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Establishing OIE H5 Reference Standard Sera



Group membership



- VLA Weybridge
- AAHL Geelong
- FLI Reims
- USDA Ames
- Hokkaido University, Japan
- IZSV, Padova
- NCFAD, Winnipeg



Aim:
to establish through a collaborative exercise amongst OFFLU partners a global reference standard serum for H5 influenza A virus

Proposed Approach



- Collect information from interested participants on their standard serum (sera) used in typing H5 influenza A virus and other serological applications
 - Strain Identity (using standard nomenclature)
 - Subtype
 - Host of origin for serum
 - HI titre with homologous antigen
- Participants to supply seven to ten ml of serum to VLA-Weybridge
- VLA will produce an anonymised panel of sera for distribution to all participants. This panel will include one reference standard antigen for comparability measurement between participants
- Each participant should test the full panel of sera against a selected range of H5 viruses held at their Institute representing contemporary viruses of wide diversity taking into account
 - Geography
 - Antigenicity
 - Genetic lineage
 - Host
- All testing done using the HI test as specified in the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

Proposed Approach



- All results should be returned to VLA (aiwrl@vla.defra.gsi.gov.uk) to collate the data and supply to all participating laboratories
- Upon distribution of results, participants will have an opportunity to comment: a teleconference will be held to make a selection of the best sera that meets the following criteria:
 - high specificity to H5 virus
 - producing broad cross reactivity with a diverse panel of virus strains
 - preferably representative of all geographical regions
- It will be necessary to specify two reference standard sera with heterologous NA subtypes in order to accommodate the problem of cross interference from anti neuraminidase antibody
- Upon selection of a reference standard serum, a candidate laboratory will be identified to undertake supply and production
- OIE reference standard serum distributed to all OIE laboratories and standardisation data collected from each participant before it's available for general release

Timeline for establishing OIE H5 reference standard sera



| Activity | Due Date |
|---|------------|
| Indicate willingness to participate and provide information on data sheet for sera that will be supplied. | 23/Dec/08 |
| Submit sera to VLA Weybridge | 31/Jan/09 |
| Panel of reagents distributed to all participants | 13/Mar/09 |
| Test results returned from all participants | 31/May/09 |
| Data distributed to all participants | 15/Sept/09 |
| Final selection of standard serum (sera) | 30/Sept/09 |
| Reference serum distributed and re-assessed by participants | 31/Dec/09 |
| Reference standard available to global community | 31/Mar/10 |

Data subject to normalisation and statistical analysis



| | Subtype | Homologous Titre | H5N1 Tk/Tky/05 | H5N1 Ck/Sco/59 | H5N2 Ost/Den/96 | H5N9 Tk/Ont/69 | H5N8 Tk/Ire/83 | H5N1 Tk/Eng/07 | H5N3 Te/Eng/06 | H5N7 Dk/Den/03 | H5N1 Tk/Tky/05 | H5N1 Tk/Eng/07 |
|---------|---------|------------------|----------------|----------------|-----------------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|
| A | H5N1 | 4096 | 256 | 512 | 256 | 256 | 4096+ | 256 | 1024 | 1024 | 256 | 256 |
| B | H5N1 | 1024 | 64 | 128 | 16 | 64 | 1024 | 64 | 256 | 64 | 64 | 64 |
| C | H5N3 | 256 | 128 | 128 | 64 | 64 | 512 | 128 | 256 | 256 | 128 | 64 |
| D | H5N2 | 1024 | 128 | 128 | 128 | 128 | 1024 | 128 | 256 | 512 | 256 | 64 |
| E | H5N1 | 256 | 4 | 16 | <2 | 8 | 16 | 4 | 16 | 16 | 8 | 4 |
| F | H5N1 | 320-640 | 128 | 64 | 32 | 64 | 128 | 128 | 32 | 128 | 256 | 128 |
| G | H5N2 | 512 | 128 | 256 | 128 | 128 | 2048 | 128 | 256 | 512 | 256 | 128 |
| H | H5N1 | 1280 | 256 | 32 | 4 | 64 | 128 | 128 | 32 | 128 | 512 | 256 |
| I | H5N1 | 640-1280 | 64 | 64 | 16 | 32 | 256 | 64 | 128 | 128 | 128 | 64 |
| J | H5N1 | 128 | <2 | <2 | <2 | <2 | <2 | <2 | <2 | <2 | <2 | <2 |
| K | H5N1 | 256 | 32 | 64 | 256 | 32 | 512 | 64 | 128 | 256 | 64 | 32 |
| L | H5N1 | 256 | 64 | 256 | 64 | 64 | 512 | 64 | 256 | 256 | 128 | 64 |
| M | H5N9 | 32 | 32 | 32 | 128 | 128 | 256 | 64 | 128 | 256 | 64 | 32 |
| SPF Neg | | <2 | <2 | <2 | <2 | <2 | <2 | <2 | <2 | <2 | <2 | <2 |

