Medicines & Healthcare products Regulatory Agency

NIBSC Confidence in Biological Medicines

Influenza Reagent
Influenza Virus infectious IVR-165
NIBSC code: 12/204
Instructions for use
(Version 1.0, Dated 01/11/2012)

1. INTENDED USE

The influenza reference virus IVR-165 is a reassortant prepared by CSL Ltd using classical reassortant methodology from A/Victoria/361/2011 virus and A/Puerto Rico/8/34 virus. Reagent 12/204 is prepared from IVR-165 and 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 IVR-165 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 handled only in appropriate containment facilities by fully trained competent staff. It should be used and disposed of in accordance with national safety guidelines and your laboratory's safety procedures.

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 freeze dried allantoic fluid from embryonated SPF hen's eggs.

5. STORAGE

Store in the dark at -20^{0}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

A range of dilutions (e.g. 10^{-3} to 10^{-5}) should be made in a suitable medium for initial cutivation.

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
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 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 and waste disposal procedures should follow those outlined in your facility standard laboratory operating procedures. Appropriate disinfectants would include Chlorine based chemicals, 70% Ethanol and phenolic compounds when used according to manufacturer's specified recommendations.

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

National Institute for Biological Standards and Control,
Potters Bar, Hertfordshire, EN6 3QG. T +44 (0)1707 641000, nibsc.org
WHO International Laboratory for Biological Standards,
UK Official Medicines Control Laboratory

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: NA
Toxicity Statement: Non-toxic
Veterinary certificate or other statement if applicable**.** **Attached:** No

Passage history of IVR-165

Passage level	Lot	Laboratory
E1-E3		VIDRL, Melbourne, Australia
D1 - D6	VI-1555	CSL, Melbourne, Australia
E6	34520	NIBSC, Hertfordshire, UK

E = SPF eggs

Attached derivation as received from CSL
HI data as received from VIDRL, Melbourne.

CSL

Derivation of IVR-165
A/Victoria/361/2011 – like High Growth Reassortant

PREPARATION
Preparation of IVR-165, lot VI-1555, an A/Victoria/361/2011 (H3N2) high growth reassortant influenza virus was conducted in the Influenza Development department, R&D, CSL Limited.

MATERIALS
The following materials of biological origin were used during preparation of high growth reassortant IVR-165:

Virus Isolate: The virus isolate was obtained from the WHO Collaborating Centre for Reference & Research on Influenza, Melbourne (WHO-CC).
Supply details are:
A/Victoria/361/2011 (Type A, Subtype H3N2)
WHO-CC Laboratory number: SL/1110498
Passages prior to receipt at WHO-CC: Nil
Passages undertaken in WHO-CC: E3, HA=128(guinea pig red cells)

Eggs: SPF Premium Plus eggs were used for all passages.

Antiserum: Trypsin-periodate treated sheep hyperimmune antiserum Lot# AS367, sub-lot # 4720, raised against influenza virus A/Puerto Rico/8/34.
The sheep antiserum was derived from sheep born and raised in Australia.

Note on Transmissible Spongiform Encephalopathies (TSEs):
Australia and New Zealand have been declared TSE free in accordance with OIE guidelines. Detailed information on Australia's animal health status can be obtaind from the following Animal Health Australia website link:
www.animalhealthaustralia.com.au/aahc/status/ahia.cfm
The trypsin used is 10x solution of gamma irradiated porcine pancreatic trypsin;
Invitrogen / Gibco Cat # 15090, Lot No. 798572

PASSAGE HISTORY:
Mixed infection passage: A/Victoria/361/2011 (H3N2) wild type virus @10^{-5} x A/Puerto Rico/8/34 (H1N1) ($@10^{-3}$ HA titre 1040 (with CRBC

↓

1st Antiserum Passage Inoculum @ 10^{-3} with antiserum to A/Puerto Rico/8/34 (H1N1) HA titre ND (with CRBC)

↓

2nd Antiserum Passage Inoculum @ 10^{-3} with antiserum to A/Puerto Rico/8/34 (H1N1) HA titre 502 (with CRBC)
HA titre ≥1154 (with GPRBC)

↓

1st Limit dilution passage Inoculum @ 10^{-8} HA titre 710 (with CRBC)

↓

2nd Limit dilution passage Inoculum @ 10^{-9} HA titre 1114 (with CRBC)

↓

Preparation of IVR-165 Lot VI-1555
Inoculum @ 10^{-5} mean HA titre 454 (with CRBC)

Total number of passages post mixed infection = 5
Total number of passages since this virus was received from an approved laboratory = 6
HA titres were determined using chicken red blood cells at room temperature.
Guinea Pig red cells were also used at 2nd Antiserum passage.

