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Comparison of lentiviral and vesicular stomatitis virus core SARS-CoV-2 pseudotypes and generation of a stable cell line for use in antibody neutralisation assays

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Betacoronavirus SARS-CoV-2, the causative agent of COVID19, is a single stranded positive sense RNA virus. Since its emergence there has been great efforts to identify correlates of protection,which is crucial for vaccine evaluation studies. However, handling SARS-CoV-2 requires BSL-3 containment facilities slowing research efforts. Pseudotype viruses (PV) are a safe alternative to authentic virus that can be handled at low containment. PVs are chimeric viruses containing the core of a virus where its genome has been completely or partially replaced by a reporter gene, displaying a correctly folded SARS-CoV-2 spike on its surface. We developed lentiviral and vesicular stomatitis virus (VSV) core PVs alongside a stable A549 cell line expressing receptor ACE2 and protease TMPRSS2 responsible for S protein priming, for use in neutralization assays. Lentiviral PVs were generated by transfection with plasmids encoding the spike, HIV-1 gag-pol and a luciferase reporter. For VSV PVs, producer cells pre-transfected with the spike were infected with recombinant VSV expressing luciferase,before harvesting. The stable A549 cell line was generated by sequential infection of VSV-G PVs bearing lentiviral vectors encoding ACE2 and TMPRSS2 genes followed by antibiotic selection, before being tested in neutralization assays. We compared lentiviral and VSV PV platforms using monoclonal antibodies and convalescent sera with our stable A549 cells or HEK293T cells pre-transfected with plasmids encoding ACE2 and TMPRSS2. Antibody titres showed equivalence however VSV had the advantage of a shorter incubation therefore enabling a higher throughput. PVs offer a robust platform for future seroepidemiology and vaccine evaluation studies.

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