Medicines & Healthcare products Regulatory Agency

NIBSC Confidence in Biological Medicines

WHO International Standard
Luteinizing Hormone, Human, Pituitary.
NIBSC code: 81/535
Instructions for use
(Version 2.0, Dated 22/10/2014)

1. INTENDED USE

The 2nd International Standard (IS) for Luteinizing Hormone (LH), human, pituitary, in ampoules coded 80/552, has been widely used for the calibration of immunoassays of LH. Stocks of the 2nd are exhausted and the WHO Expert Committee on Biological Standardization (ECBS) has recognised (2011) the need for a replacement IS.

This reference material consists of a batch of ampoules, coded 81/535, which contain purified LH of human, pituitary origin. The ampouled preparation has been calibrated in an international collaborative study in terms of the 2nd IS, 80/552. The 3rd IS, coded 81/535, was established at the 65th meeting of the ECBS in 2014. This material replaces the 2nd IS which is discontinued.

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.
However, as with all materials of human origin, the preparation cannot be assumed to be free from infectious agents. The container and its contents should be used and discarded according to your own laboratory procedures. Such procedures probably will include the wearing of protective gloves and avoiding the generation of aerosols. Care should be exercised in opening the container to avoid cuts.

3. UNITAGE

Each ampoule contains 33 INTERNATIONAL UNITS of human, pituitary LH

4. CONTENTS

Country of origin of biological material: Netherlands.
Each ampoule contains the residue, after freeze-drying, of 0.5ml of a solution which contained:

LH extract	approx	5.8 µg
Lactose	"	5 mg
Human plasma albumin	"	1 mg
Sodium chloride	"	90 µg

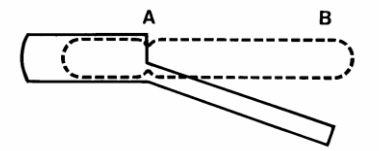
Nitrogen gas at slightly less than atmospheric pressure.

5. STORAGE

Unopened ampoules should be stored 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

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.
For practical purposes each ampoule contains the same amount of human, pituitary LH. Depending on the intended use, dissolve the total contents of the ampoule in a known amount of a suitable diluent. If excessive dilutions are prepared, a carrier protein (0.05 – 0.1% w/v BSA or HSA) should be added. The ampoules do not contain bacteriostat and a solution of the reagent should not be assumed to be sterile.

8. PREPARATION OF AMPOULES

Some 50mg of highly purified pituitary LH, batch no. NM15, were generously donated to WHO by Drs R.M. Lequin and J.G. Loeber (Nijmegen, the Netherlands) and Dr G. Hennen (Liége, Belgium). The batch of ampoules, coded 81/535, was prepared as described in Storring and Gaines Das, 1993. A collaborative study was organised (2013) by NIBSC in which eleven laboratories in seven countries participated, with the aims being:
1) to calibrate 81/535 in terms of the current IS, 80/552
2) to demonstrate the suitability of 81/535 to serve as the IS for the calibration of human, pituitary LH immunoassays by examining its behaviour in immunoassays
3) to assess the stability of 81/535 after accelerated thermal degradation
From this study, the geometric mean estimate of immunoreactivity of 81/535 in terms of 80/552 was 33.2 IU per amp (n=21; 95% confidence limits 32.1 – 34.3; GCV 7.4%). Preparation 81/535 is sufficiently stable to serve as an IS. Analysis of thermally-accelerated degradation samples gave a predicted loss of immunoreactivity per year of 0.014% when stored at -20°C.

9. STABILITY

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. Unopened ampoules should be stored on receipt as indicated on the label. In addition, once reconstituted, diluted or aliquoted, users should determine the stability of the material according to their own method of preparation, storage and use.

10. REFERENCES

Storring PL & Gaines Das, RE (1993). Journal of Endocrinology 138, 345-359

11. ACKNOWLEDGEMENTS

We gratefully acknowledge the important contributions of all the participants in the collaborative study, Dr J G Loeber, Dr R M Lequin and Dr G.Hennen who kindly donated the human, pituitary LH.

12. 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:

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

World Health Organization

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

13. 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

14. 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.

15. 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: Freezed dried powder	Corrosive: No
Stable: Yes	Oxidising: No
Hygroscopic: Yes	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.	

16. 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.

17. 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: 6mg
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_biolefstandardsrev2004.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.