STA® - THROMBIN

Determination of the Thrombin Time

STA® - THROMBIN ②

- Kit Containing 12 x 2-ml Vials

(REF 00611)

STA® - THROMBIN ⑩

- Kit Containing 12 x 10-ml Vials

(REF 00669) IVD CE

September 2013 *English 2*

1/ INTENDED USE

The **STA® - Thrombin** kits provide a reagent for the determination of the thrombin time (TT) with STA-R® and STA Compact®.
(In the USA this procedure has been assigned to the moderate complexity category per CLIA 1988 - CDC Analyte Code 6105; CDC Test System Codes 4677 and 4875).

2/ SUMMARY AND EXPLANATION

- The thrombin time is a rapid and simple test designed for the assessment of fibrin formation. The thrombin time remains normal in deficiencies of factor XIII (fibrin stabilizing factor) (1, 2).
  Thrombin time should first be performed before any another specific assays are attempted, when a prolongation of the overall tests (PT, APTT) cannot be explained.
- Prolongation of the thrombin time indicates (1, 2):
  – an abnormality of fibrinogen
  ◊ qualitative: dysfibrinogenaemia
  ◊ quantitative: severe hypofibrinogenaemia or congenital afibrinogenaemia
  acquired hypofibrinogenaemia (DIC, fibrinolysis, liver diseases)
  – the presence of antithrombins
  ◊ therapeutic: heparin, hirudin, argatroban...
  ◊ abnormal: FDP
  appearing during myelomas...

3/ TEST PRINCIPLE

In the presence of a predetermined quantity of thrombin, a normal plasma will consistently clot in a finite time.

4/ KIT REAGENT

An Assay Value insert with a barcode is provided in the box. This barcode contains the following information: lot number, kit code number, reagent code number and expiration date.

The **STA® - Thrombin** reagent contains titrated calcium thrombin (human), approximately 1.5 NIH unit/ml, lyophilized.

WARNING - POTENTIAL BIOHAZARDOUS MATERIAL
The reagent provided in this kit contains materials of human and/or animal origin. Whenever human plasma is required for the preparation of this reagent, approved methods are used to test the plasma for the antibodies to HIV 1, HIV 2 and HCV, and for hepatitis B surface antigen, and results are found to be negative. However, no test method can offer complete assurance that infectious agents are absent. Therefore, users of reagents of these types must exercise extreme care in full compliance with safety precautions in the manipulation of these biological materials as if they were infectious.

5/ CAUTION

Store at 2-8 °C. For *in vitro* diagnostic use only. This reagent is to be used only by certified medical laboratory personnel authorized by the laboratory.
The STA® - Thrombin kits are designed for use with analyzers of the STA® line suitable with this reagent. Read the Reference Manual of the analyzer model carefully before starting.
Exercise great care in the handling of this reagent and of patient samples. The disposal of waste materials must be carried out according to current local regulations.
In the USA, wherever appropriate, observe CLIA-88 requirements.

6/ SPECIMEN COLLECTION AND TREATMENT

Sample collection must be in conformity with the recommendations for haemostasis tests.

- Blood (9 vol.) is collected in 0.109 M (i.e., 3.2 %) trisodium citrate anticoagulant (1 vol.) (in the USA follow CLSI guidelines H3-A6 and H21-A5).
- Centrifugation: 15 minutes at 2,500 g.
- Plasma storage: 8 hours at 20 ± 5 °C.
  If on heparin therapy, plasmas remain stable for 2 hours at 20 ± 5 °C when collected with citrate anticoagulant and for 4 hours at 20 ± 5 °C when collected with CTAD tubes.

7/ REAGENT PREPARATION AND STORAGE

- **Preparation**
  Reconstitute the reagent as indicated below:
  – each vial of STA® - Thrombin ② (REF 00611) with 2 ml of distilled water. Allow the reconstituted material to stand at room temperature (18-25 °C) for 30 minutes. Then, mix reagent well by swirling the vial without creating any bubbles before use.
  – each vial of STA® - Thrombin ⑩ (REF 00669) with 10 ml of distilled water. Allow the reconstituted material to stand at room temperature (18-25 °C) for 30 minutes. Then, mix reagent well by swirling the vial without creating any bubbles, place in it a new STA® - maxi Reducer (REF 00801) and install the perforated plastic cap before use.
- **Storage**
  The reagents in intact vials are stable until the expiration date indicated on the box label, when stored at 2-8 °C.
  Once reconstituted:
  – the STA® - Thrombin ② vial is stable for 8 hours on STA-R® and STA Compact®.
  – the STA® - Thrombin ⑩ vial with STA® - maxi Reducer and perforated cap in place is stable for 7 days on STA-R® and STA Compact®.

8/ REAGENTS AND EQUIPMENT REQUIRED BUT NOT PROVIDED

- **STA® - Coag Control** N (REF 00679), **STA® - Coag Control** N + ABN* (REF 00676), **STA® - System Control** N (REF 00678) or **STA® - Routine QC 2 ml** N** (REF 00554).
- STA-R® or STA Compact®.
- STA® - maxi Reducer (REF 00801) in case STA® - Thrombin ⑩ is used.
- Common clinical laboratory equipment and materials.

