Influenza Reagent
Influenza anti- A/Chick/Scotland/59 (H5N1) Serum
NIBSC code: 00/542
Instructions for use
(Version 2.0, Dated 29/03/2008)

1. INTENDED USE
Influenza antisera reagent 00/542 is prepared in sheep for serological assay of A/Chick/Scotland/59-like antigens. For single-radial-diffusion assay (SRD) an appropriate NIBSC antigen reagent should be included in each test.

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 reagent was prepared in a sheep (SH4) to the purified haemagglutinin (HA) of A/Chick/Scotland/59 virus. The HA was extracted from purified virus by treatment with bromelain and purification on sucrose gradients (Brand, C N and Skehel, J J. Nature New Biology, 1972, 238, 145-147). One dose of approximately 50µg HA with Freund's complete adjuvant (FCA) was given intramuscularly, followed two weeks later with a 25µg dose with FCA and two further 25µg doses with FCA at weekly intervals.
Six weeks after the initial immunization, serum was collected, diluted 1/7.5 with phosphate buffered saline containing sodium azide (0.05% w/v) and filled into vials in 2ml volumes. The mean weight of 7 vials test weighed was 2.0008g with a coefficient of variation of 0.27%.

5. STORAGE
+2-8°C
Please note: because of the inherent stability of lyophilized material, NIBSC may ship these materials at ambient temperature.

6. DIRECTIONS FOR OPENING
Vials have a ‘flip-up’ circular cap. Either on the cap or the collar of the vial, there is an indication of the point at which to lever off the cap. This exposes an area of the stopper through which reconstitution and withdrawal of the preparation can be made using a hypodermic needle and syringe. If use of a pipette is preferred, then fully remove the metal collar using, for example, forceps, taking care to avoid cuts by wearing appropriate gloves. Remove the stopper for access. Care should be taken to prevent loss of the contents.

7. USE OF MATERIAL
No attempt should be made to weigh out any portion of the material
Antiserum reagent 00/542 is broadly reactive in haemagglutination-inhibition tests with a variety of influenza A (H5) viruses and has no detectable HI activity with influenza A (H3N2), (H1N1) and b viruses (see appendix). Before use in HI tests, non-specific inhibitors should be removed from antiserum reagent 00/542 by normal procedures.

Antiserum reagent 00/542 may be used for SRD assay of influenza H5 HA antigens. For the assay of A/Duck/Singapore-Q/F119-3/97 (H5N3) (Dk/Sing) HA antigens containing 20-50µg of HA activity in 1ml, 5-10µl of undiluted Reagent should be added to 1ml agarose. For the assay of A/Hong Kong/489/97 (H5N1) (HK/489) HA antigens containing 20-50µg of HA activity in 1ml, 10-20µl of undiluted reagent should be added to 1ml agarose. Antigens of lower concentration (5-20µg HA/ml) are assayed by adding 2.5-5 or 5-10µl (Dk/Sing or HK/489 respectively) of the reagent to 1ml agarose. It may be necessary to change these antiserum concentrations according to local laboratory conditions.

Antiserum reagent 99/834 should be used according to the method described by Wood JM, Schild GC, Newman RW and Seagroatt VA. Journal of Biological Standardisation, 1977, 5, 237-247.

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 aliquoted, 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. Reference materials are held at NIBSC within assured, temperature-controlled storage facilities. Reference Materials should be stored on receipt as indicated on the label.

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/jc/jctlm/
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: Not applicable or not classified

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

Toxicological properties

Effects of inhalation: Avoid inhalation
Effects of ingestion: Avoid ingestion
Effects of skin absorption: 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
* 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: No