and molecular diagnosis in nanoscale and single molecular level, and explore new device concepts and self-assembly techniques for the development of biomedical nanodevices and sensors for biosensing. Dr. MA Yifan's group focuses on cutting-edge research into nanoparticle-based adjuvant and vaccine delivery systems immune regulation mechanism of nanomaterials, the in vivo tracking of virus and cell using NIR imaging technology, as well as cancer immunotherapy.



The effect of antiviral agents on the dynamics of respiratory QD-H5N1p infection in mice.