



Manufactured By: HYPHEN BioMed

# BIOPHEN AT anti-IIA ON STA-R

Assay of AT anti-IIa activity on STAR



## 1. Reconstitution of Biophen AT anti-IIa (#A221122) reagents.

	NAME	Reconstitution	Stability*	T° Stabilization
R1	thrombin	2.5 ml of distilled water *	Refer insert	** 30 mn on board before any use
R2	Substrate	2.5 ml of distilled water *		
R3	Tris-Heparin buffer	Ready to use		

\*The stability data claimed on the insert were obtained on reconstituted vials, kept closed, protected from and provided any contamination or evaporation is avoided. Stability must be controlled, and can be adjusted and validated if required, according to the exact use conditions for each laboratory.

**Reconstitution:** (\*) After reconstitution with distilled water, let the reagent to stabilize for 30 minutes at room temperature (18-25°C).

**Stabilization of reagents:** (\*\*) It is necessary to let the reagent temperature to stabilize for at least 30 minutes on the automate board before any use.

**Storage of reagents:** Take care of putting up the specific caps back on the bottles before storing them at 2°-8° C, and of strictly respecting the temperature stabilization time of 30 minutes before using the reagents on the automate.

If the reagents are kept on the automate board, take care and use reducers to limit as much as possible any evaporation of the reagents.

Homogenize the reagents before each use.

Any reagent of biological origin must be handled with all the required cautions, as being potentially infectious.

**Do not interchange the reagents from different lots.**

### **Reagents required but not provided:**

- Plasma Calibrator titrated for AT (eg: **Biophen Plasma Calibrator #A222101**), or **international or internal reference material**.
- Normal and Abnormal quality control plasmas titrated for AT (eg: **BIOPHEN Normal Control Plasma #A223201** and **BIOPHEN Abnormal Control Plasma #A223301**).
- Distilled water, preferentially sterile.



## **2. Preparation of the calibration curve and controls/samples.**

- **Calibration curve:**

- Calibration is performed with normal pooled citrated plasma with the assigned value of 100% AT. The assay includes a standard plasma dilution of **1:40 (managed by the automate)**. By definition, this later dilution of the pool represents the **100% AT** activity. The calibration curve is from 0 to 100% AT.

**Or**

-Calibration is performed with a commercially available plasma calibrator, with a known AT Concentration "C" (**eg Biophen Plasma Calibrator #A222101**). The 1:40 dilution (managed by the automate) corresponds to the indicated AT concentration (C).

- **Tested plasma and controls:**

In these conditions, tested plasmas and controls are loaded "undiluted" and assayed at the 1:40 dilution (managed by the STAR).

Controls: The control is performed with commercially available control plasmas, titrated for AT. Various control plasma are available: **Biophen Normal Control Plasma (#A223201)**, and **Biophen Abnormal Control Plasma (#A223301)**

Nota : For lyophilized calibrators and controls, following reconstitution with distilled water, let the reagent to stabilize 30 minutes at room temperature. It is recommended to run the calibration curve with a freshly reconstituted calibrator. It is necessary to let the reagent temperature to stabilize for at least 30 minutes onto the automate before any use. Take care avoiding any contamination or evaporation of the reagents. Stability can be adjusted according to the exact use conditions.

**Homogenize before each use.**

**Do not freeze calibrators and quality control plasmas.**

**Quality controls must be run regularly, and for each new batch of reagents, after an important maintenance of the instrument, or if measured values are not in compliance with the one expected for the method.**

**Performances may present slight variations according to the instrument used. Validate the expected values in the exact laboratory working conditions.**

**Check the validity of the series by including quality control plasmas at different levels in each one.**

## **3. Results:**

- The calibration curve (working range) is of the Lin (OD/min) – Lin (concentration, AT%) type.
- The values obtained for patients and controls are directly calculated from the calibration curve (when the standard 1:40 dilution is used for the test).
- The results are expressed as %AT activity. (100 % being equivalent to 1 IU/ml)

**The calibration curve is validated when linearity (r2), as well as measured control values, are in compliance.**

**A new calibration curve must be carried out for each new batch of reagents, after each important maintenance of the instrument, or when measured values for controls are out of the acceptance range for the method (after checking all other parameters for the system).**

**Performances may present variations according to the instrument used, and to its adjustment. Validate the expected values in the laboratory working conditions. Performances, as well as values for each new lot of quality controls used, must then be confirmed (and adjusted if necessary) in the exact laboratory working conditions.**

#### 4. Programming the STA-R analyser:

Click on the icon **set up software** for the **manager program** and create the program according to:

This calibration allows measuring concentrations in the range of about ... -....% of Factor ....

