Influenza Reagent
Influenza anti B/Guangdong/120/2000 Serum
NIBSC code: 01/450
Instructions for use
(Version 4.0, Dated 05/03/2008)

1. INTENDED USE
Influenza reagent 01/450 is prepared in sheep for the single radial diffusion assay of B/Guangdong/120/2000 antigens. Assays should contain an appropriate NIBSC antigen reagent.

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 antiserum was prepared in SHEEP (SH412/413) to the purified HA of B/Guangdong/120/2000 virus. The HA antigen was extracted from purified virus by treatment with bromelain and purified by sedimentation on sucrose gradients (Brand, C N and Skehel, JJ, Nature, New Biology, 1972, 238, 145-147). One dose of approximately 50 micrograms of HA with Freund's complete adjuvant (FCA) was given intramuscularly into each of two sheep, followed two weeks later with a 10 microgram dose in the presence of Freund's incomplete adjuvant (FIA) and two further doses at weekly intervals.

Six weeks after the initial immunization, each serum (SH412 and SH413) was collected, sodium azide (0.05% w/v) was added and the sera were exposed to pH5 for a two hour period in order to comply with EC foot and mouth disease decisions (see section 4). The sera were pooled as reagent SH412/413, diluted 1:4 with PBS buffer containing sodium azide (0.05% w/v) and filled into vials in 2ml volumes. The mean weight of 43 vials test weighed was 2.047g with a coefficient of variation of 0.49%.

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
For the assay of antigens containing 20-50 micrograms of HA activity in 1ml reagent, approximately 11µl of Reagent 01/450 should be added to 1ml agarose. Antigens of lower concentration (5-20 micrograms HA/ml) are assayed by adding 5.5µl of the diluted reagent to 1ml agarose. It may be necessary to change these antiserum concentrations according to local laboratory conditions.

Antiserum Reagent 01/450 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.

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

Reference Materials should be stored on receipt as indicated on the label.

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

9. REFERENCES

10. ACKNOWLEDGEMENTS

11. FURTHER INFORMATION
Further information can be obtained as follows:
This material: enquiries@nibsc.org
WHO Biological Standards: http://www.who.int/biologicals/en/
Derivation of International Units: http://www.nibsc.org/standardisation/international_standards.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>
</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.

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### 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**: Yes SH412-413
Veterinary Certificate

Reagent 01/450

This is to certify that sheep numbers 2476 (SH412) and 2499 (SH413) were used for the production of blood antiserum between April and June 2001 to prepare reagent 01/450.

The sheep were ewes on the Royal Veterinary College Boltons Park Farm, Brokmans Park, Hertfordshire, UK that were surplus to breeding requirements, in good health and showed no signs of clinical disease.

From April to May 2001, there have been no cases of foot and mouth disease within a 20 mile radius of the farm.

Reagent 01/450 has been inactivated by exposure to pH5 for 2 hours and has been tested negative for foot and mouth disease virus by the Central Public Health Laboratory, Colindale, UK. The reagent thus complies with the European Commission Decision of 8 March 2001 (2001/190/EC), concerning protection measures with regard to foot and mouth disease in the UK.

Reagent 01/450 is only for laboratory use as an in vitro diagnostic reagent.

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Bvet Med MRCVS
Named Veterinary Surgeon

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* Information from http://www.maff.gov.uk