3.2.S.5

Certificate of Analysis - Pertussis Identity (PT) Reference - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

A.N.M.A.T. FOLIO 002589 MESA DE ENTRADAS

sanofi pasteur

Product Quality and Disposition Department

The vaccines division of sanofi-aventis Group

QUALITY CONTROL RELEASE

NATIVE PURIFIED PERTUSSIS TOXIN

Lot number **FA313027**

Protein content	: **1190.50** µg/ml
SDS-page	: **Conforms**
Pertussis toxin identification	: **Positive**
Clustering activity on Chinese Hamster Ovary cells	: **204800** CPU/µg of protein
Haemagglutining activity	: **102357** HAU/µg IHA with cholesterol : **0 % - Conforms**

Expiry date : **JUNE 2012**

GENERAL CONCLUSION : Conforms

Release date : **01.10.2008**

C FEUILLET
20 JUIL. 2010

Product Quality and Disposition Department

004-VRAC PERT - TPn CC2.dot/20.07.10

This document is intended for health authority biological release purposes only

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

3.2.S.5

Certificate of Analysis - NIBSC 90 518 Standard Reference - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

Resuelto 06/03/09

NIBSC
Assuring the quality of biological medicines

Blanche Lane
South Mimms
Potters Bar
Hertfordshire EN6 3QG
United Kingdom

Tel: +44 (0)1707 641000
Fax: +44 (0)1707 641050
Email: enquiries@nibsc.ac.uk
Web: http://www.nibsc.ac.uk

Non WHO Reference Material
Bordetella pertussis Toxin (PT)
NIBSC code: 90/518
Instructions for use
(Version 5.0, Dated 10/04/2008)

1. INTENDED USE

This material is not for in vitro diagnostic use.
The British Reference Preparation of Pertussis Toxin is an extract of *Bordetella pertussis* culture supernatant. It is a reference preparation for use in the control testing of acellular pertussis vaccines.

2. CAUTION

This preparation is not for administration to humans.

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

This material has an assigned unitage of 2100 IU per ampoule based on its calibration in terms of preparation JNIH-5.

4. CONTENTS

Country of origin of biological material: United Kingdom.
Each ampoule contains the freeze dried residue of 0.5ml of 0.01M sodium phosphate pH 7.6, 0.5M NaCl which contained:

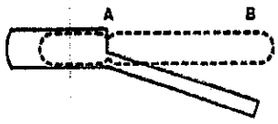
Pertussis toxin	20 micrograms
Trehalose	5 mg

5. STORAGE

Unopened ampoules should be stored at -20°C.

6. DIRECTIONS FOR OPENING

Tap the ampoule gently to collect the material at the bottom (labelled) end. Ensure ampoule is scored all round at the narrow part of the neck, with a diamond or tungsten carbide tipped glass knife file or other suitable implement before attempting to open. Place the ampoule in the ampoule opener, positioning the score at position 'A'; shown in the diagram below. Surround the ampoule with cloth or layers of tissue paper. Grip the ampoule and holder in the hand and squeeze at point 'B'. The ampoule will snap open. Take care to avoid cuts and projectile glass fragments that enter eyes. Take care that no material is lost from the ampoule and that no glass falls into the ampoule.



Side view of ampoule opening device containing an ampoule positioned ready to open. 'A' is the score mark and 'B' the point of applied pressure.

7. USE OF MATERIAL

Purified PT was generously donated by SmithKline Biologicals, Rixensart, Belgium, through the good offices of Dr C Capiau. The material was >98% pure by silver stained SDS-PAGE, and contained <0.1ng of endotoxin per mg of protein by LAL assay.

Ampoules coded 90/518 were prepared according to the procedures used for International Biological Standards (29th ECBS Report, 1978). A sodium buffered solution of PT, of known concentration, was diluted with a sterile solution of NaCl and trehalose to yield a solution of 0.001M sodium phosphate pH 7.6, 0.5M NaCl containing 1% trehalose and 0.004% PT. The solution was distributed in 0.5 ml aliquots into ampoules. The ampouled solution was lyophilised, and the ampoules sealed under nitrogen by heat fusion of the glass and stored at -20°C in the dark.

For practical purposes each ampoule contains the same quantity of PT. The entire contents of each ampoule should be completely dissolved in an accurately measured amount of solvent (distilled water, saline or buffer) and the solution kept cool (e.g +4°C) prior to use. No attempt should be made to weigh out proportions of the freeze dried powder. It is recommended that the solution, not for immediate use, is stored at -20°C or below. Repeated freezing and thawing should be avoided. The ampoules contain no bacteriostat and the preparation should not be assumed to be sterile.