TESTING OF INFLUENZA VIRUS SPF LOT VI-1555:

Test	Result	
Sterility	Pending	
Antigenicity	Seed lot VI-1555 (IVR-165) has a HI reactivity pattern that is consistent with the wild type A/Victoria/361/2011 virus. See WHO report on one-way HI testing attached	
Genotype (by real time RT-PCR)	6:2 H1, N1 from A/Victoria/361/2011 Remaining 6 internal genes from PR-8	
	A/Victoria/361/2011	**PR8**
	H3	
	N2	

Disclaimer:
The material i.e. high growth reassortant virus IVR-165 and the information provided in this derivation report are provided on an "as is" basis and as such without any warranty or representation of any kind (express or implied) including, without limitation, of satisfactory quality or fitness for a particular purpose.

Prepared by:
Leonora Pancho
Manager
Influenza Development, R&D, CSL Limited
Wednesday, 15th February 2012

WHO COLLABORATING CENTRE FOR REFERENCE AND RESEARCH ON INFLUENZA MELBOURNE AUSTRALIA 10 Wreckyn St, North Melbourne, Victoria, 3051, Australia Phone: +61 3 9342 3900 Fax: +61 3 9342 3939 www.influenzacentre.org

National Institute for Biological Standards and Control,
Potters Bar, Hertfordshire, EN6 3QG. T +44 (0)1707 641000, nibsc.org
WHO International Laboratory for Biological Standards,
UK Official Medicines Control Laboratory

Medicines & Healthcare products Regulatory Agency

NIBSC Confidence in Biological Medicines

Influenza Virus Seed Lot

Identity Test Report for: CSL Limited

Sample ID No.	1144782	Test Code	CSL: QA 0050
Seed Lot No.	VI-1555	Date submitted	14/02/2011
Sample name	IVR-165 (A/Victoria/361/2011)	WHO ID No.	1202012

Test applied	**Haemagglutination Inhibition Assay**	Assay Date	15 February 2012
Assay performed by	T. Mastorakos		

		HI titre with reference antisera							
Reference antigen		A1	A2	A3	A4	A(H1N1) pdm	B VIC	B YAM	H1
A/PERTH/16/2009	A(H3)	**320**	<20	320	20	<40	<40	<20	<20
A/BRISBANE/10/2007	A(H3)	20	**320**	80	20	<40	<40	<20	<20
A/PERTH/10/2010	A(H3)	80	<20	**640**	40	<40	<40	<20	<20
A/BRISBANE/59/2007	A(H1)	<20	<20	<20	<20	<40	<40	<20	**320**
A/CALIFORNIA/07/2009	A(H1N1) pdm	<20	<20	<20	<20	**1280**	<40	<20	<20
B/BRISBANE/33/2008	B VIC	<20	<20	<20	<20	<40	**1280**	<20	<20
B/WISCONSIN/1/2010	B YAM	<20	<20	<20	<20	<40	<40	**160**	<20
A/VICTORIA/361/2011	A(H3) (WT)	40	80	160	**640**	<40	<40	<20	<20
Test antigen									
VI-1555		160	160	640	640	<40	<40	<20	<20

Actual antisera used were raised to:	
A1	A/PERTH/16/2009
A2	A/BRISBANE/10/2007
A3	A/PERTH/10/2010
A4	A/VICTORIA/361/2011
A(H1N1) pdm	A/CALIFORNIA/07/2009
B VIC	B/BRISBANE/33/2008
B YAM	B/WISCONSIN/1/2010
H1	A/BRISBANE/59/2007

Conclusion: Seed lot VI-1555 (IVR-165) has a HI reactivity pattern that is consistent with the wild type A/Victoria/361/2011 virus.

Pass	Fail	Warn
■	☐	☐

Ian Barr
Deputy Director
15.02.2012

National Institute for Biological Standards and Control,
Potters Bar, Hertfordshire, EN6 3QG. T +44 (0)1707 641000, nibsc.org
WHO International Laboratory for Biological Standards,
UK Official Medicines Control Laboratory