

TECHNOSCREEN ADAMTS13 Activity - English

INTENDED USE

Semiquantitative flow through assay for the determination of ADAMTS13 activity levels in human citrated plasma. This assay is intended as a screening tool for estimating ADAMTS13 activity. Deficiency of ADAMTS13 needs to be confirmed by a quantitative system.

SUMMARY

ADAMTS13 (a disintegrin-like and metalloproteinase with thrombospondin type 1 motif 13) is an enzyme (vWF-cleaving protease or vWF-CP) that specifically cleaves unusually large von Willebrand factor (vWF) multimers, which induce platelet thrombus formation under high shear stress. If the activity of ADAMTS13 is lowered for some reason, unusually large vWF multimers may accumulate within blood causing thrombosis due to platelet aggregation, which in turn may lead to TTP (thrombotic thrombocytopenic purpura).

Assay principle: Patient plasma is incubated with a vWF fragment. ADAMTS13 specific cleavage of this vWF fragment is then detected with a cleavage site specific antibody by the amount of color development. The color intensity directly correlates with the amount of cleaved substrate and thus with ADAMTS13 activity level in the plasma sample.

The TECHNOSCREEN ADAMTS13 Activity contains:

	Reagent / Content	Description
10 x 1	TECHNOSCREEN test device	Device with test and control well including membrane and absorbent paper
10 x 1	TECHNOSCREEN pre-incubation tubes	Tubes with vWF fragments, lyophilized
1 x 4 mL	TECHNOSCREEN R1	Reagent 1: Wash solution PBS and detergent; liquid, ready to use
1 x 1 mL	TECHNOSCREEN R2	Reagent 2: biotinylated anti-GST antibody; liquid, ready to use
1 x 1 mL	TECHNOSCREEN R3	Reagent 3: streptavidin coated gold particles; liquid, ready to use
11 x 1	Large pipette (300 µl)	300 µl single use pipette to transfer plasma sample into pre-incubation tube
22 x 1	Small pipette (60 µl)	60 µl single use pipette to transfer pre-incubation mixture onto test device
1 x 1	Short Instruction Pad	Short protocol including stand for pre-incubation tube
1 x 1	Reference Color Chart	Lot specific color chart with colored zones indicating four levels of ADAMTS13 activity from 0.0 – 0.8 IU/mL. For visual interpretation of results.

Warning and precautions

- RUO for research use only
- Do not mix components from different lots.
- Do not use test kits after the expiry date
- The pipettes are single use only. Do not re-use.
- All blood and plasma samples have to be regarded as potentially infectious and handled with appropriate care and in compliance with the biosafety regulations in force. Dispose the same way as hospital waste.
- A Material Safety Data Sheet for this product is available from www.technoclone.com.

Stability and storage

The expiry date printed on the labels is only applicable to storage of the unopened containers at 2...8 °C.

Stability opened/ in use:

Reagent	2...8 °C
TECHNOSCREEN test device	For immediate use
TECHNOSCREEN pre-incubation tubes	For immediate use
TECHNOSCREEN R1	Until expiry date
TECHNOSCREEN R2	Until expiry date
TECHNOSCREEN R3	Until expiry date
Reference color chart	Store in kit box (in dark)

TEST PROCEDURE

Preparation of plasma samples

Refer to CLSI Document H21-A5 or DIN 58905 for instructions on specimen collection, handling, and storage.

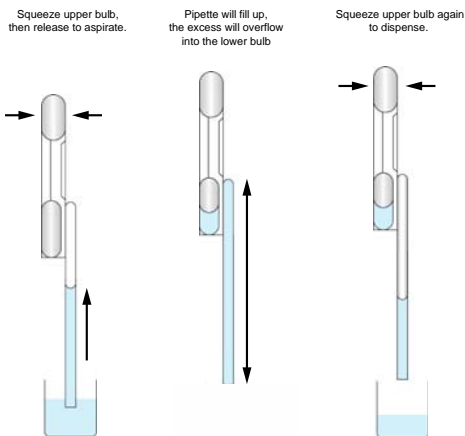
Thaw frozen samples rapidly at 37 °C and centrifuge if necessary. Gently mix before testing. After thawing, the assay must be performed within 2 hours.

Preparation of reagents

Bring pre-incubation tube and reagents to room temperature (18...25 °C) before use.

Make sure that you hold the dropper bottles vertically when dropping reagents on the test device.

Using the pipettes



Performance of the test

- Step 1 Add 300 µl of plasma sample to a fresh pre-incubation tube using the large pipette. Mix by inverting the closed tube at least 3 times. Place the pre-incubation tube into the designated stand on the short instruction pad.
- Step 2 Incubate the pre-incubation mixture (from Step 1) for 20 minutes at room temperature (18...25 °C).

For all subsequent steps, ensure that the liquid is completely absorbed before continuing with the next step.

- Step 3 Add 1 drop of reagent (R1) to both test and control well on the test device.
- Step 4 Mix the pre-incubation mixture by inverting before use. Add 60 µl of pre-incubation mixture to the test well on the test device using a small pipette. Do not squeeze the small pipettes too firmly.
- Step 5 Using a second fresh pipette add 60 µl of the pre-incubation mixture to the control well on the test device.
- Step 6 Add 1 drop of reagent (R2) to both test and control well on the test device.
- Step 7 Add 1 drop of reagent (R1) to both test and control well on the test device.
- Step 8 Add 1 drop of reagent (R3) to both test and control well on the test device.
- Step 9 Add 1 drop of reagent (R1) to both test and control well on the test device.
- Step 10 Let the test device dry for 1-2 minutes prior to interpretation. Read color development using the lot specific reference color chart within 10 minutes.

LIMITATION OF THE TEST

Samples containing EDTA can not be used. EDTA is a strong inhibitor of ADAMTS13.

Samples with excessive hemolysis, icterus or lipemia, should not be used.

Flow through of plasma should take 30 – 50 seconds. Prolonged flow through might indicate plasma interferences such as lipemia, interfering with the assay result. Please disregard the result and determine ADAMTS13 activity by a different system such as TECHNOZYM ADAMTS13 Activity ELISA.

INTERPRETATION OF RESULTS

To ensure the validity of the test, confirm that the color intensity in the control well matches or exceeds the color on the lot specific reference color chart.

For the interpretation of the test well, consult the flip side of the color chart included in the kit. Results may be expressed in % activity or IU/mL (10 % equals 0.1 IU/mL).

Normal range for ADAMTS13 Activity: **0.40 – 1.30 IU/mL** (as determined in TECHNOZYM ADAMTS13 Activity ELISA)

Deficiency of ADAMTS13 activity should be confirmed with a quantitative test system.

PERFORMANCE CHARACTERISTICS

Performance data are given below. Results obtained in individual laboratories may differ.

170 patient samples with different ADAMTS13 levels were tested both using the TECHNOSCREEN ADAMTS13 Activity Test and the TECHNOZYM ADAMTS13 Activity ELISA. Visual interpretation was performed by independent investigators. Based on a cut off of 0.1 IU/ml: positive predictive value 99 %, negative predictive value 80 %, sensitivity 94 % and specificity 97 %.

STANDARDISATION

The reference color chart is calibrated against the International Standard for ADAMTS13 in plasma (12/252).

LITERATURE

Please contact Technoclone www.technoclone.com or your local distributor.

EDITORIAL NOTE

This document is available in several languages. The translations have been done using the master document in English. In the event of doubts or discrepancies, the wording in the master document in English shall take precedence.



TECHNOSCREEN ADAMTS 13 Activity - YouTube

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