Correction for Fernández de Castro et al., Reovirus Forms Neo-Organelles for Progeny Particle Assembly within Reorganized Cell Membranes

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In our article (1), we reported that smooth membranes connected with rough endoplasmic reticulum (ER) cisternae are incorporated within inclusion structures formed in reovirus-infected cells using both two-dimensional and three-dimensional electron microscopy (Fig. 1, 3, 5, and 6). Using rabbit polyclonal antisera raised against ER-Golgi intermediate compartment 53 (ERGIC-53) (H-245) and the KDEL receptor (KDEL-R) (FL-212), both of which were purchased from Santa Cruz Biotechnology, we observed staining attributable to ERGIC-53 (Fig. 4G to L) and KDEL-R (Fig. S2J to L) in reovirus inclusions in both HeLa cells and MDCK cells. We concluded from these observations that ERGIC, a membranous system that functionally links the ER and Golgi compartment, is a source of inclusion-associated membranes.

As part of our ongoing studies of the biogenesis of reovirus inclusions, we performed immunofluorescence assays using two monoclonal antibodies specific for ERGIC-53 (C-6 [Santa Cruz Biotechnology] and G1/93 [Enzo Life Sciences]) and a monoclonal antibody specific for KDEL-R (KR-10 [Thermo Scientific]). With these antibodies, we were unable to detect distribution of either ERGIC-53 or KDEL-R within viral inclusions. Immunoblotting experiments using either ERGIC-53 (H-245) or KDEL-R (FL-212) polyclonal antiserum revealed multiple protein bands in both uninfected and infected cells (Fig. 1). These new findings do not alter the main conclusion of our paper, which is that reovirus inclusions are embedded in membrane. The origin of the membranous elements that build reovirus inclusions is a focus of our current research.

Poor specificity of commercial antibodies has been reported previously (2, 3). Our findings using the H-245 and FL-212 antisera are most likely attributable to the nonspecific immunoreactivity of these reagents. We apologize for the erroneous conclusions made from the experiments using these antisera and alert readers to potential problems with the use of polyclonal antisera for immunohistochemical staining.

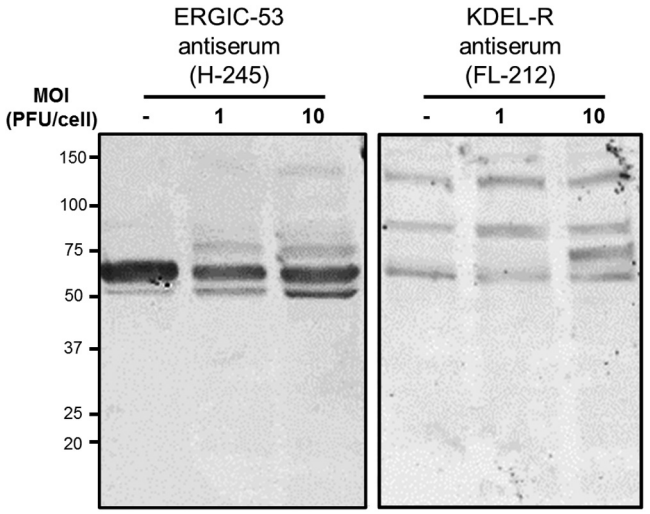

FIG 1 ERGIC-53 (H-245) and KDEL-R (FL-212) polyclonal antisera from Santa Cruz Biotechnology detect multiple protein bands. HeLa cells were infected with reovirus at a multiplicity of infection (MOI) of 1 or 10 PFU/cell. Cell lysates were prepared 24 h postinfection, resolved by SDS-PAGE, and transferred to nitrocellulose membranes. Membranes were blocked with 5% skim milk in TBS and immunoblotted using polyclonal antiserum H-245 (against ERGIC-53, left panel) or FL-212 (against KDEL-R, right panel). Anti-rabbit secondary antibody (IRDye 800CW goat anti-rabbit IgG [LI-COR]) was used for detection. Molecular mass markers (in kilodaltons) are indicated on the left.

REFERENCES

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