Methodist Health Services Corporation &
UnityPoint Health Methodist|Proctor
Department of Pathology
Peoria, IL 61636

Effective Date: January 29, 2017
NEW

Protime
Sta Compact Max

I. PRINCIPLE

The **STA® - Neoplastine® CI Plus** Plus kit provides reagents for the determination of the prothrombin time (PT) in plasma. The principle of the test consists of the use of calcium thromboplastin to measure the clotting time of the patient's plasma and to compare it with that of a normal standard. The test measures, as a whole, the activity of the coagulation factor II (prothrombin), factor V (proaccelerin), factor VII (proconvertin), factor X (Stuart factor) and factor I (fibrinogen).

II. CLINICAL SIGNIFICANCE

A prolonged PT has been observed in the following clinical states:

A. congenital or acquired deficiencies of factor II, V, VII, X or fibrinogen
B. liver failure (cirrhosis, hepatitis)
C. treatments with vitamin K antagonists
D. hypovitaminosis K: nutritional intake deficiency, disorders in absorption or metabolism of vitamin K (hemorrhagic disease of the newborn, cholestasis, treatment with antibiotics)
E. fibrinolysis
F. DIC

The PT is commonly used for monitoring vitamin K antagonist therapy (3) because of its sensitivity to variations in the concentration of the vitamin-K dependent factors II, VII and X. Consequently, the comparability of results of this test is essential for finding the therapeutic range.
It is well known that the PT value of a plasma may vary according to the origin of the thromboplastin reagent and to the instrument used to measure it. In this system the PT ratio is converted into the International Normalized Ratio (INR). The INR value corresponds to the value of the ratio of the patient's PT to that of the standard PT raised to the ISI (International Sensitivity Index) power of the thromboplastin used:

$$\mathbf{INR} = \left(\text{Patient's PT} / \text{Geometric Mean PT}\right)^{\mathbf{ISI}}$$

The use of the INR is recommended for the assessment of the vitamin K antagonist therapy in patients

III. SPECIMEN TYPE:

Mix nine parts of freshly collected blood with one part of 0.11 mol/L (3.2%) sodium citrate anticoagulant. Invert the tube gently three or four times immediately after venipuncture to ensure proper mixing of blood an anticoagulant. A syringe or evacuated tubes (blue top) may be used with caution for collection. If multiple specimens are collected, the coagulation sample should be the second or third tube

collected. If blood is drawn from an indwelling catheter, the line should be flushed with 5.0 mL saline and the first 5 mL of blood or six dead space volumes of the catheter discarded or used for other laboratory tests. The citrate concentration must be adjusted in patients who have hematocrit values above 55%. Specimens that are clotted, collected in the wrong tube, have visible hemolysis or have less than a 90% fill should be rejected.

IV. HANDLING/STORAGE CONDITIONS:

The whole blood specimen is checked for clot formation by gentle inversion and observation. Centrifuge the capped blood specimen as soon as possible after collection for 4 Min at 4000 RPM or at a speed and time required to produce platelet-poor plasma (platelet count <10x10^9/L). The plasma may remain on the packed cells if testing immediately or separated if freezing. To separate plasma, use a plastic transfer pipette; remove the plasma to a polypropylene/plastic tube until ready to test. If testing is not complete within 24 hours, the plasma must be removed to a polypropylene/plastic tube and frozen. A frost-free freezer should not be used. Frozen plasma samples must be rapidly thawed at 37°C while gently mixing and tested immediately after thawing. If testing is delayed, the sample may be held for 24 hours at room temperature. Specimens should be stored on board the analyzer or at room temperature after testing. Once removed from the analyzer, caps must be removed if additional testing is ordered.

V. REAGENTS

A. STA® - Neoplastine® CI Plus kit

1. **Reagent 1:** STA® - Neoplastine® CI Plus , lyophilized thromboplastin prepared from fresh rabbit cerebral tissue. The ISI value of STA® - Neoplastine® CI Plus, correlated with a secondary standard of the RBT (rabbit brain thromboplastin) by instruments of the STA® line, is indicated on the Assay Value insert provided in the box.
2. **Reagent 2:** solvent containing calcium 10-ml per vial.
3. P**reparation:** Transfer the entire contents of one vial of Reagent 2 into one vial of Reagent 1 of the same kit. Allow the reconstituted reagent to stand at room temperature (18-25 °C) for 30 minutes. Swirl the Reagent 1 vial gently to obtain a homogeneous suspension. Then, add a stirring-bar to the vial, place a new STA®- Maxi Reducer and install the perforated cap
4. **Storage:** The reagents in intact vials are stable until the expiration date indicated on the box label, when stored at 2-8 °C.
   a. Once reconstituted, Reagent 1 is stable: 48 hours on board
   b. add the stir-bar, STA® - Reducer and put the perforated plastic cap in place
   c. **Do not freeze.**

