

THROMBINEX[®]

BOVINE THROMBIN

For Evaluation of Plasma Coagulation

PRODUCT DESCRIPTION

Thrombinex is bovine thrombin and stabilizing buffer. One vial of Thrombinex provides 12-20 units of thrombin activity and is reconstituted to a working concentration of 6-10 units/mL.

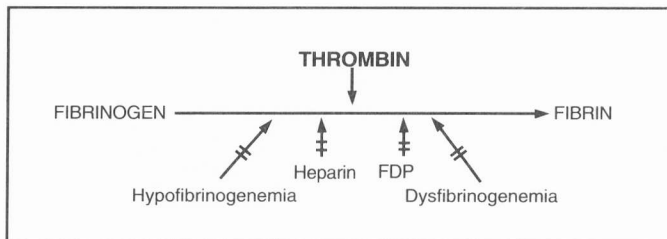
When Thrombinex is reconstituted and used according to directions, normal plasma will clot in approximately fifteen (15) to twenty-two (22) seconds.

INTENDED USE

Thrombinex is for use in thrombin time testing for the evaluation of disseminated intravascular coagulation (DIC), quantitative and qualitative fibrinogen abnormalities, heparin therapy, and streptokinase or urokinase therapy.

PRINCIPLE

When added to citrated plasma, thrombin causes the conversion of fibrinogen to fibrin! Extension of the time required for fibrin formation is an indication of hypofibrinogenemia, dysfibrinogenemia, the presence of circulating fibrin degradation products (FDP), or heparin!¹⁻³



—||— Inhibition of fibrin formation

PRECAUTIONS

Thrombinex is FOR IN VITRO DIAGNOSTIC USE ONLY and is NOT FOR INGESTION, INJECTION OR TOPICAL APPLICATION.

MATERIAL PROVIDED

Thrombinex (Bovine Thrombin) 20 x 2.0mL, 200 determinations. Store at 2° - 8°C prior to reconstitution.

MATERIALS REQUIRED BUT NOT PROVIDED

1. Purified water (distilled, deionized or reagent grade), pH = 5.3 - 7.2
2. Pipettes (2.0mL and 0.2mL volume capacities)
3. Normal citrated plasma (see QUALITY CONTROL)

INSTRUMENTATION

Thrombin time endpoints may be detected by manual methods or with any automated or semi-automated coagulation timer. Follow the manufacturer's instructions for operating the timer in use.

COLLECTION AND PREPARATION OF TEST PLASMA

Test plasma for thrombin times should be prepared from citrated whole blood. The minimum sample volume required for each duplicate test is 0.4mL (0.2mL per test).

A. Blood Collection

Blood collection for the thrombin time, as for any coagulation test, should be performed with care to avoid hemolysis or contamination by tissue fluids.

1. Syringe technique:
Draw blood. Mix immediately with 0.11M sodium citrate by adding 9 parts whole blood to 1 part anticoagulant. Invert gently to mix.⁸
2. Vacuum blood collection tube technique:
Draw blood using tubes containing sodium citrate anticoagulant. Invert gently to mix.

B. Centrifuge blood at 2500 x g for 7 minutes.⁷

C. Remove plasma from cells being careful not to disturb the buffy coat. Plasma should be free of red cells and platelets.

D. If testing is delayed, refrigerate the plasma at 2° - 8°C for a maximum of 4 hours. Beyond 4 hours, freeze the plasma at -20°C or lower for up to 8 weeks.

RECONSTITUTION

1. Tap the vial to dislodge material adhering to the stopper.
2. Remove the aluminum seal by lifting the plastic safety cap.
3. Remove the stopper and reconstitute the vial contents with 2.0mL purified water.
4. Replace the stopper (see RESULTS). Allow 10 minutes for rehydration. Invert gently to ensure complete mixing.

Each reconstituted Thrombinex vial provides sufficient materials for 10 determinations and is stable for 72 hours when stored in its covered plastic container at 2° - 8°C. BRING RECONSTITUTED THROMBINEX TO ROOM TEMPERATURE FOR TESTING.

TEST PROCEDURE

NOTE THE FINAL VOLUME OF THE TEST MIXTURE IS 0.4mL. IF NECESSARY, ADJUST COAGULATION INSTRUMENT TO ACCOMMODATE THIS VOLUME.

1. Pipette 0.2mL of a normal control plasma into a test cuvette (see QUALITY CONTROL).
2. Incubate for 2 minutes at 37°C.
3. Add 0.2mL of reconstituted Thrombinex, simultaneously starting the timer.
4. Record the clotting time.
5. Repeat Steps 1 through 4 for a duplicate sample. Duplicate results should correlate with \pm 5%.
6. Repeat Steps 1 through 5 for each test plasma, substituting the test plasma for the normal control plasma in Step 1.