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

1) D.Xing, R G Das and P.Newland
Collaborative Study; evaluation of proposed international reference preparation of pertussis toxin, Code JNIH-5
WHO BS/03.1978

2) D.Xing et al. Vaccine 20 (2002) 3535 - 3542

10. ACKNOWLEDGEMENTS

Grateful acknowledgements are due to: Dr C Capiau and SmithKline Biologicals for providing the material and Standards Processing Division for the filling.

11. FURTHER INFORMATION

Further information can be obtained as follows;
This material: enquiries@nibsc.ac.uk
WHO Biological Standards: Http://www.who.int/biologicals/en/
JCTLM Higher order reference materials: http://www.bipm.org/jctlm
Derivation of International Units:
Http://www.nibsc.ac.uk/products/faq.asp
Ordering standards from NIBSC:
Http://www.nibsc.ac.uk/products/faq.asp
NIBSC Terms & Conditions: Http://www.nibsc.ac.uk/terms.html

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.ac.uk

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

Physical and Chemical properties

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

NIBSC
Assuring the quality of biological medicines

Blanche Lane
South Mimms
Potters Bar
Hertfordshire EN6 3QG
United Kingdom

Tel: +44 (0)1707 641000
Fax: +44 (0)1707 641050
Email: enquiries@nibsc.ac.uk
Web: http://www.nibsc.ac.uk

Physical appearance: Freeze dried powder	Corrosive:	No
Stable: Yes	Oxidising:	No
Hygroscopic: No	Irritant:	No
Flammable: No	Handling:	See caution, Section 2
Other (specify):	Contains material of biological origin	

Toxicological properties

Effects of inhalation: Not established, avoid inhalation
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 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

Information provided by the Institute is given after the exercise of all reasonable care and skill in its compilation, preparation and issue, but it is provided without liability to the Recipient in its application and use.

It is the responsibility of the Recipient to determine the appropriateness of the standards or reference materials supplied by the Institute to the Recipient ("the Goods") for the proposed application and ensure that it has the necessary technical skills to determine that they are appropriate. Results obtained from the Goods are likely to be dependant on conditions of use by the Recipient and the variability of materials beyond the control of the Institute.

All warranties are excluded to the fullest extent permitted by law, including without limitation that the Goods are free from infectious agents or that the supply of Goods will not infringe any rights of any third party.

The Institute shall not be liable to the Recipient for any economic loss whether direct or indirect, which arise in connection with this agreement.

The total liability of the Institute in connection with this agreement, whether for negligence or breach of contract or otherwise, shall in no event exceed 120% of any price paid or payable by the Recipient for the supply of the Goods.

If any of the Goods supplied by the Institute should prove not to meet their specification when stored and used correctly (and provided that the Recipient has returned the Goods to the Institute together with written notification of such alleged defect within seven days of the time when the Recipient discovers or ought to have discovered the defect), the Institute shall either replace the Goods or, at its sole option, refund the handling charge provided that performance of either one of the above options shall constitute an entire discharge of the Institute's liability under this Condition.

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: 0.5 - 1.0 g
Toxicity Statement: Toxic
Veterinary certificate or other statement if applicable. Attached: No

A World Health Organization International Laboratory For Biological Standards

National Institute for Biological Standards and Control

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

3.2.S.5

Certificate of Analysis - In-House Haemagglutinating Activity (FHA)
Reference Material - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

sanofi pasteur

La division vaccins du Groupe sanofi aventis

A.N.M.A.T. 00239 FOLIA DE ENTRADAS

CERTIFICATE OF ANALYS

Produit : FHA validity control

Lot : 11-2001

Date 27/11/2001

Production

Native purified FHA FA071446 solubilized, diafiltered, and filtered.

Utilisation

- Storage at -70°C
- Protein content: 1.35 mg / mL

November, 25th 2010

Eric Deshaies
Virological QC Lab Manager

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

3.2.S.5

Certificate of Analysis - In-House Haemagglutinating Activity (PT) Reference Material - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

sanofi pasteur

La division vaccins du Groupe sanofi-aventis

A.N.M.A.T. FOLIO 259 ADUANA DE ENTRADAS

CERTIFICATE OF ANALYS

Produit : Toxin Pertussis validity Control
for IHA test

Lot : FA287985
Date 20/09/2007

Production

Solution of Pertussis Toxin : sample B (after filtration) at 692,33µg/mL diluted in CTW buffer to have a final solution at 100µg/mL (realized by manufacturing team)
At lab : sampling in 0,5mL volume.

