Use of Pseudotyped Viruses for the Production of Reference Materials as part of Emerging Viral Outbreak Preparedness

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International Reference Materials

- NIBSC is a World leader in the production of biological reference material and standards
  - Produces >90% WHO International standards
- Used to monitor assay function and calibrate results into International Units
- Established via a multi-lab collaborative study and assessment of candidate materials:
  - *Ability to harmonise data*
  - *Commutability*
  - *Stability*

Biological reference materials

Catalogue available: www.nibsc.org

Emerging Virus Antibody Reference Material

WHO Blueprint priority diseases

- Crimean-Congo haemorrhagic fever (CCHF)
- ✓ Ebola virus disease
- Marburg virus disease
- Lassa fever
- Middle East respiratory syndrome coronavirus (MERS-CoV)
- Severe Acute Respiratory Syndrome (SARS)
- Nipah viral disease
- Rift Valley fever (RVF)
- ✓ Zika
- Disease X

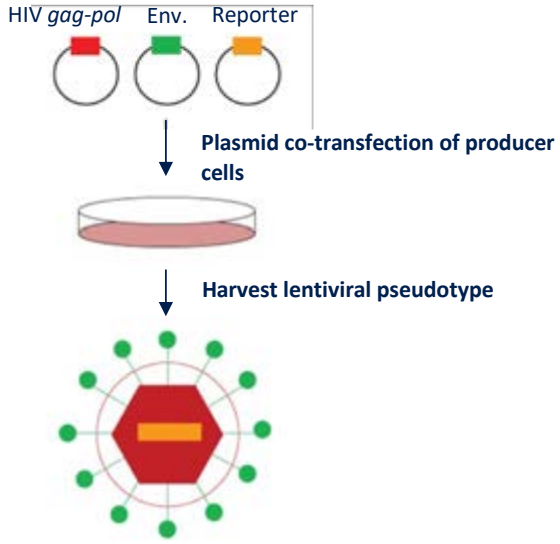
15/220	Anti-EBOV plasma, human (WHO Reference Reagent) International Reference Reagent
15/262	1st International Standard for Ebola virus (EBOV) antibodies Sierra Leone Convalescent Plasma Pool International Standard
16/344	WHO Anti-EBOV Convalescent Plasma (International Reference Panel) International Reference Preparation

Require an alternative source of antigen to characterise material for BSL4 pathogens at a low containment level…

- Assist comparison of results from treatment/vaccine efficacy clinical trials
- Preferred candidate is a pool of plasma/sera from convalescent patients

Pseudotyped Virus

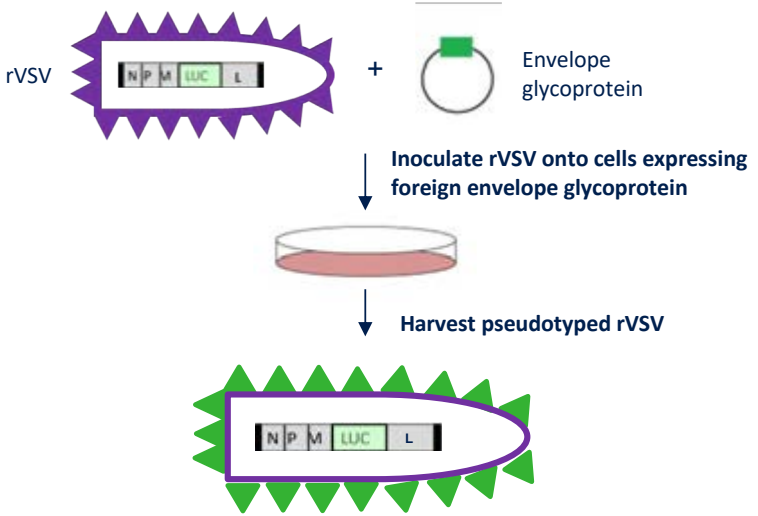
Lentiviral core



Temperton *et al.,* (2015) Encyclopedia of Life Sciences

- Spherical morphology
- Reporter gene integrated within cell genome
- Results acquired 48-72hrs post-infection

Vesiculoviral core



Whitt, (2010) Journal of Virological Methods, 169 [2]

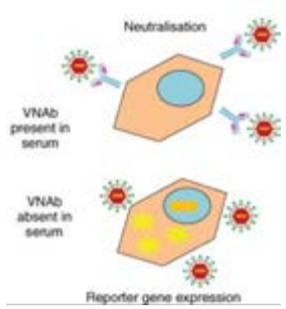
- Bullet shaped
- Transient expression of reporter gene
- Results acquired 24hrs post-infection