QUALITY CONTROL

1. Performance of a thrombin time on a normal control plasma is necessary to provide a reference point for interpretation of patient test results. This control may be prepared by pooling fresh citrated plasma obtained from several normal donors, dividing the pool into plastic test tubes, and freezing at -20°C or lower for up to eight weeks.

An acceptable thrombin time range should be established by each laboratory for a normal control plasma. Once established, thrombin times performed with this control must fall within the acceptable range to indicate optimal thrombin concentration.

2. A plasma with a known abnormal thrombin time (low fibrinogen, heparin, elevated FDP), used as a test plasma, will verify the sensitivity of the test system. Such plasmas are commercially available (see PRODUCT AVAILABILITY).

RESULTS

The clotting time of the test plasma is compared to the clotting time of the normal control plasma. A normal thrombin time is within \pm 2 seconds of the normal control plasma value.

Thrombin time results will vary with the coagulation timing system in use. Thrombinex, used as indicated under TEST PROCEDURE, generally yields normal thrombin times within a range of 15 to 22 seconds. To obtain shorter thrombin time result, Thrombinex may be reconstituted with 1.0mL to 1.5mL purified water (distilled, deionized or reagent grade), pH = 5.3 - 7.2

EXPECTED VALUES

A test plasma with a thrombin time more than 2 seconds beyond the normal control plasma time is considered abnormal and requires further investigation. Abnormal results may be caused by a low fibrinogen concentration, the presence of an abnormal fibrinogen molecule (dysfibrinogenemia), heparin, or high concentrations of fibrin degradation products!¹⁻³

When the thrombin time is used to monitor thrombolytic therapy with drugs such as streptokinase or urokinase, a thrombin time of 2.0 to 5.0 times the normal control value is indicative of the lytic state.^{4,5}

CORRECTION STUDIES

If the thrombin time of the test plasma is prolonged, correction studies with fresh normal plasma may provide additional diagnostic information.

Mix 0.1mL of test plasma and 0.1mL of fresh normal plasma in a test cuvette. Repeat Steps 2 through 5 under TEST PROCEDURE.

Interpretation (see Table):

- A. Complete correction (thrombin time within ± 2.0 seconds of the normal control plasma value) will be observed if the abnormal time is due to a low fibrinogen concentration (less than 100mg/dL)⁶.
- B. No correction or partial correction will be observed in the presence of heparin or heparin-like circulating anticoagulants, or in cases of increased fibrinolysis. Performance of a Reptilase[®] time (see PRODUCT AVAILABILITY) will differentiate between the effects of heparin and increased fibrinolysis.¹

Table EXPECTED RESULTS

RESULTS SPECIMEN	THROMBIN TIME	CORRECTION STUDIES	REPTILASE TIME
Hypofibrinogenemia	+	Normal	++
Dysfibrinogenemia	++	Normal to ++	+++
FDP	++	++	++
Heparin	++++	+++	Normal

+ = slight prolongation of the clotting time

++++ = marked prolongation of the clotting time

LIMITATIONS

Incorrect ratios of blood to anticoagulant may result in spurious test results. The quantity of sodium citrate added to blood must be proportionally decreased in patients with hematocrit values above 53 and increased in patients with hematocrit values below 25.

As thrombin time results are extremely sensitive to variations in thrombin concentration, it is necessary that test interpretation always be made in reference to a normal control plasma.

Traumatic venipunctures and heparin contamination will yield erroneous results.

PERFORMANCE CHARACTERISTICS

Thrombinex has been tested on hypofibrinogenemic plasma (less than 100 mg/dL), dysfibrinogenemic plasma, heparinized plasma, and plasmas with elevated levels of fibrin(ogen) degradation products. Studies have shown that Thrombinex is sufficiently sensitive to these conditions.

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PRODUCT AVAILABILITY

PRODUCT	CATALOG NUMBER	NET CONTENTS
Thrombinex [®] (Bovine thrombin)	101628	20 x 2.0mL
ALSO AVAILABLE		
Plastinex [®] , Thromboplastin Reagent	101158 102672	20 x 4.0mL 15 x 10.0mL
Cephalinex [®] , APTT Reagent (Silica Activated)	101162 102677	20 x 3.0mL 15 x 10.0mL
Coagulation Control Plasma		
Citrex [®] I (Normal)	101166	20 x 1.0mL
Citrex II (Abnormal)	101170	20 x 1.0mL
Citrex III (Abnormal)	101174	20 x 1.0mL
Citrex H (Heparin Control)	102682	20 x 1.0mL
Low Fibrinogen Control	101303	5 x 1.0mL
Reptilase [®] -PC (plus control)	101623	10 test
Thrombolytic Control Plasma	101625	5 x 1.0mL
Calcium Chloride Solution, 0.025M	100989	1 x 473mL

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Printed in U.S.A.

Technical Bulletin No. 101629 Rev. H