ID No.	K3005
Project Title	A nanocaged nanobody display platform for infectious diseases detection and therapy
Principal Investigator	Xiyun Yan (Prof., Institute of Biophysics(IBP), Chinese Academy of Sciences (CAS))
Project Members IMSUT Host Researcher Members	 Yasushi Kawaguch (Prof., IMSUT) Kelong Fan (Prof., IBP, CAS) Jiuyang He (Postdoc, IBP, CAS) Bing Jiang (Associate Prof., Zhengzhou Univ.)

Report

During the past 2021, we have achieved great progress on our research project. Since we have successful constructed fenobody protein nanocage by displaying 24 H5N1-specific nanobodies on the surface the ferritin nanocage through genetic engineering approach, we then verified the specificity and binding affinity of fenobody to the H5N1 virus in the past year. Fenobody system has been proved to have great potential in improve the apparent affinity and *in vivo* half-life of nanobody.

Then we tried to expend the applications of fenobody system. First, we loaded Fe_3O_4 nanoparticles into the cavity of fenobody nanocage by biomineralization synthesis. Fe^{2+} can enter the cavity of ferritin protein shell through hydrophilic channels freely, then Fe^{2+} can be catalyzed to form metallic oxide core at the ferroxidase site of the cavity of fenobody. By adding the additional oxidizing agents H_2O_2 and NaOH, the oxidation of metal ions was accelated, and finally Fe_3O_4 core was formed in the cavity of fenobody nanocage. The synthesized Fe_3O_4@Fenobody possesses peroxidase-like activity which catalyzed the oxidation of peroxidase substrate TMB and produce colored products. Fe_3O_4@Fenobody acted as an enzyme-labeled antibody, which may recognize H5N1 antigen and catalyze substrate color reaction. Through double-antibody sandwich ELISA, we used H5N1 as capture antibody, and Fe_3O_4@Fenobody was used as detection antibody, without the addition of enzyme-labeled secondary antibody, Fe_3O_4@Fenobody successfully recognized H5N1 (1:100 dilution).

Next, we will further test the availability of Fe_3O_4@Fenobody to be used in the test strip rapid detection technology for rapid virus detection.