



**Influenza Reagent  
Influenza virus infectious NIB-91  
NIBSC code: 14/322  
Instructions for use  
(Version 0.0, Dated 21/04/2015)**

**1. INTENDED USE**

Reagent 14/322 is prepared from NIB-91 which was processed for freeze drying in 250µl volumes as described by Campbell, PJ, Journal of Biological Standardisation, 1974, 2,249-267. The known passage history of NIB-91 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 assigned 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**

DIN ampoules have an 'easy-open' coloured stress point, where the narrow ampoule stem joins the wider ampoule body. Various types of ampoule breaker are available commercially. To open the ampoule, tap the ampoule gently to collect material at the bottom (labelled) end and follow manufactures instructions provided with the ampoule breaker.

**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<sup>-3</sup> to 10<sup>-5</sup>) 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:  
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

Physical and Chemical properties	
Physical appearance: white powder	Corrosive: No
Stable: Yes	Oxidising: No
Hygroscopic: No	Irritant: No
Flammable: No	Handling: See caution, Section 2
Other (specify): Live influenza virus	
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](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> NA
<b>Toxicity Statement:</b> Non-toxic
<b>Veterinary certificate or other statement</b> if applicable.
<b>Attached:</b> No

**Passage history of NIB-91 (Post mixed infection)**

Passage	Lot	Laboratory
E1-E5		NIBSC, Hertfordshire, UK
E6	40250	NIBSC, Hertfordshire, UK

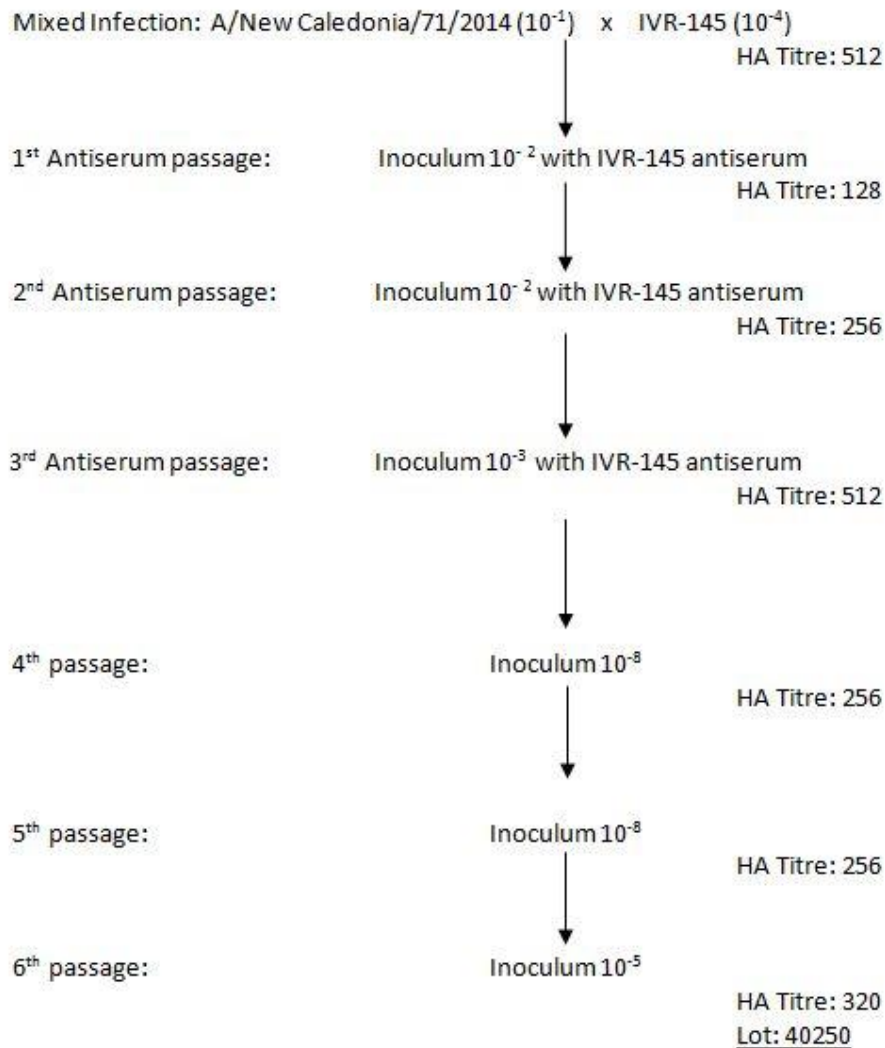
Sterility: no visible contamination was detected in a variety of media (tryptose soya broth, thioglycolate broth, Sabouraud's broth and blood agar plates) after 14 days incubation.



