AMERICAN BIOCHEMICAL & PHARMACEUTICALS LTD.

REF ABP-TRA-1
1 X 5.0mg
TRAP-6-amide

IVD

RESEARCH USE ONLY

abp
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PRODUCT DESCRIPTION

TRAP-6-amide is a synthetic hexapeptide. (SFLLRN-NH_2) that mimics the action of thrombin. Trap-6 activates the thrombin receptors PAR-1 and PAR-4.[1,2,3,4] It is not inhibited by P2 Y_{12} anti-platelet agents. TRAP-6 is sensitive to GP II_b III_a antagonists.[1-4]

TEST PRINCIPLE

Light Transmission Aggregometry

LTA is the reference method for the measurement of platelet function. The dynamic range is established by the difference in light transmission between Platelet Poor Plamsa (PPP) which sets the 0% baseline and Platelet Rich Plasma (PRP) which sets the 100% limit. The PRP is maintained at 37°C and stirred at about 1,000 rpm. When an agonist is added to stimulate the PRP, platelets aggregate, allowing an increase in the amount of light passing through the PRP to increase compared to the PPP baseline. The change in light transmission is a measure of the platelet response to an agonist.

When added to normal Platelet Rich Plasma, or certain other platelet preparations, TRAP-6 reagent elicits a strong, monophasic response in a concentration dependent manner.[7,8,9]

MATERIALS PROVIDED

A single vial containing 5.0 mg of TRAP-6-amide powder. Its molecular weight is 747.90.

MATERIALS REQUIRED BUT NOT PROVIDED

1. Platelet Aggregometer
2. Preservative Free physiologic saline (0.85 or 0.90%)
3. Pipette and tips
4. Sample tubes and caps
5. Siliconized Aggregometer Cuvettes
6. Plastic coated micro stir bars

REAGENT STORAGE

TRAP-6-amide may be transported at ambient temperature but should not be exposed to prolonged periods of high temperatures. Once received, TRAP-6 should be storted at -15° to -20°C or colder temperatures.

Reconstituted TRAP-6 may be stored for extended periods at -80°C

RECONSTITUTION

Frozen TRAP-6 reagent MUST BE WARMED to room temperature prior to use. Reconstituted reagent should be kept at 2°-8°C while not in use.

Reconstitute the vial of TRAP-6 with preservative free, physiologic saline (0.85 or 0.9%).

FREQUENTLY USED CONCENTRATIONS

Working Concentration	1 mg TRAP-6	5mg TRAP-6	10mg TRAP-6
	Diluent Vol.	Diluent Vol.	Diluent Vol.
1.0mM	1.3353 mL	6.6767 mL	13.3535 mL
5.0mM	0.2671 mL	1.3353 mL	2.6707 mL
10.0mM	0.1335 mL	0.6677 mL	1.3353 mL

REAGENT DISPOSAL

Unused TRAP-6 must be disposed of as a hazardous waste in accordance with local regulations and laboratory policy.[9]

PROFESSIONAL LABORATORY USE ONLY

INSTRUMENTATION

TRAP-6 reagent will perform as described when used on most Light Transmission Aggregometers.

Follow the aggregometer manufacturer's INSTRUCTIONS for USE and sample size and agonist volume requirements.

PATIENT PREPARATION[5,7,8]

1. Clinical, medication, family and social histories required prior to testing.
2. Patients should refrain from taking aspirin or other anti-platelet medications for 7-10 days, or as directed by their physician.
3. Patients should avoid supplements, herbal preparations, energy drinks or other products known to affect platelet function.
4. Patients should avoid fatty meals and food products prior to specimen collection.

SPECIMEN COLLECTION

Refer to the current CLSI Approved Guideline H58-A: Platelet Function Testing by Aggregometry for detailed specimen collection and sample preparation instructions and related references.[7]

EVACUATED SPECIMEN COLLECTION TUBE TECHNIQUE (PREFERRED) [7,8,9,10]

1. Use a 21 or 23 gage winged needle set specifically labeled for specimen collection use
2. Remove the tourniquet as soon as blood starts to flow.
3. Collect the blood specimen in 2.7uL plastic evacuated specimen collection tubes containing 0.105/0.11M (2.3%) buffered sodium citrate anticoagulant.
4. Gently invert each tube 4-5 times to assure complete mixing.
5. Maintain specimens at room temperature without removing the caps.
6. Observe Standard Precautions throughout the specimen collection process and follow appropriate laboratory policies for post phlebotomy patient care and disposal of sharps and supplies.
7. Evacuated specimen collection tubes with light blue tops may contain 3.2% or 3.8% sodium citrate. Check the label for the proper concentration.[3,6,7,9]
8. Underfilled tubes should be rejected.
9. Blood collection should be performed with care to avoid patient anxiety, stasis, hemolysis and contamination by tissue fluid, or any exposure to glass.
10. Make sure the winged needle set is intended for phlebotomy use.
11. Each of the following can cause test results to be inaccurate.
    a. Visible RBC contamination
    b. Hemolysis
    c. Icterus
    d. Lipemia
    e. Clots

These are unacceptable specimens and should be rejected.

