Influenza Reagent
Influenza anti-A/Victoria/4897/2022-like (H1N1) HA serum
NIBSC code: 23/100
Instructions for use
(Version 1.0, Dated 24/04/2023)

1. INTENDED USE
Influenza antisera reagent 23/100 is prepared for single radial diffusion assay of H1N1 A/Victoria/4897/2022-like antigens using an appropriate NIBSC antigen reagent.

The antiserum was prepared in sheep SH812, SH813, SH814 and SH815 using the purified HA of a A/Victoria/4897/2022-like virus. The HA antigens were extracted from purified virus by treatment with bromelain and purified by sedimentation on sucrose gradients (Brand, CN and Skehel, JJ, Nature, New Biology, 1972, 238, 145-147).

2. CAUTION
This preparation is not for administration to humans or animals in the human food chain.

The material is not of human or bovine origin.

As with all materials of biological origin, this preparation should be regarded as potentially hazardous to health. It should be used and discarded according to your own laboratory’s safety procedures. Such safety procedures should include the wearing of protective gloves and avoiding the generation of aerosols. Care should be exercised in opening ampoules or vials, to avoid cuts.

3. UNITAGE
No unitage is assigned to this material.

4. CONTENTS
Country of origin of biological material: United Kingdom.

The immunization schedule for sheep SH812, SH813, SH814 and SH815 was as follows: one dose of approximately 100 µg of A/Norway/31694/2022 virus HA with Freund’s Complete Adjuvant (FCA) was given intramuscularly, followed two weeks later with a 50 microgram dose of A/Victoria/4897 (IVR-238) including Freund’s Incomplete Adjuvant (FIA). Three further 50 microgram doses of A/Victoria/4897/2022 (IVR-238) HA including FIA were given over a week. Six weeks after the initial immunization, serum was collected and sodium azide (0.05% w/v) added.

The antiserum was then treated by an APHIS approved method for the inactivation of FMDV.

The antisera obtained from sheep SH812, SH813, SH814 and SH815 were pooled, diluted 1:6 with PBS buffer containing sodium azide (0.05% w/v), and filled into vials in 2ml volumes.

5. STORAGE
+2-8°C

However, if it is intended to store the reagent for long periods i.e. >2 years, they may be stored at -20°C. The antiserum can be frozen and thawed without any adverse impact on its use in the SRD assay.

6. DIRECTIONS FOR OPENING
Vials have a screw cap; an internal stopper may also be present. The cap should be removed by turning anti-clockwise. Care should be taken to prevent loss of the contents. Please note: If a stopper is present on removal of the cap, the stopper should remain in the vial or be removed with the cap.

7. USE OF MATERIAL
For the assay of antigens containing 20-50 micrograms of HA activity in 1ml, approximately 10-20 µl of the undiluted reagent should be added to 1ml agarose. It may be necessary to change the antiserum concentrations depending on the A/Victoria/4897/2022-like antigen standard used or according to local laboratory conditions.

Antiserum Reagent 23/100 should be used according to the method described by Wood, JM, Schild, GC, Newman, RW and Seagroatt, VA. Journal of Biological Standardisation, 1977, 5, 2.

8. STABILITY
It is the policy of WHO not to assign an expiry date to their international reference materials. They remain valid with the assigned potency and status until withdrawn or amended.

Reference materials are held at NIBSC within assured, temperature-controlled storage facilities. Reference Materials should be stored on receipt as indicated on the label. Once reconstituted, diluted or aliquotted, users should determine the stability of the material according to their own method of preparation, storage and use.

NIBSC follows the policy of WHO with respect to its reference materials.

Users who have data supporting any deterioration in the characteristics of any reference preparation are encouraged to contact NIBSC.

9. REFERENCES
None

10. ACKNOWLEDGEMENTS
None

11. FURTHER INFORMATION
Further information can be obtained as follows;
This material: enquiries@nibsc.org
WHO Biological Standards:
http://www.who.int/biologicals/en/
JCTLM Higher order reference materials:
http://www.bipm.org/en/committees/jcctlm/
Derivation of International Units:
http://www.nibsc.org/standardisation/international_standards.aspx
Ordering standards from NIBSC:
http://www.nibsc.org/products/ordering.aspx
NIBSC Terms & Conditions:
http://www.nibsc.org/terms_and_conditions.aspx

12. CUSTOMER FEEDBACK
Customers are encouraged to provide feedback on the suitability or use of the material provided or other aspects of our service. Please send any comments to enquiries@nibsc.org
13. CITATION
In all publications, including data sheets, in which this material is referenced, it is important that the preparation's title, its status, the NIBSC code number, and the name and address of NIBSC are cited and cited correctly.

