

**1st INTERNATIONAL STANDARD FOR ACTIVATED FACTOR IX (FIXa), HUMAN
ESTABLISHED 1999**

NIBSC code: 97/562

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1. THE STANDARD

The 1st International Standard for Activated Factor IX (FIXa), Human, coded 97/562 consists of ampoules containing aliquots of a freeze-dried purified human FIXa prepared from activated recombinant human Factor IX. This preparation was established as the 1st International Standard for Activated Factor IX, Human, by the Expert Committee on Biological Standardisation of the World Health Organisation in October 1999

2. POTENCY

The potency of the 1st International Standard for FIXa was calibrated by 9 laboratories from 8 different countries against the NIBSC reference preparation, 92/720 by chromogenic and clotting methods specific for FIXa. The assigned potency of this preparation is

11.0 IU/ampoule.

The unit of FIXa as defined by this standard is not identical to a unit of purified Factor IX when fully activated. Studies are in progress to determine the exact relationship between the unit of FIXa as defined by this standard and the International Unit of Factor IX.

3. CAUTION

THIS PREPARATION IS NOT FOR ADMINISTRATION TO HUMANS.

The recombinant Factor IX used for preparation of the 1st International Standard for FIXa was derived from Chinese hamster ovary cells. The clinical grade human albumin used for the formulation of the standard was manufactured from plasma tested and found negative for HBsAg, anti-HCV and anti-HIV 1/2. The final product has been tested and found negative for HCV RNA by PCR. However, as with all preparations of biological origin, this material cannot be assumed to be free from infectious agents and 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 probably will include the wearing of protective gloves and avoiding the generation of aerosols. Care should be exercised in opening ampoules to avoid cuts.

4. DIRECTIONS FOR OPENING THE AMPOULE

Tap the ampoule gently to collect the material at the bottom (label) 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 the first finger of the other end.
Apply a bending force to open the ampoule.

Care should be taken to avoid cuts and projectile glass fragments that might enter one's eyes. 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.

5. BULK MATERIAL AND FILLING

Thirty-one ml of frozen activated recombinant human Factor IX (activated by Factor XIa) were thawed at 37°C and then diluted with approximately 3.5 litres of 0.05M Tris, 0.15M NaCl, 5 mg/ml Trehalose, 1.25% human albumin, pH 7.4. The solution was distributed at 4°C into 3500 ampoules, coded 97/562. The mean residual moisture content was 0.025%. The mean weight of liquid content of 69 check weight ampoules was 1.0072g, with coefficient of variation 0.15%. The contents of the ampoules were then freeze-dried under the conditions normally used for international biological standards¹.

6. STORAGE AND RECONSTITUTION

Unopened ampoules should be stored in the dark at or below -20°C. Allow ampoules to warm to room temperature. Open ampoule, taking care to ensure that all material is in the lower part, and reconstitute with 1.0ml distilled water. The reconstituted Standard should be used as soon as possible.

7. STABILITY

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.

Accelerated degradation study of 97/562 showed that after 10 months storage, when compared with samples stored at -70°C, no detectable sign of degradation was observed in samples stored at 37°C or below. Degradation of 97/562 will be monitored continually.

Studies on stability after reconstitution showed that no detectable sign of degradation was observed in samples stored on melting ice for 4 hours. However, when stored at room temperature (+22°C) after reconstitution, up to 20% and 25 % loss of activity were observed after 2 and 4 hours respectively. It is recommended that once reconstituted, the standard should be kept on melting ice, in plastic tube for no longer than 4 hours.

8. CITATION

In all publications including data sheets in which this material is referenced, it is important that the WHO status of the preparation, specified by the title of the preparation, the name and address of the WHO International Laboratory for Biological Standards at NIBSC and the NIBSC code number are cited and cited correctly.

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10. ACKNOWLEDGEMENTS ARE MADE TO

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11. REFERENCES

1. Campbell PJ. Procedures used for the production of biological standards and reference preparations. *J Biol Standardisation*. 1974, 2, 259-267.

12. MATERIAL SAFETY SHEET

Physical properties (at room temperature)			
Physical appearance	Freeze dried solid		
Fire hazard	None		
Chemical properties			
Stable	Yes	Corrosive:	No
Hygroscopic	Yes	Oxidising:	No
Flammable	No	Irritant:	No
Other (specify)	Contains recombinant and human plasma derived material		
Handling:	See caution		
Toxicological properties			
Effects of inhalation:	No adverse effects have been reported for this material		
Effects of ingestion:	No adverse effects have been reported for this material		
Effects of skin absorption:	No adverse effects have been reported for this material		
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. Seek medical advice		
Action on Spillage and Method of Disposal			
Spillage of ampoule contents should be taken up with absorbent material wetted with a virucidal agent. Rinse area with a virucidal agent followed by water.			
Absorbent materials used to treat spillage should be treated as biologically hazardous waste.			