B. STA® - Coag Control N + ABN kit: provides a normal plasma and an abnormal plasma intended for the quality control of the following tests on analyzers of the STA® brand name suitable with these reagents: prothrombin time (PT), activated partial thromboplastin time (APTT), fibrinogen.

C. Preparation: Reconstitute each vial of Reagent 1 or 2 with exactly 1 ml of fresh reagent grade Nerl water. Allow the reconstituted material to stand at room temperature (18-25 °C) for 30 minutes. Then, swirl the vial gently 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, Reagents 1 and 2 remain stable for 8 hours.

D. STA – DESORB U is a decontaminating solution for use with the STA Compact® It is designed as an integral part of the STA® analyzer system.

Preparation: Install a new STA® - maxi Reducer) and the perforated cap on a freshly opened bottle of STA® - Desorb U before loading it into the analyzer.

1. N.B.: a fine white sediment may be observed in the bottom of the bottle; this has no effect on the performance of the product. on STA Compact® model, place one bottle in the product drawer
2. **Storage:** The reagent in intact bottles is stable until the expiration date indicated on the box label, when stored at 2-8 °C and protected from light.
    a. Once opened, the STA® - Desorb U with STA® - maxi Reducer and perforated cap in place, is stable for:5 days on board STA Compact®

The STA® - Desorb U reagent contains KOH, a corrosive chemical at the concentration provided (< 1 %).

Danger:

a. Causes severe skin burns and eye damage.
b. Wear protective gloves/protective clothing/eye protection/face protection.
c. IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower.
d. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

CAUTION:

1. Store reagent kit at 2-8 °C.
2. For in vitro diagnostic use only.
3. The stirring-bar used in the reagent vial should never be the source of contamination. To ensure that stirring-bars are contamination-free, rinse the bars with distilled water and dry them carefully to remove all traces of moisture before adding them to reagent vials. In addition, decontaminate stirring-bars once a week according to the following procedure:
4. immerse the stirring bars in a vial of STA® - Desorb U and let them soak. Pour through sieve into sink and run water over them to rinse off. Thoroughly dry them and put them away.

VI. INSTRUMENTATION/EQUIPMENT

STA-R®, STA Compact® or STA Satellite®
Stirring-bar
STA® - mini Reducer or STA® - maxi Reducer
Cuvette roll – 1000
Centrifuge
Distilled Water
Pipettes & tips

VII. CALIBRATION

The ISI value for the Prothrombin time must be the value indicated on the insert included in the STA® line product. The operator must check the ISI value before leaving the menu if there has been a lot change, a software update, or any other major change. An incorrect ISI value can lead to inaccurate INR (International Normalization Ratio) results. See analyzer Reference Manual. Entering or Modifying the ISI ratio and/ or the reference time (geometric mean): Refer to START-

UP or Reference Manual for further information.

VIII. QUALITY CONTROL

STA®- Coag Control N + Abn. It is necessary to run controls in order to ensure accuracy and reproducibility of the results. Two different levels of control should be used. Prepare the control reagents and scan the information contained in the barcode printed on their respective Assay Value insert to the instrument.

IX. PROCEDURE:

A. Refer to START-UP/Operating procedure for the analyzer before running patient and QC specimens at the start of each shift.

X. CALCULATIONS:

A. The International Normalized Ratio (INR) is calculated by STA Compact® and when INR is selected as a reporting unit in Methodologies.

$$INR = (\text{Patient's PT/Mean Normal PT})^{ISI}$$

B. 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, expiration date, calibration values and ISI value. The ISI appears in the Calibration screen for the PT.