TESTS						Configuration	
<b>Method</b>							
IDENTIFICATION							
Abbreviation				ATalla		Last up date	
Name		ATalla		Method		Colo-Kinetics OD /min	
SAMPLE			DILUENT				
Vol. µl	Incu.	Dilution	Id.	Name		Stab. h	Continued
140 µl	0 sec	1/40	R3	R3 ATalla buffer		(*)	
REAGENTS							
	Id.	Name		Stab. h	Vol. µl	Incu. sec	Prec.
Ra	R1	R1 ATalla Thrombin		(*)	70	60	<input checked="" type="checkbox"/>
Rb							<input type="checkbox"/>
Rc							<input type="checkbox"/>
Rd	R2	R2 ATalla Substrat		(*)	70		

Enter data concerning washing

REAGENTS									
	Id.	Name		Stab. h	Vol. µl	Incu. sec	Prec.	Vial. ml	Min. Vol ml
Diluent	R3	R3 ATalla buffer		(*)				25	1.5
Ra	R1	R1 ATalla Thrombin		(*)	70	60	<input checked="" type="checkbox"/>	3	0.5
Rb							<input type="checkbox"/>		
Rc							<input type="checkbox"/>		
Rd	R2	R2 ATalla Substrat		(*)	70			3	0.5
Washing									
	Ra		Rb		Rc		Rd		
Before	Non						Non		
After	Special						intensive		
Wash	12227						12227		
Name	STA-DESORB U						STA-DESORB U		
Stab. H	120						120		
Vial ml	15						15		
Vol in. ml	0.9						0.9		

(\*) To be filled by the user, according to the instructions of the insert, and to the exact use conditions.

Click on the icon **Result software** for the **manager program** and create the program according to:

TESTS		Configuration							
		<b>Result</b>							
METHOD		RESULT							
Chro KINETICS OD/mn		Primary Unit	%						
Linearity Min.	0.95	Correction factor	1.00						
First Point	10	Single/Duplicate							
Second Point	60	<input checked="" type="radio"/> Single <input type="radio"/> Duplicate							
		Precision (%)	5.00						
		Redilution	Condition						
		1/	<						
		1/	>						
<input checked="" type="checkbox"/> <b>Rd Heating</b>		<table border="1"> <thead> <tr> <th colspan="2">VALIDATION %</th> </tr> <tr> <th>Min.</th> <th>Max.</th> </tr> </thead> <tbody> <tr> <td>0*</td> <td>120*</td> </tr> </tbody> </table>		VALIDATION %		Min.	Max.	0*	120*
VALIDATION %									
Min.	Max.								
0*	120*								

Write calibration data.

TESTS		Configuration	
		<b>Calibration</b>	
MODE :		Linear regression	Determination
			<input type="checkbox"/> Single <input checked="" type="checkbox"/> Duplicate
ASSAYS		Concentrations	
SCALE :	<input checked="" type="checkbox"/> Linear <input type="checkbox"/> Log	<input checked="" type="checkbox"/> Linear <input type="checkbox"/> Inverse <input type="checkbox"/> Log	
	Id.	Key	Name
Std 1	CAL		CAL
Std 2	CAL		CAL
Std 3	CAL		CAL
Std 4	0**		Cal 0 (R3 buffer)
Ctrl, Niv. 1			
Ctrl, Niv. 2			

\*According to the calibrator used (homogenize before each use).  
 Note\*\*: Calibrator 4 is "R3 buffer" alone.

Click on the icon **Printout/Transmission software** for the **manager program** and create the program according to:

TESTS						Configuration
					Printout/Trans.	
<b>PARAMETERS</b>						
	Units	Factor Convers	Print	Transmission Test numbers	Usual Values Min. Max.	
Main	%		v	0	80*	120*
Aux1	DO/min		v	0		
Aux2						
Aux3						
<b>Printout Limits</b>						
Min.	<input type="text" value="0"/>	Max.	<input type="text" value="150"/>			

(\*) to be adjusted by the user.

Show the last page and enter data concerning the quality control.

TESTS						Configuration
					Q.C	
<b>LEVEL 1</b>						
	Id.	Key	Name	Stab. h	Vial. ml	Min Vol ml
Control	(*)		(*)	*	*	*
<b>LEVEL 2</b>						
	Id.	Key	Name	Stab. h	Vial. ml	Min Vol ml
Control	(*)		(*)	*	*	*
<b>LEVEL 3</b>						
	Id.	Key	Name	Stab. h	Vial. ml	Min Vol ml
Control				*	*	*
Period : <input type="text"/> hours <input type="text"/> Tests <input type="text"/> Vial						

\*According to the controls used (homogenize before each use).

Example of calibration curve obtained:

% AT	OD/min
100	0.60
50	1.37
25	1.76
0	2.17
$r^2$ ( lin - lin )	0.994