Is Lgr4 essential for VSV– and VSV-G–pseudotyped lentiviral vector entry to cells?

DOI 10.1074/jbc.L117.000471

Menachem Rubinstein[1]

From the Department of Molecular Genetics, The Weizmann Institute of Science, Rehovot 7610001, Israel

Edited by Charles E. Samuel

A recently published paper concluded that Lgr4 is essential for vesicular stomatitis virus (VSV) entry to cells (1), in contrast with independently confirmed studies, showing that VSV infects cells through the LDL receptor (LDLR) and/or its family members (2–4). This report raises many concerns, suggesting that Lgr4 is not involved in VSV entry. I will address three key issues.

1. VSV-G and Lgr4 ectodomains (ECD) have been expressed without the signal peptide. Hence, they did not translocate into the ER and, therefore, did not form correct disulfide bridges, nor acquire the oligosaccharide side chains, which are important for correct folding of the VSV-G ectodomain. Therefore, the described co-immunoprecipitations (Fig. 5*B* of Ref. 1) could stem from aggregation of the misfolded VSV-G–ECD and LGR4-ECD.

2. The interpretation of the Biacore experiments (Fig. 5*D* of Ref. 1) is very difficult, as most of the binding shown is irreversible. The graph shows only association curves, whereas the dissociation curves are missing for all but the highest concentration. A quick look suggests a half-effect at 500–700 nM, and, therefore, the claimed K_d of ~70 nM cannot be correct.

3. Furthermore, the claimed 70 nM value is not consistent with the concentration required for VSV neutralization (200 μg/ml, ≅4 μM, Fig. 6*A* of Ref. 1). By comparison, the IC_{50} of LDLR-ECD is 55 ng/ml (2). Thus, on weight basis, LGR4-ECD is ~3600 times less potent than LDLR-ECD. Therefore, the author's statement that "soluble LGR4-ECD … could inhibit VSV infection… showing great potential for treating vesicular stomatitis" is unfounded.

The author declares that he has no conflicts of interest with the contents of this article.

[1] To whom correspondence should be addressed. E-mail: Menachem.rubinstein@weizmann.ac.il.

References

1. Zhang, N., Huang, H., Tan, B., Wei, Y., Xiong, Q., Yan, Y., Hou, L., Wu, N., Siwko, S., Cimarelli, A., Xu, J., Han, H., Qian, M., Liu, M., and Du, B. (2017) Leucine-rich repeat-containing G protein–coupled receptor 4 facilitates vesicular stomatitis virus infection by binding vesicular stomatitis virus glycoprotein. *J. Biol. Chem.* **292,** 16527–16538 CrossRef Medline
2. Finkelshtein, D., Werman, A., Novick, D., Barak, S., and Rubinstein, M. (2013) LDL receptor and its family members serve as the cellular receptors for vesicular stomatitis virus. *Proc. Natl. Acad. Sci. U.S.A.* **110,** 7306–7311 CrossRef Medline
3. Amirache, F., Lévy, C., Costa, C., Mangeot, P. E., Torbett, B. E., Wang, C. X., Nègre, D., Cosset, F. L., and Verhoeyen, E. (2014) Mystery solved: VSV-G-LVs do not allow efficient gene transfer into unstimulated T cells, B cells, and HSCs because they lack the LDL receptor. *Blood* **123,** 1422–1424 CrossRef Medline
4. Scott, C. C., Vossio, S., Vacca, F., Snijder, B., Larios, J., Schaad, O., Guex, N., Kuznetsov, D., Martin, O., Chambon, M., Turcatti, G., Pelkmans, L., and Gruenberg, J. (2015) Wnt directs the endosomal flux of LDL-derived cholesterol and lipid droplet homeostasis. *EMBO Rep.* **16,** 741–752 CrossRef Medline