Medicines & Healthcare products Regulatory Agency

NIBSC Confidence in Biological Medicines

Non WHO Reference Material
Anti-Nucleolar Factor Plasma, Human
NIBSC code: 68/340
Instructions for use
(Version 5.0, Dated 04/04/2008)

This material is not for in vitro diagnostic use.

1. INTENDED USE

In collaboration with the Immunology Unit, WHO, Geneva, the National Institute for Biological Standards and Control has been developing research standards for a number of human autoimmune antibodies and other materials. A Research Standard for Anti-Nuclear Factor Serum (Homogeneous) Human has already been established (Anderson, Addison, and Dixon, 1971). A Research Standard for Anti-Nucleolar Plasma Human has been developed and is described here.

Research Standard A for Anti-Nucleolar Factor Plasma, Human 68/340, is a freeze-dried preparation of citrated plasma. The source material was obtained in Holland by plasmapheresis of a patient with mild rheumatoid arthritis. It was supplied by Dr. T.E.W Feltkamp of the Central Laboratory for Blood Transfusion, Amsterdam. It was received on 6th February 1968, in the liquid state, and was then stored at -20°C.

On 27th August 1968, the bulk plasma was thawed and diluted to a final concentration of approximately 33%, the diluent being 20% normal calf serum in physiological saline. The material was then clarified by passage through a coarse, non-sterilising filter (Millipore AP 20). On 28th August it was distributed into ampoules, pre-frozen in liquid nitrogen and freeze-dried. On 30th August the ampoules were fitted with plastic capillary-leak plugs and the contents were dried to constant weight by secondary dessication in vacuuo over phosphorous pentoxide. On 5th September the ampoules were filled with pure dry nitrogen and sealed by fusion of glass. They were examined the next day for presence of pinholes and leaks, and since that time have been stored at -20°C in the dark. During the fill 66 ampoules were taken at more or less regular intervals from the total of 3500 and were tested for wet weight of contents. The mean weight was 300mg and the range +/- 0.83%.

The dry weight of freeze-dried material was determined in each of 8 sealed ampoules. The mean weight was 12.35mg (range +/-1.8%). The mean estimated moisture content was 0.28% in 8 sealed ampoules (range 0 to 0.65%).

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. 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 preparation has been assigned a unitage so that 1 unit is the activity present in 0.1235mg of the freeze-dried powder and, for practical purposes, each ampoule contains 100 units.

4. CONTENTS

Country of origin of biological material: United Kingdom.

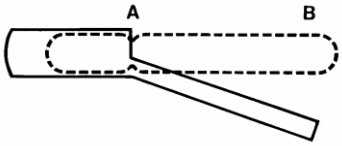
5. STORAGE

Store at unopened ampoules 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

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

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

In order to use this research standard the contents of 1 ampoule should be reconstituted by adding 0.3ml of distilled water. The contents should then be repeatedly and completely washed out with suitable diluent and the diluted plasma made up to a convenient volume. The reconstituted diluted standard should be titrated alongside the test serum in any assay.. The relative titres of standard and test sera can be estimated and the relative potencies of the test sera can be expressed in units per given volume.

8. STABILITY

Two sets of stability studies were done. Activities were estimated in an immunofluorescence test on sections of frozen rat liver. The technique was based on that described by Anderson et al (1971). In the first set the source material was diluted 1 in 5 and 1 in 10 in 20% calf serum in saline and freeze-dried. Both dilutions were held at 37°C for 6 months. No loss of activity was demonstrated during the freeze drying process nor during the storage at 37°C.

In the second set, the freeze-dried preparation 68/340 was examined, using dilution intervals of 2-fold or less. The preparation was not found to have lost activity during the freeze-drying nor on storage at +4°C for 19 months. However, ampoules of 68/340 stored for 19 months at +37°C had lost 50% to 75% of their activity in comparison with those stored at -20°C. The 37°C material was also incompletely soluble in 0.3ml of distilled water per ampoule, whereas the 4°C and the -20°C materials were completely soluble.

NIBSC follows the policy of WHO with respect to its reference materials. It is the policy of WHO not to assign an expiry date to their international reference materials. They remain valid with the assigned potency and status until withdrawn or amended.

Reference materials are held at NIBSC within assured, temperature-controlled storage facilities. Reference materials should be stored on receipt as indicated on the label. For information specific to a particular biological standard, contact NIBSC.

Users who have data supporting any deterioration in the characteristics of any reference preparation are encouraged to contact NIBSC.

9. REFERENCES

Anderson, S.G Addison, I.E and Dixon H.G (1971) Antinuclear Factor Serum (Homogeneous) Ann. New York Acad. Sci., 177, 337

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WHO International Laboratory for Biological Standards,
UK Official Medicines Control Laboratory

10. ACKNOWLEDGEMENTS

We are very pleased to acknowledge the assistance in the production of this standard given by Dr J. Swanson Beck, Dr. T.E.W Feltkamp and Dr W. Hijmans.

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: No
Flammable: No	Handling: See caution, Section 2
Other (specify): Contains material of human 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

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: 0.01235g
Toxicity Statement: Toxicity not assessed
Veterinary certificate or other statement if applicable**.** **Attached:** No

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