Medicines & Healthcare products Regulatory Agency

NIBSC Confidence in Biological Medicines

WHO International Standard
Fragile X Syndrome, Human gDNA , 1st International Genetic Reference Panel
NIBSC code: 08/158
Instructions for use
(Version 5.0, Dated 13/12/2012)

1. INTENDED USE

The ampoules contain freeze-dried purified gDNA samples extracted from EBV transformed cell lines. They are intended for use as a reference panel in genetic tests for Fragile X syndrome. This panel was established in 2008 as the 1st International Genetic Reference Panel for Fragile X syndrome by the Expert Committee on Biological Standardization (ECBS) of the World Health Organization (WHO).
N.B. these materials should not be put to any other use.

2. CAUTION

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

Each material was tested and found to be negative for HIV1, HTLV1, HBV and HCV by PCR. 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

There is no unitage assigned to these materials.

4. CONTENTS

Country of origin of biological material: United Kingdom.
The DNA samples were extracted using a 'salting out' method and suspended in Tris/EDTA buffer with 2.5 mg/ml Trehalose as an excipient before freeze-drying.
The panel comprises five individually coded ampoules, each containing approximately 23 µg of human gDNA;

07/120 Female, wild-type,
07/122 Female, pre-mutation,
07/168 Female, full mutation,
07/170 Male, full mutation,
07/174 Male, pre-mutation

The panel was tested in an international collaborative study involving 21 laboratories and the following numbers of CGG repeats in the 5' untranslated region of the FMR1 gene were obtained;

Material	CGG repeats	
	Range	Mean
07/120	19-24, 28-33	22, 31
07/122	30-36, 100-132	33, 113
07/168	33-41, 300-401	38, 346
07/170	353-960	754
07/174	97-127	114

In addition, two laboratories detected a 7 CGG repeat in the 07/170 male, full mutation sample.

5. STORAGE

Store all unopened ampoules of the freeze-dried preparations 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. 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 manufactures 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.

a. Open ampoules as described in section 6. above.
b. Reconstitute freeze-dried material at room temperature with 40µL of sterile nuclease-free water.
c. Transfer the entire contents to nuclease-free tubes.
d. Allow the material to reconstitute for 1 hour at room temperature and pipette well to mix before use.
e. Measure DNA concentration before use.
f. We recommend that the material is used directly after reconstitution and is not stored beyond this point, but if this is desired, then the material should be stored in sealed tubes between +2 to +8°C if the samples are to be tested within 3 months. For longer periods, store in aliquots at -20°C or below. Care should be taken to avoid cross-contamination with other samples.

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 -20ºC or below, for the assigned values 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.

9. REFERENCES

The following publication describes the International Collaborative Study which was carried out in order to characterise the panel;
M. Hawkins, J. Boyle, K. Wright, R. Elles, S. Ramsden, A. O'Grady, M. Sweeney, D. Barton T. Burgess, M. Moore, C. Burns, G. Stacey, E. Gray, P. Metcalfe, J.R. Hawkins. Preparation and validation of the first WHO International Genetic Reference Panel for Fragile X Syndrome. European Journal of Human Genetics, advance on-line publication 2010.

10. ACKNOWLEDGEMENTS

We would like to thank the staff of the UK National Genetics Reference Laboratories and the CRMGEN consortium for supplying materials and assistance.

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

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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

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 solid		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 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: 0.011g
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.

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