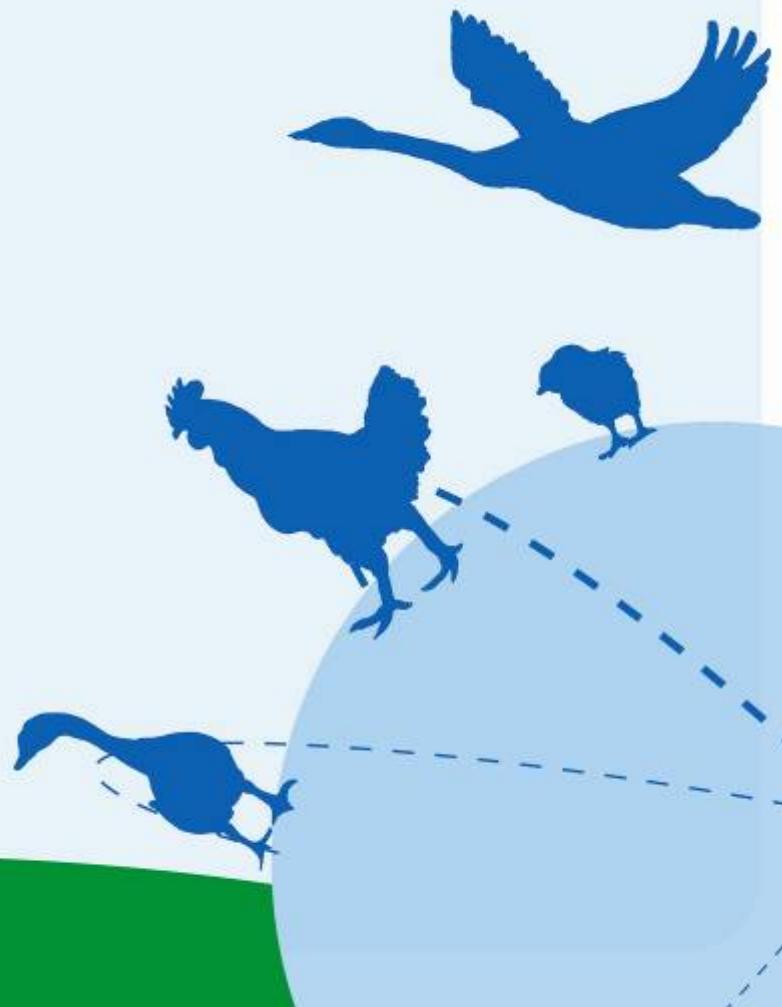
Evaluation after „normalization“:

| | Lab A | Lab B... | Σ | % | X/ref. Antigen | WEY H7N1 | WEY H7N7 |
|----------------|--------------|-----------------|-----------|----------|-----------------------|-----------------|-----------------|
| Serum A | 0 | 3 | 17 | 22,37 | 8,7 | 7 | <1 |
| Serum B | 1 | 1 | 9 | 11,8 | 6,4 | <1 | <1 |
| Serum C | 0 | 7 | 12 | 15,8 | 7,9 | <1 | <1 |
| Serum D | 0 | 2 | 12 | 15,8 | 8,6 | <1 | <1 |
| Serum E | 0 | 0 | 6 | 7,9 | 3,7 | 9 | <1 |
| Serum F | 0 | 9 | 24 | 31,6 | 8,4 | 7 | <1 |
| Serum G | 0 | 3 | 10 | 13,2 | 7,1 | 8 | <1 |
| Serum H | 2 | 11 | 49 | 64,5 | 7,9 | 8 | <1 |
| Serum I | 0 | 3 | 8 | 10,5 | 7,9 | 9 | <1 |
| Serum J | | | | 0,3 | | | |
| Serum K | 0 | 2 | 9 | 11,8 | 7,1 | <1 | <1 |
| Serum L | 0 | 3 | 10 | 13,2 | 7,7 | 10 | <1 |
| Serum M | 1 | 2 | 9 | 11,8 | 6,4 | <1 | <1 |

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Thank you for your attention



OFFLU secretariat

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