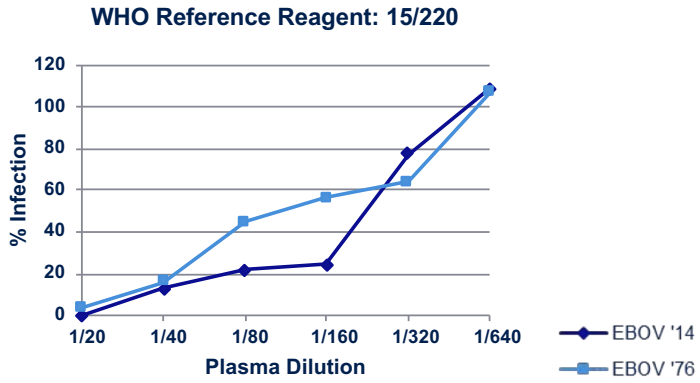
Antibody Characterisation with Pseudotypes

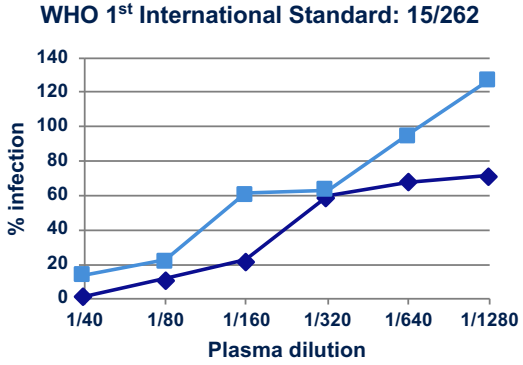
Neutralisation Assay

Titrate antibody + pseudotype onto target cells

- *Positive correlation between reporter gene signal and cells infected*



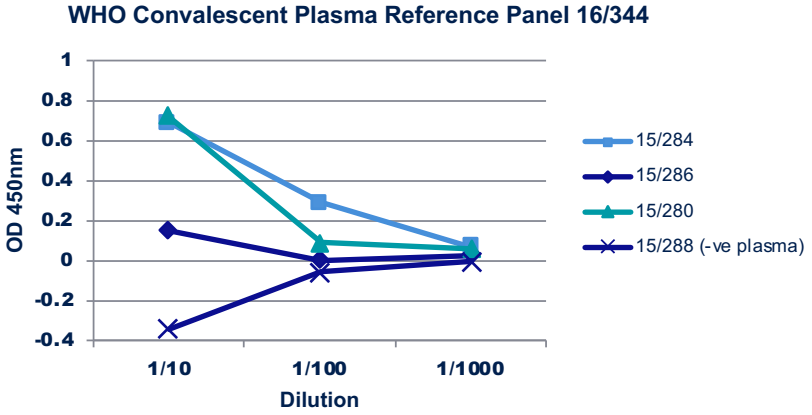




ELISA

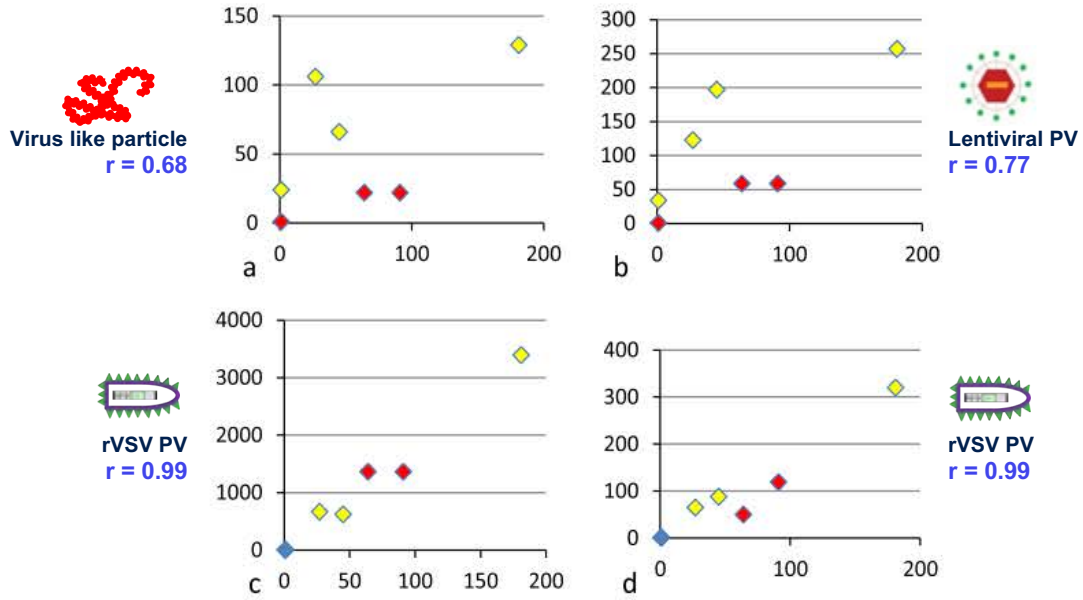
Pseudotype as the coating antigen

- *Allows conformational presentation of glycoprotein instead of monomeric recombinant protein*



Comparison of Pseudotyping Systems

- Collaborative study participants reported neutralisation titres of candidate material using other pseudotyping systems

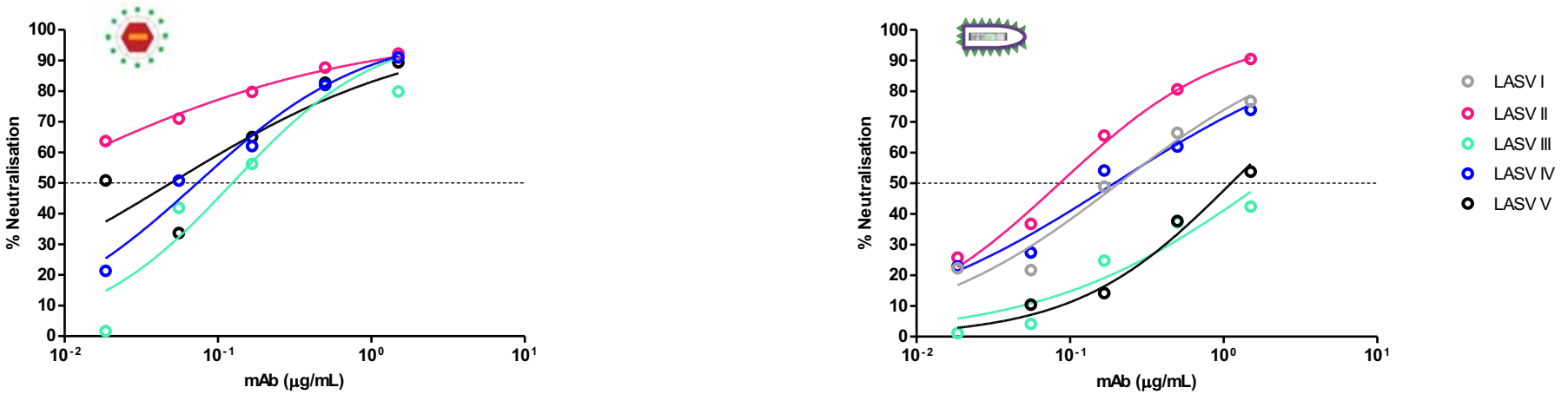


Wilkinson *et al.,* (2017) Vaccine, 35 [9]

Correlation between wildtype virus and pseudotype-based assays was better when using the VSV core system

Pseudotyping The Next Targets

- Produced Lassa pseudotyped virus with a lentiviral and vesiculoviral core
- Comparing neutralisation patterns using monoclonal antibodies



IC_{50} correlation coefficient (r) = 0.76

Conclusion

- Pseudotyped virus allowed candidate antibody material against Ebola virus to be characterised at a low containment level, supporting the establishment of International Reference Reagents
- Use of a vesiculoviral core component demonstrated better correlation with wild-type Ebola virus assays
- Requirement to evaluate the most appropriate system for each high containment virus
- Currently establishing both systems in work towards producing antibody reference material against Lassa and Nipah virus

Acknowledgements

NIBSC
Mark Hassall
Sophie Myhill
Peter Rigsby
Lindsay Stone
Dianna Wilkinson
Mark Page
Giada Mattiuzzo

University of Sussex
Edward Wright

Collaborative Study Participants

Can you donate candidate material for Lassa/Nipah/Marburg/Sudan/CCHF virus?

Would you like to participate in collaborative studies?

Please get in touch: emma.bentley@nibsc.org

Visit poster number 21.107 tomorrow!

Microbiology Society Travel Grant