Influenza Reagent
Influenza virus infectious CNIC-1902
NIBSC code: 19/148
Instructions for use
(Version 2.0, Dated 15/07/2019)

1. INTENDED USE
Reagent 19/148 is prepared from CNIC-1902 (A/beijing/2019-15554/2018 (H3N2) x A/PR8/34 ) which was processed for freeze drying in 250μl volumes as described by Campbell, PJ. Journal of Biological Standardisation, 1974, 2, 249-267. The derivation and known passage history of CNIC-1902 is attached.

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 attached to this material

4. CONTENTS
Country of origin of biological material: United Kingdom.
Each ampoule contains 250μl (nominal) of infectious influenza virus as allantoic fluid from SPF embryonated hen’s eggs.

5. STORAGE
Store in the dark at -20°C or below

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
Reconstitute the contents of one ampoule of reagent with 250μl of sterile distilled water. Leave for a minimum of 5 minutes before use to allow for complete solution of freeze-dried material. A range of dilutions (e.g. 10⁻³ to 10⁻⁷) should be made in a suitable medium for initial cultivation.

8. STABILITY
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
NA

10. ACKNOWLEDGEMENTS
NA

11. FURTHER INFORMATION
Further information can be obtained as follows:

WHO Biological Standards:
http://www.who.int/biological/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

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:</td>
</tr>
<tr>
<td>Corrosive: No</td>
</tr>
<tr>
<td>Stable: Yes</td>
</tr>
<tr>
<td>Oxidising: No</td>
</tr>
<tr>
<td>Hygroscopic: No</td>
</tr>
<tr>
<td>Irritant: No</td>
</tr>
<tr>
<td>Flammable: No</td>
</tr>
<tr>
<td>Handling: See caution, Section 2</td>
</tr>
<tr>
<td>Other (specify): Live influenza virus</td>
</tr>
</tbody>
</table>

Toxicological properties

Effects of inhalation: Likelihood of influenza virus infection
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 contents should be taken up with absorbent material wetted with an appropriate virucidal agent. Rinse area with an appropriate virucidal agent followed by water.

Absorbent materials used to treat spillage should be treated as biologically hazardous 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: 0.25g per ampoule |
| Toxicity Statement: Non-toxic |
| Veterinary certificate or other statement if applicable. |
| Attached: No |

Passage history of CNIC-1902

<table>
<thead>
<tr>
<th>Passage</th>
<th>Lot</th>
<th>Laboratory</th>
</tr>
</thead>
<tbody>
<tr>
<td>E1-E2</td>
<td>Unknown</td>
<td>Unknown</td>
</tr>
<tr>
<td>E3-E9</td>
<td>Unknown</td>
<td>CNIC, China</td>
</tr>
<tr>
<td>E10</td>
<td>CNIC-1902 (44320)</td>
<td>NIBSC, Hertfordshire, UK</td>
</tr>
</tbody>
</table>

Sterility: No visible contamination was detected in a variety of media (tryptose soya broth, thioglycolate broth, Sabouraud’s broth and blood agar plates) after 14 days incubation. The HA and NA sequence of this virus is available at GISAID with the accession number EPI_ISL_363643.
Derivation of CNIC-1902

A/Kansas/14/2017 (H3N2)-like High Growth Reassortant

Strain: A/beijing/2019-15554/2018

Passage undertaken at CNIC

1\textsuperscript{st} Mixed Infection: A/beijing/2019-15554/2018 (10\textsuperscript{3}) X PR8 (10\textsuperscript{5})

HA Titer: 1024

\rightarrow

2\textsuperscript{nd} Antiserum passage: Inoculum 10\textsuperscript{3} with PR8 HA & NA antiserum

HA Titer: 2048

\rightarrow

3\textsuperscript{rd} Antiserum passage: Inoculum 10\textsuperscript{3} with PR8 antiserum

HA Titer: 2048

\rightarrow

4\textsuperscript{th} Antiserum passage: Inoculum 10\textsuperscript{3} with PR8 antiserum

HA Titer: 2048

\rightarrow

5\textsuperscript{th} passage: Inoculum 10\textsuperscript{4}

HA Titer: 512

\rightarrow

6\textsuperscript{th} passage: Inoculum 10\textsuperscript{9}

HA Titer: 1024
7th passage: Inoculum $10^5$

HA Titer: 1024

Total number of passages since mixed infection=E7

SPF eggs were used for all passages.