

tPA assay kit

Human tPA total antigen ELISA Kit

CATALOG #
AAA71823

PRODUCT SYNONYMS

tPA; Human tPA total antigen ELISA Kit; Total Human tPA Assay; tPA assay kit

PRODUCT SPECIFICATIONS

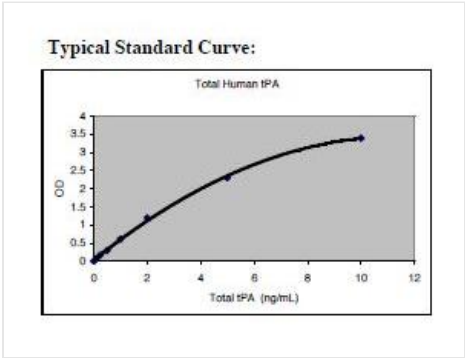
Sequence Length	19
Standard Preparation	The standard curve can be made in either 3% BSA Blocking Buffer or Depleted Plasma. If your samples are serum or plasma, the Depleted Plasma should be reconstituted at 1.0 mL per vial and used as the diluent. All other samples should use 3% BSA Blocking Buffer as the diluent To prepare standard, reconstitute with 1.0 mL of 3% BSA Blocking Buffer or Depleted Plasma depending on your sample type. Prepare an initial dilution by adding 50 uL of 1,000 ng/mL stock with 150 uL of diluent. Standard tPA Concentration (ng/mL) Amount of Diluent (L) Transfer Volume (L) Transfer Source Final Volume (L) S7 25 450 50 250 ng/mL Stock 300 S6 10 300 200 S7 250 S5 5 250 250 S6 300 S4 2 300 200 S5 250 S3 1 250 250 S4 250 S2 0.5 250 250 S3 300 S1 0.2 300 200 S2 500 S0 0.0 250 --- --- 250
Sample Collection, Storage and Preparation	Samples should be collected using trisodium citrate, acidified citrate or Stabilitetm (DiaPharma) collection media. Collection should be in accordance with the collection vials manufacturers instructions or in a 1:10 ratio of collection media to blood. Immediately, upon collection of blood, the samples should be centrifuged at 3000 x g. This should ensure the removal of platelets as they can release PAI-1 that in turn complexes with uPA. The plasma can be transferred to a clean plastic tube and stored frozen for up to one month. Samples are stable for approximately 5 hours when stored at 4 degree C with the Sabilytetm media. Note: Detergents such as Triton X cause interference with the assay. If using detergent extracted samples, it is necessary to dialyze the samples overnight to remove the detergent.
Procedural Notes	1. This assay should be run at room temperature. 2. Use aseptic technique when opening and dispensing reagents. 3. To minimize error due to handling, wipe the exterior bottom of the microplate wells with a lint-free paper towel.

Reagent Preparation	1. 10x Wash Buffer: Dilute the 50 mL of concentrate to 1x with 450 mL of DI water prior to use. 2. TBS Buffer: 0.1 M Tris, 0.15 M NaCl, pH 7. 4. 3. 3% BSA Blocking Buffer: 3% BSA in TBS Buffer. 4. Primary Antibody: Reconstitute with 10 mL of 3% BSA Blocking Buffer as directed on the vial and vortex gently to mix. Prepare immediately prior to use. 5. Secondary Antibody: Dilute with 10 mL of 3% BSA Blocking Buffer as directed on the vial and vortex gently to mix. Prepare immediately prior to use.
Preparation and Storage	1. Store the kit and all of its components at 4 degree C before use. 2. If not using the entire plate at once, prepare only the appropriate amount of Primary Antibody and PAI-1 Standard. The remaining stock solutions should be frozen and stored at At -70C. Primary Antibody should be used within two weeks. All other components should remain refrigerated. 3. Store unused portions of the microplate in a pouch with a desiccant at 4 degree C.

IMAGES

Scheme I:

	1	2	3	4	5	6	7	8	9	10	11	12
A	S7	S7	U1	U1	U9	U9	U17	U17	U25	U25	U33	U33
B	S6	S6	U2	U2	U10	U10	U18	U18	U26	U26	U34	U34
C	S5	S5	U3	U3	U11	U11	U19	U19	U27	U27	U35	U35
D	S4	S4	U4	U4	U12	U12	U20	U20	U28	U28	U36	U36
E	S3	S3	U5	U5	U13	U13	U21	U21	U29	U29	U37	U37
F	S2	S2	U6	U6	U14	U14	U22	U22	U30	U30	U38	U38
G	S1	S1	U7	U7	U15	U15	U23	U23	U31	U31	U39	U39
H	B0	B0	U8	U8	U16	U16	U24	U24	U32	U32	U40	U40



Application Data

Application Data

ADDITIONAL INFORMATION

Related Product Information for tPA assay kit	Introduction: Tissue-type Plasminogen Activator (tPA) is a member of the serine proteinase family. tPA functions to lyse fibrin clots into soluble plasmin fragments. tPA is active in two forms, single chain and two chain. The two-chain tPA is created via interaction with the plasmin product cleaving the single chain. This twochain form is regarded as the more active form. Both single chain and two-chain tPA are complexable with PAI-1. PAI-1 acts as an inhibitor for tPA by binding to the tPA and thus stifling its ability to lyse fibrin. tPA can serve as an indicator of both myocardial infarction for patients with impaired fibrinolytic systems as well as a marker for type-II diabetes. Principle of assay: This is an ELISA (Enzyme-Linked Immunosorbent Assay) for the quantitative analysis of tPA levels in biological fluid. This test kit operates on the basis of sandwich ELISA where free, latent and complexed tPA is quantified with the use of an HRP labeled secondary antibody. The various forms of tPA present in the standard or unknown is captured by the tPA capture antibody coated on the well. A primary antibody specific for tPA is then added to each well followed by the HRP conjugated secondary antibody. The bound conjugated secondary antibody is detected by the addition of substrate, which generates an optimal color after 10 minutes. Quantitative test results may be obtained by the measure and comparison of the sample and standard absorbance readings when read with a microplate reader at 450 nm.
References	1. Declerck, P., et al., (1995) Thromb Haemosts. Nov:74(5):1305-1309 2. Eitzman, D., et al., (2000) Blood 95(2):577-580 3. Kawasaki, T., et al. (2000) Blood 96(1):153-160 4. Declerck, P., et al., (1988) Blood 71(1):220-225 5. Schafer, K., et al., (2001) FASEB 15:1840-1842 6. Samad, F., et al., (1996) Proc Natl Acad Sci USA 96(12):6902-6907

NCBI AND UNIPROT INFORMATION

NCBI GI #	2285954
NCBI GeneID	5327
UniProt Accession #	Q6LBF5; P00750
Molecular Weight	2,342 Da
NCBI Official Full Name	tPA, partial
NCBI Official Synonym Full Names	plasminogen activator, tissue
NCBI Official Symbol	PLAT
NCBI Official Synonym Symbols	TPA; T-PA
NCBI Protein Information	tissue-type plasminogen activator; alteplase; plasminogen activator, tissue type; plasminogen/activator kringle; reteplase; t-plasminogen activator; tissue plasminogen activator (t-PA)
UniProt Protein Name	TPA protein
UniProt Gene Name	tPA
UniProt Entry Name	Q6LBF5_HUMAN

For Research Use Only. Not for use in diagnostic or therapeutic procedures. Small volumes of product may become entrapped in the seal of the vial during shipment and storage; if necessary, briefly centrifuge the vial on a tabletop centrifuge to dislodge any liquid in the cap.

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Online datasheet: <https://stage.aaabiotech.com/assay-kits/tpa/71823>