WHO International Standard
First International Standard for anti-Hepatitis B core antigen (anti-HBc), plasma, human
NIBSC code: 95/522

Instructions for use
(Version 2.0, Dated 15/04/2013)

1. INTENDED USE
Antibodies to hepatitis B virus core antigen (anti-HBc) are produced during acute hepatitis B virus (HBV) infection and persist lifelong so that HBV infection can be detected in chronic carriers even when negative for hepatitis B surface antigen (HBsAg) and HBV-DNA. Anti-HBc screening therefore has the potential to detect the majority of occult HBV infection. In the absence of anti-HBc testing HBV transmission occurred in blood recipients as well as after organ transplants. This has led some countries to improve blood safety by mandatory blood screening for anti-HBc (e.g., Argentina, Brazil, France, Germany, Japan, Paraguay, Peru, Uruguay, USA and Venezuela). In addition, anti-HBc may be the only positive serological marker in chronic HBV infections.

Anti-HBc persists also in those who have cleared the virus so that isolated anti-HBc can be indistinguishable from serological profile of resolved HBV infection not easy to differentiate from potential false positive reactions in particular in low prevalence HBV countries. Therefore high quality anti-HBc tests with high sensitivity and specificity are required.

A WHO Collaborative Study organised by the Paul Ehrlich Institute was undertook to assess the suitability of a candidate reference material (NIBSC code 95/522) for detection of antibodies to hepatitis B core antigen (anti-HBc) in diagnostic assays. Thirteen laboratories from 10 countries tested the above described materials using 20 different anti-HBc assays.

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

The preparation contains material of human origin, and either the final product or the source materials, from which it is derived, have been tested and found negative for HBsAg, anti-HIV and HCV RNA. its material contains high levels of anti-HBs. It has also been tested and found negative for HBV DNA and is therefore considered non-infectious. 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 is assigned a unitage of 50IU/ampoule. (50 IU/ampoule are equivalent to the 100 PEI-U/ml of the PEI 82 anti-HBc standard)

4. CONTENTS
Country of origin of biological material: United Kingdom. This preparation contains the freeze dried residue of 1ml plasma from UK blood donors which is reactive for anti-HBc and also for anti-HBs at high levels.

5. STORAGE
Ampoules should be stored 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. Tap the ampoule gently to collect the material at the bottom (labeled) end. Ensure that the disposable ampoule safety breaker provided is pushed down on the stem of the ampoule and against the shoulder of the ampoule body. Hold the body of the ampoule in one hand and the disposable ampoule breaker covering the ampoule stem between the thumb and first finger of the other hand. Apply a bending force to open the ampoule at the coloured stress point, primarily using the hand holding the plastic collar.

Care should be taken to avoid cuts and projectile glass fragments that might enter the eyes, for example, by the use of suitable gloves and an eye shield. Take care that no material is lost from the ampoule and no glass falls into the ampoule. Within the ampoule is dry nitrogen gas at slightly less than atmospheric pressure. A new disposable ampoule breaker is provided with each DIN ampoule.

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

Each ampoule should be reconstituted in 1ml distilled water. It is recommended that human serum or defibrinated human plasma (anti-HBc negative) is used in the preparation of a series of dilutions of the IS when calibrating secondary standards as the use of diluents such as PBS/BSA may interfere with the test results.

8. STABILITY
Reference materials are held at NIBSC within assured, temperature-controlled storage facilities and they 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 have indicated that this material is suitably stable, when stored at -20°C or below, for the assigned value to remain valid until the material is withdrawn or replaced. These studies have also shown that the material is suitably stable for shipment at ambient temperature without any effect on the assigned values. Users who have data supporting any deterioration in the characteristics of any reference preparation are encouraged to contact NIBSC.

Users should determine the stability of reconstituted material according to their own method of preparation, storage and use. Stability studies on reconstituted material in the Collaborative Study showed that there was no loss of activity after 14 days storage at +2-8°C and after two freeze/thaw cycles. However, multiple freeze/thaw cycles should be avoided.

9. REFERENCES
WHO Expert Committee on Biological Standardization report WHO/BS/08.2098 available online. http://apps.who.int/iris/bitstream/10665/69975/1/WHO_BS_08.2098_eng.pdf

10. ACKNOWLEDGEMENTS

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:
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

<table>
<thead>
<tr>
<th>Physical and Chemical properties</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Physical appearance: Freeze dried powder</td>
<td>Corrosive: No</td>
</tr>
<tr>
<td>Stable: Yes</td>
<td>Oxidising: No</td>
</tr>
<tr>
<td>Hygroscopic: No</td>
<td>Irritant: No</td>
</tr>
<tr>
<td>Flammable: No</td>
<td>Handling: See caution, Section 2</td>
</tr>
<tr>
<td>Other (specify):</td>
<td>Contains material of human origin</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Toxicological properties</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Effects of inhalation:</td>
<td>Not established, avoid inhalation</td>
</tr>
<tr>
<td>Effects of ingestion:</td>
<td>Not established, avoid ingestion</td>
</tr>
<tr>
<td>Effects of skin absorption:</td>
<td>Not established, avoid contact with skin</td>
</tr>
</tbody>
</table>

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
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: 1.0g
- 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_biol_efstandardsrev2004.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.