Human Tissue Plasminogen Activator ELISA Kit
ab190812

Overview

Product name: Human Tissue Plasminogen Activator ELISA Kit
Detection method: Colorimetric

Precision

<table>
<thead>
<tr>
<th>Sample</th>
<th>n</th>
<th>Mean</th>
<th>SD</th>
<th>CV%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Human serum</td>
<td>5</td>
<td></td>
<td></td>
<td>3.7%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Sample</th>
<th>n</th>
<th>Mean</th>
<th>SD</th>
<th>CV%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Human serum</td>
<td>3</td>
<td></td>
<td></td>
<td>10.5%</td>
</tr>
</tbody>
</table>

Sample type: Cell culture supernatant, Serum, Hep Plasma, EDTA Plasma, Cit plasma
Assay type: Sandwich (quantitative)

Sensitivity: 3.5 pg/ml
Range: 78 pg/ml - 5000 pg/ml

Recovery

<table>
<thead>
<tr>
<th>Sample type</th>
<th>Average %</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
<td>104</td>
<td>100% - 107%</td>
</tr>
<tr>
<td>Cell culture media</td>
<td>119</td>
<td>116% - 123%</td>
</tr>
<tr>
<td>Hep Plasma</td>
<td>97</td>
<td>94% - 99%</td>
</tr>
<tr>
<td>EDTA Plasma</td>
<td>84</td>
<td>82% - 86%</td>
</tr>
<tr>
<td>Cit plasma</td>
<td>99</td>
<td>97% - 107%</td>
</tr>
</tbody>
</table>
**Assay time**  
1h 30m

**Assay duration**  
One step assay

**Species reactivity**  
Reacts with: Human  
Does not react with: Goat, Cow, Pig

**Product overview**  
Human Tissue Plasminogen Activator ELISA Kit has been re-developed. We have identified new recombinant monoclonal antibodies to provide improved performance and consistency. This kit will be discontinued when stock is depleted. The new version of the kit is available as **ab282306**.

Human Tissue Plasminogen Activator ELISA Kit (ab190812) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of Tissue Plasminogen Activator protein in cell culture supernatant, cit plasma, edta plasma, hep plasma, and serum. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human Tissue Plasminogen Activator with 3.5 pg/ml sensitivity.

SimpleStep ELISA® technology employs capture antibodies conjugated to an affinity tag that is recognized by the monoclonal antibody used to coat our SimpleStep ELISA® plates. This approach to sandwich ELISA allows the formation of the antibody-analyte sandwich complex in a single step, significantly reducing assay time. See the SimpleStep ELISA® protocol summary in the image section for further details. Our SimpleStep ELISA® technology provides several benefits:

- Single-wash protocol reduces assay time to 90 minutes or less  
- High sensitivity, specificity and reproducibility from superior antibodies  
- Fully validated in biological samples  
- 96-wells plate breakable into 12 x 8 wells strips

A 384-well SimpleStep ELISA® microplate (**ab203359**) is available to use as an alternative to the 96-well microplate provided with SimpleStep ELISA® kits.

**Notes**  
Tissue Plasminogen Activator (Tissue-type plasminogen activator or tPA) is a circulating serine protease involved in the breakdown of clots. tPA converts inactive plasminogen to active plasmin; in turn plasmin degrades the fibrin matrix in clots. In addition, plasmin can cleave tPA at Arg-310 with results in a two chain disulphide linked tPA that has even greater proteolytic activity. tPA is synthesized in many tissues and is secreted into most extracellular body fluids. Recombinant tPA is used medically to resolve or prevent blood clots in ischemic stroke or myocardial infarction.

**Platform**  
Microplate

**Properties**

**Storage instructions**  
Store at +4°C. Please refer to protocols.

<table>
<thead>
<tr>
<th>Components</th>
<th>1 x 96 tests</th>
</tr>
</thead>
<tbody>
<tr>
<td>10X Human TPA Capture Antibody</td>
<td>1 x 600µl</td>
</tr>
<tr>
<td>10X Human TPA Detector Antibody</td>
<td>1 x 600µl</td>
</tr>
</tbody>
</table>
Function
Converting the abundant, but inactive, zymogen plasminogen to plasmin by hydrolyzing a single Arg-Val bond in plasminogen. By controlling plasmin-mediated proteolysis, it plays an important role in tissue remodeling and degradation, in cell migration and many other physiopathological events.

Play a direct role in facilitating neuronal migration.

Tissue specificity
Synthesized in numerous tissues (including tumors) and secreted into most extracellular body fluids, such as plasma, uterine fluid, saliva, gingival crevicular fluid, tears, seminal fluid, and milk.

Involvement in disease
Note=Increased activity of TPA results in increased fibrinolysis of fibrin blood clots that is associated with excessive bleeding. Defective release of TPA results in hypofibrinolysis that can lead to thrombosis or embolism.

Sequence similarities
Belongs to the peptidase S1 family.
Contains 1 EGF-like domain.
Contains 1 fibronectin type-I domain.
Contains 2 kringle domains.
Contains 1 peptidase S1 domain.

Domain
Both FN1 and one of the kringle domains are required for binding to fibrin.
Both FN1 and EGF-like domains are important for binding to LRP1.
The FN1 domain mediates binding to annexin A2.
The second kringle domain is implicated in binding to cytokeratin-8 and to the endothelial cell surface binding site.

Post-translational modifications
The single chain, almost fully active enzyme, can be further processed into a two-chain fully active form by a cleavage after Arg-310 catalyzed by plasmin, tissue kallikrein or factor Xa.
Differential cell-specific N-linked glycosylation gives rise to two glycoforms, type I (glycosylated at Asn-219) and type II (not glycosylated at Asn-219). The single chain type I glycoform is less readily converted into the two-chain form by plasmin, and the two-chain type I glycoform has a lower activity than the two-chain type II glycoform in the presence of fibrin.
N-glycosylation of Asn-152; the bound oligomannosidic glycan is involved in the interaction with the mannose receptor.
Characterization of O-linked glycan was studied in Bowes melanoma cell line.

Cellular localization
Secreted > extracellular space.

Images
SimpleStep ELISA technology allows the formation of the antibody-antigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.

Background-subtracted data values (mean +/- SD) are graphed.

Example of TPA standard curve

Background-subtracted data values (mean +/- SD, n = 2) are graphed.

Titration of Human serum and plasma (heparin) within the working range of the assay
The concentrations of Tissue Plasminogen Activator were measured in duplicates, interpolated from the Tissue Plasminogen Activator standard curves and corrected for sample dilution. Undiluted samples are as follows: serum 25%, plasma (citrate) 12.5%, plasma (EDTA) 25%, and plasma (heparin) 25%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean Tissue Plasminogen Activator concentration was determined to be 5.25 ng/mL in serum, 4.86 ng/mL in plasma (citrate), 4.45 ng/mL in plasma (EDTA), and 4.65 ng/mL in plasma (heparin).

Serum from 10 apparently healthy male donors was measured in duplicate. The mean TPA concentration was determined to be 2,741 pg/mL with a range of 1,836-4,012 pg/mL in male donors.
PBMC were grown in the absence or presence of phytohemagglutinin (PHA) for 2 days. TPA was measured in 4-fold diluted cell culture supernatants of unstimulated and PHA stimulated PBMC. Measured values were interpolated from the TPA Standard Curve diluted in Sample Diluent NS and DF corrected. Mean +/-SD, n=2, are graphed.

To learn more about the advantages of SimpleStep ELISA® kits see [here](#).

Sandwich ELISA - Human Tissue Plasminogen Activator ELISA Kit (ab190812)

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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