14. MATERIAL SAFETY SHEET
Classification in accordance with Directive 2000/54/EC, Regulation (EC) No 1272/2008: 1

<table>
<thead>
<tr>
<th>Physical and Chemical properties</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Physical appearance: Liquid</td>
<td>Corrosive: No</td>
</tr>
<tr>
<td>Stable:</td>
<td>Oxidising: No</td>
</tr>
<tr>
<td>Yes</td>
<td></td>
</tr>
<tr>
<td>Hygroscopic:</td>
<td>Irritant: No</td>
</tr>
<tr>
<td>No</td>
<td>Handling: See caution, Section 2</td>
</tr>
<tr>
<td>Flammable:</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Other (specify):</td>
<td></td>
</tr>
<tr>
<td>Contains Sheep Serum and Sodium Azide (0.05% w/v)</td>
<td></td>
</tr>
</tbody>
</table>

Toxicological properties
- Effects of inhalation: Not established, avoid inhalation
- Effects of ingestion: Not established, avoid ingestion
- Effects of skin absorption: Not established, avoid contact with skin

Suggested First Aid
- Inhalation: Seek medical advice
- Ingestion: Seek medical advice
- Contact with eyes: Wash with copious amounts of water. Seek medical advice
- Contact with skin: Wash thoroughly with water.

Action on Spillage and Method of Disposal
Spillage of ampoule contents should be taken up with absorbent material wetted with an appropriate disinfectant. Rinse area with an appropriate disinfectant followed by water. Absorbent materials used to treat spillage should be treated as biological waste.

15. LIABILITY AND LOSS
In the event that this document is translated into another language, the English language version shall prevail in the event of any inconsistencies between the documents.

Unless expressly stated otherwise by NIBSC, NIBSC’s Standard Terms and Conditions for the Supply of Materials (available at http://www.nibsc.org/About_Us/Terms_and_Conditions.aspx or upon request by the Recipient) (“Conditions”) apply to the exclusion of all other terms and are hereby incorporated into this document by reference. The Recipient’s attention is drawn in particular to the provisions of clause 11 of the Conditions.

16. INFORMATION FOR CUSTOMS USE ONLY
| Country of origin for customs purposes*: United Kingdom |
| Toxicity Statement: Non toxic                          |
| Veterinary certificate or other statement if applicable. |
| Attached: Yes SH812 SH813 SH814 SH815 |

Country of origin for customs purposes*: Defined as the country where the goods have been produced and/or sufficiently processed to be classed as originating from the country of supply, for example a change of state such as freeze-drying.

Net weight: 2g

Toxicity Statement: Non toxic

Veterinary certificate or other statement if applicable.

Attached: Yes SH812 SH813 SH814 SH815
VETERINARY CERTIFICATE OF ANIMAL HEALTH

This is to certify that I have examined a Sheep with ear tag number UK 014795407863 [Virology no SF3812], which has been used in the production of blood antiseraum between 3rd March 2023 and 12th April 2023. Both the ear tag number and the animals’ record show that it is of UK origin.

This animal was a breeding Ewe which became surplus to requirements. In my opinion at the time of clinical examination, the ewe was in good health and showed no clinical signs of infectious disease.

Alex McSloy MA VetMB DipACVIM PhD MRCVS
Named Veterinary Surgeon

Date signed: 14 April 2023

Alex McSloy
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WHO International Laboratory for Biological Standards,
UK Official Medicines Control Laboratory

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VETERINARY CERTIFICATE OF ANIMAL HEALTH

This is to certify that I have examined a Sheep with ear tag number UK 014795405602 [Virology no 3F81.3], which has been used in the production of blood antiserum between 3rd March 2023 and 12th April 2023. Both the ear tag number and the animal's record show that it is of UK origin.

This animal was a breeding Ewe which became surplus to requirements. In my opinion at the time of clinical examination, the ewe was in good health and showed no clinical signs of infectious disease.

Alex McSloy
MA VetMB DipACVIM PhD MRCVS
Named Veterinary Surgeon

Date signed: 14 April 2023

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Tel: +44 (0)1707 666333, E-mail: amcslay@rvc.ac.uk
VETERINARY CERTIFICATE OF ANIMAL HEALTH

This is to certify that I have examined a Sheep with ear tag number UK 024126907226 [Virology no 573814], which has been used in the production of blood antiserum between 3rd March 2023 and 12th April 2023. Both the ear tag number and the animals' record show that it is of UK origin.

This animal was a breeding Ewe which became surplus to requirements. In my opinion at the time of clinical examination, the ewe was in good health and showed no clinical signs of infectious disease.

Alex McSloy
MA VetMB DipACVIM PhD MRCVS
Named Veterinary Surgeon

Date signed: 14 April 2023
VETERINARY CERTIFICATE OF ANIMAL HEALTH

This is to certify that I have examined a Sheep with ear tag number UK 024126907013 [Virology no. SF3815], which has been used in the production of blood antiserum between 3rd March 2023 and 12th April 2023. Both the ear tag number and the animals' record show that it is of UK origin.

This animal was a breeding Ewe which became surplus to requirements. In my opinion at the time of clinical examination, the ewe was in good health and showed no clinical signs of infectious disease.

Alex McSloy
MA VetMB DipACVIM PhD MRCVS
Named Veterinary Surgeon

Date signed: 14 April 2023