



**Influenza Reagent**  
**Influenza Virus Infectious CNIC-2206**  
**(A/Guizhou-Liuzhite/326/2022) (H3N2)**  
**NIBSC code: 22/180**  
**Instructions for use**  
**(Version 1.0, Dated 14/10/2022)**

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**1. INTENDED USE**

Reagent 22/180 is prepared from CNIC-2206 (H3N2), a reassortant of A/Guizhou-Liuzhite/326/2022 and A/PR/8/34, which was processed in 250µl volumes as liquid stock. The known passage history of CNIC-2206 is attached.

**2. CAUTION**

**The material is not of human or bovine origin. This preparation is not for administration to humans or animals**

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.  
Each vial contains 250µl (nominal) of infectious influenza virus as allantoic fluid from SPF embryonated hen's eggs.

**5. STORAGE**

Store in the dark at -70°C or below.  
Material type: Liquid – will be shipped according to the storage and shipping conditions of the product

**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**

Ready to use.

**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**

NA

**10. ACKNOWLEDGEMENTS**

NA

**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](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](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

<b>Physical and Chemical properties</b>	
Physical appearance: Clear liquid	Corrosive: No
Stable: Yes	Oxidising: No
Hygroscopic: No	Irritant: No
Flammable: No	Handling: See caution, Section 2
Other Live influenza virus (specify):	
<b>Toxicological properties</b>	
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
<b>Suggested First Aid</b>	
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.
<b>Action on Spillage and Method of Disposal</b>	
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](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**

<b>Country of origin for customs purposes*:</b> 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.
<b>Net weight:</b> 0.25g per vial.
<b>Toxicity Statement:</b> Non-toxic
<b>Veterinary certificate or other statement</b> if applicable. <b>Attached:</b> No

**Passage history of CNIC-2206 (H3N2)**

<b>Cumulative number of passages</b>	<b>Passage numbers at each stage</b>	<b>Lot</b>	<b>Laboratory</b>
E9	E9	Unknown	Unknown
E16	E9/E7	20220815A	CNIC, China
E17	E9/E7/E1	47180	NIBSC, UK

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\_15157782.



**CNIC**

09/29/2022

**CNIC-2206 HIGH GROWTH REASSORTANT REPORT**

High Growth Reassortant	Parental Wild Type virus	Conclusion
CNIC-2006 (Lot:20220815A)	A/Guizhou-Liuzhite/326/2022	TWO-WAY PASS

● **Two-way HI test**

The high growth reassortant (HGR) virus was antigenically characterized by a “two-way” Hemagglutination Inhibition (HI) test. Ferret antisera raised against the wild type parental virus A/Guizhou-Liuzhite/326/2022 well inhibited the HGR CNIC-2206 (within two-fold reduction of HI titers compared with the homologous virus). Similarly, ferret antiserum raised against the HGR CNIC-2206 well inhibited the parental virus. Therefore, CNIC-2206 is antigenically similar to A/Guizhou-Liuzhite/326/2022 and passes the two-way test.

**HEMAGGLUTINATION INHIBITION ASSAY OF INFLUENZA A VIRUSES**

TESTED 09/28/2022

ANTIGENS	REFERENCE FERRET ANTISERA		
	A/Guizhou-Liuzhite/326/2022	CNIC-2206	PASSAGE HISTORY
A/Guizhou-Liuzhite/326/2022	1280	320	E9
CNIC-2206	640	640	E9/E7

● **GENETIC CHARACTERIZATION**

Whole genome sequencing of HGR CNIC-2206 was performed. Comparing with wild type parental virus A/Guizhou-Liuzhite/326/2022 (E9), the HA gene of CNIC-2206 acquired amino acids change at positions D225G and P227S. There was no amino acid change in the NA.

For the internal genes, CNIC-2206 has M, NP, NS, PA, PB1 and PB2 genes from A/PR/8/34, making it a 6:2 reassortant.



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Derivation of CNIC-2206 High Growth Reassortant

Strain: A/Guizhou-Liuzhite/326/2022

Passage undertaken at CNIC

1<sup>st</sup> Mixed Infection: A/Guizhou-Liuzhite/326/2022 ( $10^{-1}$ ) X A/PR/8/34 ( $10^{-6}$ )

HA Titer: 2048

2<sup>nd</sup> Antiserum passage: Inoculum  $10^{-3}$  with A/PR/8/34 & NA antiserum

HA Titer: 64

3<sup>rd</sup> Antiserum passage: Inoculum  $10^{-1}$  with A/PR/8/34 & NA antiserum

HA Titer: 64

4<sup>th</sup> Antiserum passage: Inoculum  $10^{-2}$  with A/PR/8/34 & NA antiserum

HA Titer: 8

5<sup>th</sup> passage: Inoculum  $10^{-7}$

HA Titer: 512

6<sup>th</sup> passage: Inoculum  $10^{-8}$

HA Titer: 8

7<sup>th</sup> passage: Inoculum  $10^{-5}$

HA Titer: 32

Total number of passages since mixed infection=E7

SPF eggs were used for all passages.