



**WHO International Standard
PEGylated Granulocyte Colony Stimulating Factor
(Human rDNA derived)
NIBSC code: 12/188
Instructions for use
(Version 2.0, Dated 03/12/2013)**

1. INTENDED USE

The preparation coded 12/188 is the primary standard for *in vitro* biological activity of PEGylated Granulocyte Colony Stimulating Factor (PEG-G-CSF) preparations produced by conjugation of a 20kD monomethoxy linear PEG to the N-terminal methionyl residue of G-CSF and representative of the approved product, INN PEG-Filgrastim.

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 assigned potency agreed on the basis of an International Collaborative Study is 10,000 International Units of biological activity per ampoule. This unitage has been derived independently of the G-CSF International Standard.

Users should note that there is no formal relationship or conversion factor between the units for PEG-G-CSF and the units for the G-CSF International Standard (coded 09/136). Users wishing to compare the activities of the two standards may note that in the study, the average of all assay estimates (excluding the NFS-60/formazan dye assays) was 33,250 IU G-CSF per ampoule of 12/188.

Since 12/188 has only been evaluated for use in *in vitro* bioassays, it cannot be assumed to be suitable for evaluation *in vivo* or for pharmacokinetic studies without suitable validation. Users of this standard will need to perform validation studies if using the standard for purposes other than evaluation of *in vitro* biological activity.

The use of the standard for calibrating other PEG-G-CSF preparations (i.e. produced using PEG of different sizes, forms, or targeted to different sites using alternative coupling chemistries or conjugation procedures) will need to be validated by the user as this has not been sufficiently addressed in the study (WHO/BS/2013.2218).

4. CONTENTS

Country of origin of biological material: United Kingdom.

Each ampoule contains the residue after freeze-drying of 1.0 ml of a solution comprising:

PEG-G-CSF, approximately 1 microgram
10 mg L-Arginine
10 mg L-Phenylalanine
5 mg Trehalose
0.2% Human Serum Albumin
0.01 % Tween 20

The unmodified G-CSF protein was expressed in *E. coli*.

5. STORAGE

For economy of use, it is recommended that the solution be sub divided into several small aliquots and stored at -40°C or below. Avoid repeated thawing/freezing. 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

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

Dissolve the total contents of the ampoule in 1.0ml of sterile distilled water. This solution will contain PEG-G-CSF at a concentration of 10,000 International Units/ml. Use carrier protein where extensive dilution is required.

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 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. Once reconstituted, diluted or aliquoted, users should determine the stability of the material according to their own method of preparation, storage and use. Users who have data supporting any deterioration in the characteristics of any reference preparation are encouraged to contact NIBSC.

9. REFERENCES

This standard was produced under WHO guidelines cited in the WHO Technical Reports Series, No. 932, 2006, Annex 2.

Report on a Collaborative study for proposed 1st International standard for PEGylated G-CSF (PEG-G-CSF) WHO/BS/2013.2218

http://www.who.int/biologicals/expert_committee/BS_2218_1st_Inter_standard_PEGylated_G-CSF.pdf

10. ACKNOWLEDGEMENTS

We are thankful to manufacturers for their generous donations of PEG-G-CSF preparations used in the collaborative study, and to the study participants for their contributions in evaluating the preparations.

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

Physical and Chemical properties	
Classification in accordance with Directive 2000/54/EC, Regulation (EC) No 1272/2008: Not applicable or not Physical appearance: Freeze dried powder	Corrosive: No
	Stable: Yes
Hygroscopic: No	Oxidising: No
Flammable: No	Irritant: No
Other (specify):	Handling: See caution, Section 2
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 sufficient to be classed as originating from the country of supply, for example a change of freeze-drying.

Net weight: 4.6g

Toxicity Statement: Toxicity not assessed

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 compliant with the instructions for use. The reference materials are established according to the WHO Recommendations for the preparation, characterization and establishment of international reference materials for biological reference materials. http://www.who.int/bloodproducts/publications/TRS932Annex2_Inter_biolestandards.pdf (revised 2004). They are officially endorsed by the WHO Expert Committee on International Standardization (ECBS) based on the report of the international collaborative study which has established their suitability for the intended use.