



**WHO International Standard
4th WHO International Standard for Diphtheria Toxoid (Adsorbed)
NIBSC code: 07/216
Instructions for use
(Version 2.0, Dated 29/11/2012)**

1. INTENDED USE

This standard preparation was prepared from a batch of purified diphtheria toxoid, adsorbed (Lot No. DAST-BKN0701) supplied by Biken, Japan. The purified toxoid had a diphtheria antigen content of 200 Lf/ml adsorbed to aluminium phosphate (1.5 mg/ml). The purified toxoid met the minimum requirements for biological products, Japan. The standard was prepared at NIBSC and stabilised by 1:1 dilution of the purified toxoid with 3.5% w/v Polygeline (Haemaccel®, Kora Healthcare, Dublin) to give a final concentration of 1.75 % w/v Polygeline. This material was then distributed into ampoules (1 ml per ampoule) and freeze-dried in the NIBSC Standards Processing Division in August 2007. The freeze-dried product is intended for use as a primary standard in assays to measure the potency of diphtheria vaccine (adsorbed) or for calibration of secondary reference materials.

2. CAUTION

This preparation is not for administration to humans or animals in the human food chain.

The active ingredient in Haemaccel®, Polygeline, is derived from bovine gelatin. Haemaccel® is licensed for human use and does not contain bovine material sourced from a territory considered to be a BSE risk. 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

The candidate standard was calibrated in an International Collaborative Study involving 30 laboratories in 20 different countries [1]. Calibration of the standard was based on results obtained in WHO/Ph. Eur. protection methods against the 3rd WHO International Standard for Diphtheria Toxoid, Adsorbed (98/560). The replacement standard was adopted by the WHO ECBS in October 2009. The replacement standard, coded 07/216, has a defined potency of 213 International Units (IU) per ampoule based on the results returned by 19 laboratories who performed WHO/Ph. Eur. protection assays for calibration of the candidate material [1].

4. CONTENTS

Country of origin of biological material: Japan.
Each ampoule of the freeze dried standard preparation contains approximately 100 Lf of purified diphtheria toxoid, 0.75 mg aluminium phosphate and 17.5 mg Polygeline (degraded, cross-linked gelatin polypeptides). The ampoules contain no bacteriostat and should not be assumed to be sterile.

5. STORAGE

Ampoules should be stored in the dark at -20°C.
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 manufacturers instructions provided with the ampoule breaker.

7. USE OF MATERIAL

No attempt should be made to weigh out any portion of the freeze-dried material prior to reconstitution.

After opening, the entire contents of each ampoule should be completely re-suspended in 1 ml of sterile water or other suitable solution such as 0.9% NaCl. It is recommended that the reconstituted standard be used as soon as possible after reconstitution and on the same day.

DO NOT FREEZE THE RECONSTITUTED STANDARD.

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.

It is the policy of WHO not to assign an expiry date to their International Reference Materials. Accelerated degradation studies performed during collaborative study [1] suggest that this material will be suitably stable when stored at the recommended storage temperature of -20°C, and the assigned potency value remains valid until the material is withdrawn or replaced.

Users who have any data supporting any change in the characteristics of this material are encouraged to contact NIBSC.

9. REFERENCES

[1] Stickings P, Rigsby P, Coombes L, Malik K, Matejtschuk, P and Sesardic D. 2010. Collaborative Study for the Calibration of a Replacement International Standard for Diphtheria Toxoid Adsorbed. *Biologicals*, 38: p 529-538.

10. ACKNOWLEDGEMENTS

Dr Toshiyuki Onishi (Biken, Japan) is gratefully acknowledged for donation of the purified toxoid material used in preparation of the replacement standard. All participants of the collaborative study performed to calibrate this replacement standard are gratefully acknowledged.

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: Freeze-dried powder	Corrosive: No
Stable: Yes	Oxidising: No
Hygroscopic: No	Irritant: Unknown
Flammable: No	Handling: See caution, Section 2
Other (specify): N/A	
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 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 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: 0.031 g
Toxicity Statement: Non-toxic
Veterinary certificate or other statement if applicable. Attached: No

17. CERTIFICATE OF ANALYSIS

NIBSC does not provide a Certificate of Analysis for WHO Biological Reference Materials because they are internationally recognised primary reference materials fully described in the instructions for use. The reference materials are established according to the WHO Recommendations for the preparation, characterization and establishment of international and other biological reference standards http://www.who.int/bloodproducts/publications/TRS932Annex2_Inter_bi_olefstandardsrev2004.pdf (revised 2004). They are officially endorsed by the WHO Expert Committee on Biological Standardization (ECBS) based on the report of the international collaborative study which established their suitability for the intended use.