OFFICIAL ABSTRACT and CERTIFICATION

CCDC11 Acts as a Scaffold to Assemble the ESCRT Membrane Scission Machinery at Viral Budding Sites for HIV-1 Release: Identifying a Novel Therapeutic Strategy for Antiviral Therapy

Poojan Pandya and Leo Takemaru

Half Hollow Hills High School West, Dix Hills, NY and Ward Melville High School, East Setauket, NY, USA

We previously reported that Coiled-Coil Domain-Containing 11 (CCDC11) is critical for the production of Human Immunodeficiency Virus (HIV) from Human Embryonic Kidney (HEK) 293T cells likely through recruitment of the Endosomal Sorting Complex Required for Transport (ESCRT) machinery. To further extend the role of CCDC11 in this process, we attempted to knockout CCDC11 in human cervical cancer HeLa cells, which is a widely used model for HIV research, using the CRISPR-Cas9 technology. We identified three CCDC11-deficient HeLa cell lines with distinct mutations after sequencing, and a significant reduction in CCDC11 protein levels was verified by western blotting and immunofluorescence microscopy. As expected, HIV-1 release into culture media from the CCDC11-deficient cells was dramatically decreased. Consistent with our hypothesis that CCDC11 is required for viral budding, CCDC11 partially colocalized with Virus-Like Particles (VLPs), produced by ectopic expression of the HIV-1 Gag structural protein, at the plasma membrane of HeLa cells. In addition, we found that CCDC11 colocalizes with the ESCRT-I component TSG101 and the ESCRT-III components CHMP2A and CHMP4B, but not with ALIX, CHMP6, or CHMP8. In support of these findings, we found that CCDC11 physically interacts with CHMP2A and CHMP4B. Collectively, our data confirm our previous results that CCDC11 plays a key role in HIV production and provide the first evidence that CCDC11 promotes HIV budding through direct recruitment of CHMP2A and CHMP4B to viral budding sites at the plasma membrane. Protein-protein interactions between CCDC11 and CHMP2A, or CHMP4B can be exploited as a novel therapeutic strategy for antiviral therapy.

Category
Pick one only — mark an "X" in box at right

- Animal Sciences
- Behavioral & Social Sciences
- Biochemistry
- Biomedical & Health Sciences
- Biomedical Engineering
- Cellular & Molecular Biology
- Chemistry
- Computational Biology & Bioinformatics
- Earth & Environmental Sciences
- Embedded Systems
- Energy: Sustainable Materials and Design
- Engineering Mechanics
- Environmental Engineering
- Materials Science
- Mathematics
- Microbiology ■
- Physics & Astronomy
- Plant Sciences
- Robotics & Intelligent Machines
- Systems Software
- Translational Medical Sciences

1. As a part of this research project, the student directly handled, manipulated, or interacted with (check ALL that apply):

   ☐ human participants ■ potentially hazardous biological agents

   ☐ vertebrate animals ■ microorganisms ■ rDNA ■ tissue

2. I/we worked or used equipment in a regulated research institution or industrial setting: ■ Yes ☐ No

3. This project is a continuation of previous research. ■ Yes ☐ No

4. My display board includes non-published photographs/visual depictions of humans (other than myself): ☐ Yes ■ No

5. This abstract describes only procedures performed by me/us, reflects my/our own independent research, and represents one year's work only ■ Yes ☐ No

6. I/we hereby certify that the abstract and responses to the above statements are correct and properly reflect my/our own work. ■ Yes ☐ No

This stamp or embossed seal attests that this project is in compliance with all federal and state laws and regulations and that all appropriate reviews and approvals have been obtained including the final clearance by the Scientific Review Committee.