



Intended Use

Diagen Chromatographed Bovine Factor Xa is used, in the main, for measuring the concentration of anti Xa⁽¹⁾ or heparin^(2, 3) in human plasma. However, it can be useful in protein engineering due to its highly specific proteolytic activity. It is also used to measure direct Xa inhibitors (Xabans) as well as other applications in research.

Summary and Principle

Factor Xa is an endoprotease, formed by the activation of Factor X, by both factor IX (with its cofactor, factor VIII in a complex known as intrinsic Xase) and factor VII with its cofactor, tissue factor (a complex known as extrinsic Xase). It is therefore the first member of the final common pathway or thrombin pathway. Factor Xa activity converts Prothrombin into Thrombin, and is an essential protein in blood clotting. It acts by cleaving prothrombin in two places (an arg-thr and then an arg-ile bond), which yields the active thrombin. This process is optimized when factor Xa is complexed with activated co-factor V in the prothrombinase complex. Factor Xa will also cleave any peptide bond preceded by isoleucine-glutamic acid-glycine-arginine (Ile-Glu-Gly-Arg) or isoleucine-aspartic acid-glycine-arginine (Ile-Asp-Gly-Arg), unless proline occupies the P₁ site (the site after the cleavage position).⁽⁴⁾

Reagent

Chromatographed Bovine Factor Xa **6 or 10 vials**
Lyophilised, Chromatographed bovine factor Xa, stabilised with albumin. For reconstitution, remove the cap and rubber stopper and add the appropriate volume of distilled water (0.5 mL or 5.0 mL). Swirl gently and let stand undisturbed for 10 minutes, and then transfer to a stoppered plastic tube.

Warnings and precautions

Please refer to Diagen Chromatographed Factor Xa SDS for further safety and handling information. Reagents containing animal by-products should be treated as potentially infectious. All wastes containing biological material should be properly labeled and stored separately from other waste. Waste materials should be disposed of according to prescribed international, national and local regulations.

Suggestions for Use

Materials Provided

Cat. No.

AFXX300 – Chromatographed Bovine Factor Xa (10 x 0.5 mL).
AFXX301 – Chromatographed Bovine Factor Xa (6 x 5.0 mL).

Activity of Factor Xa (Bovine)

The activity of Factor Xa (Bovine) may be measured using the synthetic tetrapeptide S-2222⁽⁴⁾. This tetrapeptide contains the specified amino acid sequence with *p*-nitroanilide attached to the arginine. Factor Xa (Bovine) hydrolyzes S-2222 to release free *p*-nitroaniline, which is measured at 405 nm. The assay is performed at 37°C and the change in absorbance at 405 nm is recorded.

Diagen Bovine Factor Xa is standardised against I.C.T.H. Reference 75/595.

Lot No XA73 22.0 units/mL
Lot No XA76 22.0 units/mL

Performance characteristics

Studies have shown that this product will perform as described prior to its expiration date when storage directions are followed. Values stated on the vial are lot specific and vary from one lot to another.

Storage and stability

The unopened freeze dried vials are **best stored deep frozen**, but may be stored for up to 3 years at 2 - 8°C without deterioration. Once reconstituted the contents of the vial are then stable for up to 8 hours when held at 2 - 8°C. Although the product may be stored deep frozen at -20°C, to minimize activity loss, avoid repeated cycles of freezing and thawing.

Packaging

10 x 0.5 mL, 6 x 5.0 mL vials.

References

1. Biggs, R., Denson, K.W.E., Akman, N., Borrett, R. and Haddon, M. Antithrombin III, Antifactor Xa and Heparin. *Brit. J. Haemat.* **19**, 3 (1970).
2. Yin, E.T., Wessler, S., Butler, J.V. and Cole, S. Plasma Heparin: A Unique Practical, Submicrogram Sensitive Assay. *J. Lab. Clin. Med.* **81**, 298 (1973).
3. Denson, K.W.E., and Bonnar, J. The measurement of Heparin. A method based on the Potentiation of Anti-Factor Xa. *Thromb. Diath. Haem.* **30**, 471 (1973).
4. Aurell, L., *et al.* (1977). A new sensitive and highly specific chromogenic peptide substrate for Factor Xa. *Thrombosis Res.* **11**: 595-609.

Key guide to symbols

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|  | Manufacturers catalogue number. |  | Consult instructions for use. |
|  | Manufacturers batch number. |  | Requires reconstitution. |
|  | For <i>in vitro</i> diagnostic use only. |  | Product expiry date. |
|  | Biological risks. |  | Store at 4°C or below. Best stored deep frozen. |



Manufacturer.

Diagnostic Reagents Ltd.
Thame
Oxon, OX9 3NY
UK
Tel: +44(0)1844 212426, Fax: +44(0)1844 216162
Email: sales@diagen.co.uk, Website: www.diagen.co.uk