## Derivation of NIB-91

### A/New Caledonia/71/2014 (H3N2)-like High Growth Reassortant

Strain: A/New Caledonia/71/2014 (H3N2)  
Received from VIDRL #1409151-1, E5  
Passage undertaken at NIBSC #40020, E6



Total number of passages since mixed infection= E6

SPF eggs were used for all passages.

RT-PCR/RFLP analysis indicates that NIB-91 has HA and NA genes from A/New Caledonia/71/2014 and NP, NS, PB1, PB2, PA and M genes from IVR-145 making it a 6:2 reassortant.

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.



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Wednesday, 25 March 2015

This is to confirm that NIB-91 was tested in a 2-way HI test on 24 March 2015 and passed the test. The post infection antiserum raised against the egg-propagated cultivar of A/New Caledonia/71/2014 (NIBSC F11/15) recognised NIB-91 with a titre equal to the titre of the antiserum for the homologous virus. Antiserum raised against NIB-91 (NIBSC F12/15) recognised against the egg-propagated cultivar of A/New Caledonia/71/2014 with the same titre as it recognised the homologous virus. The results are shown below. Note that NIB-91 is recognised similarly to IVR-178.

Antigenic analyses of influenza A(H3N2) viruses (Guinea Pig RBC with 20nM Oseltamivir) 2015-03-24										NEW	NEW
Viruses	Collection	Passage	A/HK	A/HK	A/HK	A/HK	A/New Cal	NIB-91			
			History	5738/14	5738/14	7127/14	4801/14	71/14	(NC/71/14)		
	Date	History	T/C	NIB	F11/15	F12/15	NIBSC	NIBSC			
			F30/14	F53/14			F11/15	F12/15			
	Genetic group		3C.2a	3C.2a cl 121							
<b>REFERENCE VIRUSES</b>											
A/Hong Kong/5738/2014	3C.2a	2014-04-30	MDCK1/MDCK2/SIAT1	160	40	160	80	80	<		
A/Hong Kong/5738/2014	3C.2a	2014-04-30	E5/E1 clone121	320	640	1280	320	2560	2560		
150057 A/Hong Kong/7127/2014		2014-07-29	E5	160	320	640	160	640	640		
142089 A/Hong Kong/4801/2014		2014-02-26	E5 isolate 1	160	640	1280	320	1280	1280		
<b>TEST VIRUSES</b>											
150355 A/New Caledonia/71/2014		2014-08-13	E5/E1	160	640	1280	320	1280	1280		
151343 IVR-178 (A/New Caledonia/71/2014)			E5/D7/E1	160	640	1280	320	1280	1280		
151433 NIB-91 (A/New Caledonia/71/2014)			E6	160	320	640	160	1280	1280		

1. <= <40

Also shown are the titres of the antisera for both the cell-propagated and cell-propagated cultivars of A/Hong Kong/5738/2014 and two other egg-propagated

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viruses, A/Hong Kong/7127/2014 and A/Hong Kong/4801/2014.

I hope that you find the results useful.

With best wishes,  
Yours sincerely,

John McCauley.

Digitally signed by John William  
McCauley

Date: 2015.04.17 19:35:41 +01'00'