12. Test results may also be affected if the patient has thrombocytopenia (the threshold is agonist and analyzer dependent) or hypofibrinogenemia. Follow laboratory policies when such specimens have been collected.
13. If the patient's hematocrit is less than 30% or greater than 55%, the blood to anticoagulant ratio must be adjusted. (see H58-A for instructions).
14. Specimens must be tested within four hours of collection.

QUALITY CONTROL

Test samples from a drug free, known donor should be run along with the test samples to confirm system performance.

LTA TEST PROCEDURE

1. Place the appropriate number of test cuvettes in to the incubation wells.
2. Add a new, plastic coated stir bar to each cuvette.
3. Prepare the PPP blank by pipetting 0.250 mL of PPP in to a cuvette. DO NOT PLACE A STIR BAR IN THE BLANK TUBE.
4. Pipette 0.225 mL of the patient sample PRP into each pre-warmed test cuvette for each patient to be tested.
5. Place the PRP test samples for a pre-set incubation period and temperature (37°C).
6. Set the 100% baseline by placing the blank into the test well.
    a. Press the Blank Button.
    b. Remove the Blank from the test well.
7. Place the PRP sample cuvette into the test well.

a. Press the start button.

8. Add 0.25uL of the agonist/reagent into the PRP using the proper pipette and tip to assure the agonist/reagent is directed into the center of the cuvette and not allowed to run down the side of the cuvette.
9. Select inject.
10. The test will run for the pre-set test time. (~6 minutes).
11. An alarm will sound when testing in all channels is completed.

WARRANTY

This product is warranted to perform to these specifications when used in accordance with the labeling. American Biochemical and Pharmaceuticals Ltd. disclaims any implied warranty of merchantability and fitness for any other purpose and in no event shall American Biochemical and Pharmaceuticals Ltd. be liable for any consequential damages arising out of the aforesaid warranty.

SELECTED REFERENCES

1. Coughlin, S.R. 'Protease-activated receptors and platelet function', Thromb. Haemost., 1999; 82:353
2. Coughlin, S.R. 'Protease-activated receptors in vascular biology', Thromb. Haemost., 2001; 86:298
3. Vassallo, R.R. Jnr., Kieber-Emmons, T., Cichowski, K. et. al. 'Structure-function relationships in the activation of platelet thrombin receptors by receptor-derived peptides' j. Biol. Chem., 1992; 267:6081
4. Andersen, H., Greenberg, D.L., Fujikawa, K. et. al. 'Protease-activated receptor is the primary mediator of thrombin-stimulated platelet procoagulant activity.' Proc. Natl. Acad. Sci. USA, 1996; 96:11189
5. Dacie & Lewis, 'Practical Haematology', Lewis, S.M., Bain, B.J. and Bates, I. (Editors); 9th Edition, Elsevier Science Ltd., 2002 pages 383-385.
6.Scarborough, RM et. al. J. Bio Chem. 267, 13146-1992.
7. Clinical and Laboratory Standards Institute (CLSI). Platelet Function Testing by Aggregometry; Approved Guidelines. CLSI document H58-A (ISBN 1-56238-683-2), Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500, Wayne, Pennsylvania 19087 USA. 2008
8. Clinical and Laboratory Standards Institute (CLSI), Collection, Transport, and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays and Molecular Hemostasis Assays; Approved Guidelines - Fifth Edition. CLSI document H21-A5 (ISBN 1/56238-657-3) Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500, Wayne, Pennsylvania 19087 USA. 2008
9. Siegel, J.D., Rhinehart, E., Jackson, M., Chiarello, L., The Healthcare Practices Advisory Committee, 2007, Guideline for Isolation Precautions Preventing Transmission of Infectious Agents in Healthcare Settings; www.cdc.gov/infectioncontrol/guidelinesisolation/index.
10. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers From Occupationally Acquired Infections; Approved Guidelines - Fourth Edition. CLSI document M29-A4 (ISBN 1-56238-961-0) [Print]; ISBN 1-56238-962-9 [Electronic]). Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500, Wayne, Pennsylvania 19087 USA, 2014

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