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ORAL ABSTRACT PRESENTATIONS

SESSION TITLE: STRUCTURE OF VIRUSES AND CHAPERONINS

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Abstract OR-8: Cryo-EM Structure of Mature Yellow Fever Virus

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Background: Yellow fever virus (YFV) is the prototype virus of the genus *Flavivirus*. It is endemic to sub-Saharan Africa and tropical South America. YF disease ranges from asymptomatic to severe jaundice and hemorrhagic fever. The flavivirus virion core is enveloped by a lipid membrane with integrated membrane (M) proteins and envelope (E) proteins that form the outer surface of the virion. The E protein provides stability to the viral particle and is responsible for early infection stages. Flaviviruses are heterogeneous in nature, which is related to their maturation process. Samples always contain mature, immature, half-mature, and damaged particles. Thus, cryo-EM is a method of choice for their structure determination. During the early stages of cryo-EM development, structures of flaviviruses were studied at 10–20 Å resolution. However, due to the progress of recent years, it became possible to determine flavivirus structures at a resolution of 5.6-2.6 Å. The cryo-EM method was used to obtain structural data of virions of dengue fever virus, Zika virus, TBE virus, etc. For YFV, only the cryo-EM structure of the immature virions at a low resolution of 25 Å was determined (Y. Zhang et al. 2003 doi:10.1093/emboj/cdg270). However, the structure of mature (most *infectious*) YFV particles is still unknown.
Methods: Virus sample was produced in Vero cell culture. YFV-17D was inactivated and purified using ultracentrifugation. The concentration of viral particles in the target inactivated YFV-17D (iYFV-17D) was evaluated by

spectrophotometry and by estimating the concentration of E protein determined by PAGE electrophoresis. Preliminary quality control of iYFV-17D sample was performed using negative staining TEM. Cryo-EM data were collected using cryo-TEM Krios (Thermo-Fisher, USA) at 300kV using DED Falcon II. Dataset was preprocessed using Warp. Further processing was performed in Relion 3.1 and CisTEM. Model building was carried out using Isolde, Phenix and Coot software.

Results: A protocol of production and purification of highly concentrated (~$2x10^{12}$) monodisperse inactivated iYFV-17D sample was developed. Cryo-EM structure of mature iYFV was solved at 4.1 Å resolution. The structure of YFV is similar to other known flavivirus structures, with 180 copies of protein E arranged in a herringbone pattern that makes up the icosahedral shell.

Conclusion: The high-resolution structure of mature iYFV-17D allowed to elucidate special features of this flavivirus and may be useful for vaccine improvement and drug development.

Key Words: flavivirus • cryoEM • structure

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