



UMR INSERM1173 Infection et Inflammation Université de Versailles St-Quentin-en-Yvelines

Dr. Marie-Anne Rameix-Welti Tel: +33 (0)6 65 13 64 34

E-mail: marie-anne.rameix-welti@uvsq.fr

Paris, September the 26th 2018

Dear editor,

We submit a manuscript entitled "Generation, amplification and titration of recombinant respiratory syncytial viruses to monitor viral multiplication and viral factories dynamics." for publication in Jove responding to your kind invitation.

Reverse genetics is a powerful technology widely used in virology field. Rescue, amplification and titration of recombinant RSV have been long published. However, these "basic" methods remain difficult to handle and require a specific knowhow.

In this manuscript we describe in details the rescue and the amplification of recombinant RSV using 2 examples. We also detail an original titration assay method using microcrystalline cellulose (Avicel) overlay. We then illustrate how recombinant RSV expressing a reporter gene might be used to monitor viral multiplication to assess the impact on viral growth of drugs or cellular protein knock down. We also provide example of analysis of the dynamics of viral structure thanks to a recombinant RSV expressing a viral fluorescent protein.

RSV infection burden is major, leading to significant mortality in infants and elderly worldwide. There are still no vaccine neither efficient antivirals available yet. The tools and methods described here would facilitate the comprehension of RSV multiplication mechanisms and development of potential antivirals. These methods, especially the plaque assay protocol, may be adapted to other viruses.

We strongly believe that the present study will be of high interest to the readership of Jove and look forward to hearing back from you about this submission,

Yours sincerely,

Marie-Anne Rameix-Welti