C. To enter or change the ISI ratio: see StartUp procedure calibration section.

XI. REFERENCE INTERVAL

Normal values vary from one laboratory to the next, depending on reagents, instrumentation and technique. So, each laboratory must determine its own expected values of the normal healthy adult population (16) based on technique and instrumentation in use.
If PT-INRs are reported, a lot specific ISI value and Reference Time or *geometric mean* must be entered into the instrument. The ISI is automatically entered into the analyzer when the new lot of reagent is first bar coded and loaded onto the instrument.
With each new lot of PT reagent, the Reference Time or *geometric mean* must be manually entered into the analyzer in order for the correct calculation of the INR. Validation of the normal range geometric mean must be confirmed onsite with each new lot of reagent.

XII. REPORTING RESULTS:

Report results using interface/manual result entry in the LIS system.
Reference interval for UPH-Methodist for Protimes: **11.5-14.4 seconds**

A. Procedure for Abnormal Results:

Critical Value: PT > 4.5 INR
Refer to the critical values policy for how to handle critical values.

B. **Analytical Measurement Range: 10-120 seconds. Results below 10 seconds should be turned out as <10 and those above 120 seconds as >120.** Results that are outside of the

validation range (>max <min) and have a blue flag will need to be "confirmed" on the analyzer.
These do not cross into the LIS system until validated in coag expert.

To validate the result in coag expert;
coag expert→workstation→dashboard→uncheck everything except "to be validated"→double click on the patient→click the validation mode button in the upper right corner→click in the box to the left of the result that needs to be validated→save.

XIII. PROCEDURAL NOTES/PROBLEM-SOLVING TIPS

A. New lot: With each new lot of **STA® - Néoplastine® CI** Plus, the operator must enter the geometric mean time (Reference Time) as described in CALIBRATION section of the StartUp procedure, before the analyzer will allow QC to be run.

B. The PT reference range and geometric mean are validated with each change of PT reagent lot number.

C. RISK OF INCORRECT RESULTS-The ISI value for the Prothrombin time must be the value indicated on the insert included in the STA® line product. The operator must check the ISI value before leaving the menu if there has been a lot change, a software update, or any other major change. An incorrect ISI value can lead to inaccurate INR (International Normalization Ratio) results.

XIV. LIMITATIONS OF THE PROCEDURE

A. **Sample:** The slightest coagulation (micro-clots) will induce considerable shortening of the times measured (autocatalytic activation of all the factors) whereas extensive coagulation will prolong the clotting times because of consumption of factors and fibrinogen.

Do not keep plasmas at 2-8 °C because in this temperature range the factor VII may be activated by the kallikrein system (2).

B. **Anticoagulant:** Maintain the correct anticoagulant/blood sample volume ratio of 1:9. If there is any considerable variation in hematocrit, modify the quantity of anticoagulant accordingly.

C. **Heparins:** The STA® - Néoplastine® CI Plus test is insensitive to unfractionated heparin levels up to 1 IU/ml and to low molecular weight heparin levels up to 1.5 anti-Xa IU/ml.

D. **Thrombin Inhibitors:** Thrombin inhibitors (e.g., hirudin, argatroban...) present in the sample to be tested may lead to a prolonged prothrombin time for this sample.

E. **Vitamin K antagonists:** Vitamin K antagonists will depress plasma levels of factors II (prothrombin), VII (proconvertin), X (Stuart factor) and IX (antihemophilic factor B).For the assessment of the vitamin K antagonist therapy, refer to the current recommendations.

XIV. REFERENCES

A. STA Compact Max® Reference Manual June 2016.
B. STA Compact Max® User Guide November 2015.
C. STA Compact Max® Software version 106.08.01.00

For additional information, please refer to the current manufacturer's package inserts.

MMCI Laboratory is a CAP accredited facility, as of 7/1/11 the responsibility of new and/or substantially revised policies and procedures will be restricted the Laboratory Director whose name appears on the CLIA certificate, whose signature appears below. The biennial review will be completed by the Administrative Director.

POLICY CREATION :

Author: *Kim Paige, CLT* *December 20, 2016*

Medical Director: *January 29, 2017*

Elizabeth A. Bauer Marsh, M.D.

MEDICAL DIRECTOR		
DATE	NAME	SIGNATURE
January 29, 2017	Elizabeth A. Bauer-Marsh	
SECTION MEDICAL DIRECTOR		
January 26, 2017	Julia Adams, MD	

REVISION HISTORY			
Rev	**Description of Change**	**Author**	**Effective Date**
0	Initial Release	Kim Paige	12/20/16

Lead	Date	Coordinator	Date	Asst. Manager	Date	Medical Director	Date
Kim Paige	1/17/17		1/20/17		1/17/17		1/20/17