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

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

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**
- HIV gag-pol
- Env.
- Reporter

Plasmid co-transfection of producer cells

Harvest lentiviral pseudotype


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

**Vesiculoviral core**
- rVSV

Inoculate rVSV onto cells expressing foreign envelope glycoprotein

Harvest pseudotyped rVSV


- 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


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

\[ \text{IC}_{50} \text{ correlation coefficient }(r) = 0.76 \]
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
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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

University of Sussex
Edward Wright

Visit poster number 21.107 tomorrow!

Collaborative Study Participants

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