CONSISE Laboratory Working Group
Summary of discussions and future plans

John Wood and Othmar Engelhardt
CONSISE 4th International Meeting, Cape Town South Africa
3-4 September 2013
Common assays for influenza serological studies

**Haemagglutination Inhibition Assay**
- Agglutination
- No agglutination

**Microneutralization Assay**
- Infection
- No infection

**MN assay read-out:**
- 3 day HA detection
- 2 day ELISA detection (WHO protocol)

In collaborative studies HI/MN assay variability between laboratories can be substantial. How can they be standardized?
MN assay comparison

Update

• Analysis of bias influencing results of first part of MN comparison study
  • Based on information available to date, no evidence for major source of bias in H1N1pdm09 study
• Extension of MN assay comparison (phase 2):
  • Included H3N2 (5 labs) and H5N1 (1 lab)
  • Results from 5 labs submitted to NIBSC (UK) for analysis
  • Ratio of titres between 3-day and 2-day assay similar in most labs
  • Preliminary data confirm the conclusions of phase 1, i.e. there are no underlying reasons that the two assays could not be comparable

Plan

• collect remaining data from additional laboratories
• Prepare report, write manuscript for publication, post consensus assay protocols and report on CONSISE website
HI assay standardization

Background
• Karen Laurie and John Wood coordinated comparison of HI protocols and tried to develop consensus assay
  – Starting point: WHO protocol

Outcome of Cape Town meeting
▪ Following discussion, consensus reached!
  ▪ Largely in agreement with protocol as in WHO Manual
  ▪ Applicable for H1N1pdm09 for subsequent collaborative study

Plan
• Revise consensus protocol and circulate to WG for approval
(International) antibody standards

Outcome
• Pathway to developing antibody standards presented and approved in principle by WG
• Possible sources of antibody
  • Human serum/plasma
    • Convalescent – help from Epi WG requested to source sera and obtain all required ethical approvals
  • Post-vaccination
  • Animal sera
• Monoclonal antibodies
• Human antibodies produced in trans-chromosomal bovines
• Discussion on status of international antibody standards
  • Agreement that formal WHO IS status not required (and too slow) in the first instance
  • Possible post hoc certification by WHO ECBS

Plan
• Revise pathway following discussion and circulate to lab WG and Steering Committee
MN and HI assay collaborative study

Outcome and Plan

• General agreement to look at lab-to-lab variability
  – Compare consensus HI protocol with local HI methods
  – Compare consensus MN protocols with local methods where local methods are different from consensus
  – Either 2-day or 3-day assay can be used
  – To be conducted for H1N1pdm09
  – Small subgroup to develop study protocol
  – NMRC (N Martin) to contribute panels of human sera

• Use the study to evaluate various sources of antibodies as potential antibody standard
  – Existing human IS
  – Monoclonal antibody
  – Pooled ferret antisera
  – Human antibodies from trans-chromosomal bovines
NI assays

Outcome

- 4 laboratories have implemented ELLA assay
- Technical issues with antigen source and some subtypes still to be resolved
- Other assays have been assessed but need more work

Plan

- Small group to plan collaborative study
- Interested labs to contact Maryna Eichelberger to obtain protocol
New serology assays

Update

• Variations of existing assays (MN) being explored, use of different cell line (CaCO2 vs MDCK) and read-out (R Wagner)
• Pseudo particle MN assay being evaluated – correlation with ‘classical’ MN; NA pseudo particles
• Modified HI assay (stabilised RBCs)
• Protein microarray
• Point-of-care test (dual path platform lateral flow)
• Luminex multiplex platform

Conclusions

▪ Most assays at early stage, need to wait for further data
  ▪ No recommendation of Lab WG at this point
▪ Pseudo particle MN assay has potential and needs further work, e.g. standardisation of particle preparation
Thank you

Laboratory Working Group
Presenters

For interesting presentations and lively discussion