Utilisation

- Storage at -20°C
- Protein content : 100 µg / ml

November, 26th 2010

Eric Deshaies
Virological QC Lab Manager

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

sanofi pasteur. Campus Mérieux, 1541, avenue Marcel Mérieux – F-69280 Marcy l'Etoile – Tél.: +33(0)4 37 37 01 00 - Fax : +33(0)4 37 37 38 54 -
www.sanofipasteur.com

3.2.S.5

Certificate of Analysis - In-House Antigenicity (FHA) Reference Material - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

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The vaccines division of sanofi-aventis Group

Product Quality and Disposition Department

QUALITY CONTROL RELEASE

NATIVE PURIFIED FHA

Lot number **FA303375**

Protein content	: **1169.56** µg/ml
SDS-page	: **Conforms**
FHA identification	: **Positive**
Clustering activity on Chinese Hamster Ovary cells	: **No cytopathic effect**
Haemagglutining activity	: **1024385** HAU/mg IHA with cholesterol: **98% - Conforms**

Expiry date: **MARCH 2012**

GENERAL CONCLUSION: Conforms

Release date: **22.07.2008**

Product Quality and Disposition Department
Anne LAMOTTE

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

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Certificate of Analysis - In-House Antigenicity (PT) Reference Material - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

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Product Quality and Disposition Department

QUALITY CONTROL RELEASE

NATIVE PURIFIED TPN

Lot number **FA353914**

Protein content	: **1149.91** µg/ml
SDS-page	: **Conforms**
Toxine Pertussis identification	: **Positive**
Clustering activity on Chinese Hamster Ovary cells	: **102400** CPU/µg of protein
Haemagglutining activity	: **102408** HAU/mg IHA with cholesterol: **0% - Conforms**

Expiry date: **JULY 2013**

GENERAL CONCLUSION: Conforms

Release date: **17.09.2009**

Anne LAMOTTE
Product Quality and Disposition Department

094-VRAC PERT – TPn CC2.dot/20.07.10
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ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

3.2.S.5

Certificate of Analysis - Endotoxins Reference Material - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

charles river
accelerating drug development. exactly.

A.N.M.A.T. FOLIO 0241 MESA DE ENTRADAS

CERTIFICATE OF ANALYSIS

VIAL CONTENTS: Endosafe® Control Standard Endotoxin is prepared from *E. coli* strain 055:B5. Each vial contains 500 ng of purified Lipopolysaccharide, freeze dried in a stabilized matrix.

RSE/CSE RATIO: The potency of this standard in Endotoxin units, (EU) has been determined to be 22 EU/ng by the method described in Appendix C (***Gel-clot Technique***) of the GUIDELINE ON VALIDATION OF THE LIMULUS AMEBOCYTE LYSATE TEST AS AN END PRODUCT ENDOTOXIN TEST FOR HUMAN AND ANIMAL PARENTERAL DRUGS, BIOLOGICAL PRODUCTS, AND MEDICAL DEVICES, published by the U.S Food and Drug Administration.

CSE Lot: EM04972 LAL Reagent Lot: B4352L RSE Lot: EC-6-3

RSE/CSE Ratio: 22 EU/ng Vial contents: 11,000 EU/vial

Geometric Mean Sensitivity with RSE: 0.03 EU/mL

IS/CSE RATIO: The Expert Committee on Biological Standardization of WHO has assigned a potency of the IS as 10,000 IU per vial of IS, so that 1 IU = 1 EU. The potency of this endotoxin standard in International (Endotoxin) Units, IU, has been designated as 22 IU/ng.

DIRECTIONS FOR USE: Reconstitute the lyophilized material with 5.5 mL of LAL reagent grade water to obtain 2000 EU/mL or 2000 IU/mL. Vortex mix vigorously for at least 5 minutes after rehydration, and for at least 1 minute immediately prior to each use.

STORAGE: Store rehydrated material at 2-8°C for up to 4 weeks. Store lyophilized material at controlled room temperature or refrigerated as preferred. Diluted endotoxin should not be stored except under validated conditions.

CAUTION: DO NOT FREEZE ENDOTOXIN SOLUTIONS

Signature: Cynthia Rhoda Date: 26 Apr 2010

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.

endotoxin and microbial detection
1023 Wappoo Road, Suite 43B, Charleston, SC 29407 • 843.409.4900 • Fax: 843.766.7576 • www.criver.com

3.2.S.5

Certificate of Analysis - Residual Glutaraldehyde Content External Reference - FHA PTxd

ROXANA MONTEMILONE
DIRECTORA TÉCNICA
SANOFI PASTEUR S.A.

CHRISTIAN DOMINGUEZ
APODERADO
SANOFI PASTEUR S.A.