* Available in the United States only.

** Not available in the United States.

9/ PROCEDURE

9.1. Patients' Plasmas
Patients' plasmas are tested undiluted. They are loaded in the instrument (see the Reference Manual of the analyzer model).
Then select the test(s) to be performed.

9.2. Quality Control
It is necessary to run controls in order to ensure accuracy and reproducibility of the results. Controls should be included in each work-shift. Use either STA® - Coag Control N + ABN, STA® - System Control N or STA® - Routine QC 2 ml N. Prepare the control(s) and scan the information contained in the barcode printed in their respective Assay Value insert(s) to the instrument. Either control is used undiluted.

9.3. Assay
Refer to the "Standardized Operating Procedures" of the instrument for full details on how to proceed from this point.
The determination of the thrombin time of the plasmas to be tested is automatically carried out by the analyzer as soon as the samples have been loaded.

10/ RESULTS

The thrombin time value (seconds) of the plasmas being tested is displayed in the "Test Panel/Test Status" screen of the instrument (see the Reference Manual). The result is to be interpreted according to the patient's clinical and biological states.

Ensure that the value obtained for the control is within the range stated in the Assay Value insert provided in the control box. If the control value is outside the stated range, check all components of the test system to ensure that all are functioning correctly, i.e., assay conditions, reagents, integrity of the plasmas being tested, etc. If necessary, repeat the tests.

11/ LIMITATIONS

- Do not test samples that have partially clotted (micro-clots).
- Do not test samples that may have been contamined by heparin (in collection tubes, syringes, etc.).

12/ REFERENCE INTERVAL

Normal values are < 21 seconds. It is recommended, however, that each laboratory determine its own normal values based on test results obtained with its normal population.
For example, 55 normal human plasmas were tested with the STA® - Thrombin reagent on the STA® analyzer; the mean time observed was 16.4 seconds with a standard deviation of 0.98 second (the clotting times observed in 98 % of the cases were less than 18.5 seconds).

13/ PERFORMANCE CHARACTERISTICS

Normal and abnormal samples were used for the intra-assay and inter-assay reproducibility studies on the STA®. Results obtained are shown below:

	Intra-Assay Reproducibility		Inter-Assay Reproducibility	
Sample	Sample 1	Sample 2	Sample 3	Sample 4
n	21	21	10	10
$\bar{X}$ (s)	19.1	32.2	17.9	33.4
SD (s)	0.53	0.55	0.29	1.09
CV (%)	2.8	1.7	1.6	3.3

14/ ALTERNATIVE PROTOCOL

The chapters 1, 2, 3, 4, 5, 6, 11 and 12 above are still valid if the thrombin time is to be determined by means other than the analyzers of the STA® line.

14.1. Reagent Preparation and Storage
The reagent preparation of either STA® - Thrombin ② or STA® - Thrombin ⑩ is identical with that indicated in chapter 7 of this insert. However, in this case, do not place either STA® - maxi Reducer or the perforated cap on the STA® - Thrombin ⑩ reagent vial.
Once reconstituted, the reagent of both STA® - Thrombin ② and STA® - Thrombin ⑩ remains stable for 2 days at 20 ± 5 °C and 7 days at 2-8 °C in its original vial.

14.2. Reagent and Equipment Required but not Provided
- **Coag Control** N (REF 00621) or **System Control** N (REF 00617).
- Instrument such as ST art®.
- Common clinical laboratory equipment and materials.

14.3. Patients' Plasmas and Control
Plasmas to be tested and control are used undiluted.

14.4. Assay
The STA® - Thrombin reagent is kept at room temperature (18-25 °C) before use.

In a glass test tube at 37 °C:	
• Plasma (patient's or control)	200 µl
• Incubate at 37 °C for	2 min.
• Starting a stop-watch, add STA® - Thrombin	200 µl
Mix. Note the clotting time (seconds).	

14.5. Results
Note the clotting times (seconds) of the plasmas being tested and that of the control.
Ensure that the value obtained for the control is within the range stated in the Assay Value insert provided in the control box. If the control value is outside the stated range, check all components of the test system to ensure that all are functioning correctly, i.e., assay conditions, reagents, integrity of the plasmas being tested, etc. If necessary, repeat the tests.

14.6. Performance Characteristics
Normal and abnormal samples were used for the intra-assay and inter-assay reproducibility studies on the KC10. Results obtained are shown below:

	Intra-Assay Reproducibility		Inter-Assay Reproducibility	
Sample	Sample 1	Sample 2	Sample 3	Sample 4
n	21	20	10	10
$\bar{X}$ (s)	18.2	32.8	19.1	36.2
SD (s)	0.64	1.30	0.45	1.41
CV (%)	3.5	4.0	2.3	3.9

REFERENCES

1. CAEN J., LARRIEU M.-J., SAMAMA M.:
   "L'hémostase. Méthodes d'exploration et diagnostic pratique". Paris: L'Expansion scientifique, 208-209, 348-351, 1975.
2. SAMAMA M., CONARD J., HORELLOU M.H., LECOMPTE T.:
   "Physiologie et exploration de l'hémostase". Paris: Doin, 155-156, 1990.

Significant changes are indicated by dotted lines in the margin.

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