

Influenza Reagent
Influenza anti A/Wisconsin/67/2005 (H3N2)-like HA Serum (sheep)
NIBSC code: 05/236
Instructions for use
(Version 4.0, Dated 30/07/2015)

#### 1. INTENDED USE

Influenza antiserum reagent 05/236 is prepared in sheep for single radial diffusion assay of A/Wisconsin/67/2005-like antigens. An appropriate NIBSC reagent should be included in each assay

#### 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 reagent was prepared in sheep 459, 460, 461 and 462 to the purified HA of A/Hiroshima/52/2005, IVR-142 (a reassortant with HA and NA from A/Hiroshima/52/2005) and NYMCX-161 (a reassortant with HA and NA from A/Wisconsin/67/2005). The HA antigen was 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.

The immunization schedule for sheep 459 and 460 was as follows: One dose of approximately 25-50 micrograms of A/Hiroshima/52/2005 HA with Freund's Complete Adjuvant (FCA) was given intramuscularly, followed two weeks later with a 10-20 microgram dose of IVR-142 including Freund's Incomplete Adjuvant (FIA), two further 20 microgram doses of IVR-142 including FIA were given at weekly intervals. Five weeks after the initial immunization, serum was collected and sodium azide (0.05% w/v) was added.

The immunization schedule for sheep 461 and 462 was as follows: One dose of approximately 25-50 micrograms of A/Hiroshima/52/2005 HA with Freund's Complete Adjuvant (FCA) was given intramuscularly, followed two weeks later with a 10-20 microgram dose of NYMCX-161 including Freund's Incomplete Adjuvant (FIA), two further 20 microgram doses of NYMCX-161 including FIA were given at weekly intervals. Six weeks after the initial immunization, serum was collected and sodium azide (0.05% w/v) was added.

The sera were pooled, and diluted 1:5 with PBS buffer containing sodium azide (0.05% w/v) and filled into vials in 2ml volumes. The mean weight of 141 vials tested was 2.05g with a coefficient of variation of 0.99 %.

# 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

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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

For the assay of antigens containing 20-50 micrograms of HA activity in 1ml, approximately 13 $\mu$ l of the undiluted Reagent should be added to 1ml agarose. It may be necessary to change the antiserum concentrations according to local laboratory conditions.

Antiserum Reagent 05/236 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

Reference materials are held at NIBSC within assured, temperature-controlled storage facilities. 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

N/A

#### 10. ACKNOWLEDGEMENTS

N/A

#### 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

Physical and Chemical properties			
Physical	Corrosive:	No	
appearance: Liquid			
Stable:	Oxidising:	No	
Yes			





Hygroscopic: No	Irritant:	No			
Flammable:	Handling:	See caution, Section 2			
No					
Other (specify): Contains Sheep Serum and Sodium Azide (0.05%					
w/v)Please complete					
Toxicological properties					
Effects of inhalation:	Not e	stablished, avoid inhalation			
Effects of ingestion:	Not e	Not established, avoid ingestion			
Effects of skin absorp	otion: Not e	stablished, avoid contact with skin			
Suggested First Aid					
Inhalation:	Seek medical advice				
Ingestion:	Seek medical advice				
	Wash with copious amounts of water. Seek medical advice				
-	Wash thoroughly with water.				
Contact With Skin.	rvasii tilolougii	iy willi 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.	Diological 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: Not established

Veterinary certificate or other statement if applicable.

Attached: Yes SH459 SH460 SH461 SH462

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**Veterinary Certificate** 

COPY

This is to certify that Sheep no. 5122 [Virology No. SH 459] was used for the production of blood antiserum between 3<sup>rd</sup> March 2006 and 5<sup>th</sup> April 2006.

This sheep was a ewe that was surplus to breeding requirements, in overt good health, and showed no signs of clinical disease.

The ear tag identifying the animal indicated that it was of UK origin.

R or should

R.M. Hull BVSc, PhD, MRCVS Named Veterinary Surgeon 7 -April-2006



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**Veterinary Certificate** 

COPY

This is to certify that Sheep no. 5124 [Virology No. SH 460] was used for the production of blood antiserum between  $3^{rd}$  March 2006 and  $5^{th}$  April 2006.

This sheep was a ewe that was surplus to breeding requirements, in overt good health, and showed no signs of clinical disease.

The ear tag identifying the animal indicated that it was of UK origin.

R or Unell

R.M. Hull

7 -April-2006

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# **Veterinary Certificate**

COPY

This is to certify that Sheep no. 5113 [Virology No. SH 461] was used for the production of blood antišerum between 3<sup>rd</sup> March 2006 and 11<sup>th</sup> April 2006.

This sheep was a ewe that was surplus to breeding requirements, in overt good health, and showed no signs of clinical disease.

The ear tag identifying the animal indicated that it was of UK origin.

Ry Hull

R.M. Hull /8 -April-2006 BVSc, PhD, MRCVS Named Veterinary Surgeon



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**Veterinary Certificate** 

COPY

This is to certify that Sheep no. 5116 [Virology No. SH 462] was used for the production of blood antiserum between 3<sup>rd</sup> March 2006 and 11<sup>th</sup> April 2006.

This sheep was a ewe that was surplus to breeding requirements, in overt good health, and showed no signs of clinical disease.

The ear tag identifying the animal indicated that it was of UK origin.

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