

Analytical performance validation of new screening assay for the deficiency of ADAMTS13 Activity

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INTRODUCTION

Background

Severe deficiency in ADAMTS13 activity levels (< 0.1 IU/ml) is a specific marker for thrombotic thrombocytopenic purpura (TTP). TTP is an acute life threatening disorder with the highest mortality rate within the first 24 h of hospital admission. It would be beneficial to have a quick turnaround of ADAMTS13 activity reporting. We have developed and validated a simple screening assay which allows the reporting within 1 hour of blood draw.

MATERIALS AND METHODS

40 patient plasma samples with activity levels ranging from 0 - 1.1 IU/mL were tested in the TECHNOZYM® ADAMTS13 Activity ELISA and two kit lots of the TECHNOSCREEN® ADAMTS13 Activity assay. A cut off value of 0.1 IU/mL was chosen since this reflects the value used by laboratories for classifying a TTP patient. The sample results were classified as either >0.1 IU/mL or <0.1 IU/mL in order to calculate assay performance. One sample was highly lipemic so did not flow through the test device and therefore was excluded.

Classification	TECHNOSCREEN / ELISA result	TECHNOSCREEN IU/mL	TECHNOZYM ELISA IU/mL
A	POSITIVE / POSITIVE	≥0.1	≥0.1
B	POSITIVE / NEGATIVE	≥0.1	≤0.1
C	NEGATIVE / POSITIVE	0	≥0.1
D	NEGATIVE / NEGATIVE	0	≤0.1

Tab 1. sample classification

The summed values for each sample class were placed in a 2x2 contingency table and the following standard calculations were made:

$$\text{Positive predictive value} = A / (A+B)$$

$$\text{Negative predictive value} = D / (D+C)$$

$$\text{Sensitivity} = A / (A+C)$$

$$\text{Specificity} = D / (D+B)$$

$$\text{Accuracy} = (A+D) / (A+B+C+D)$$

RESULTS

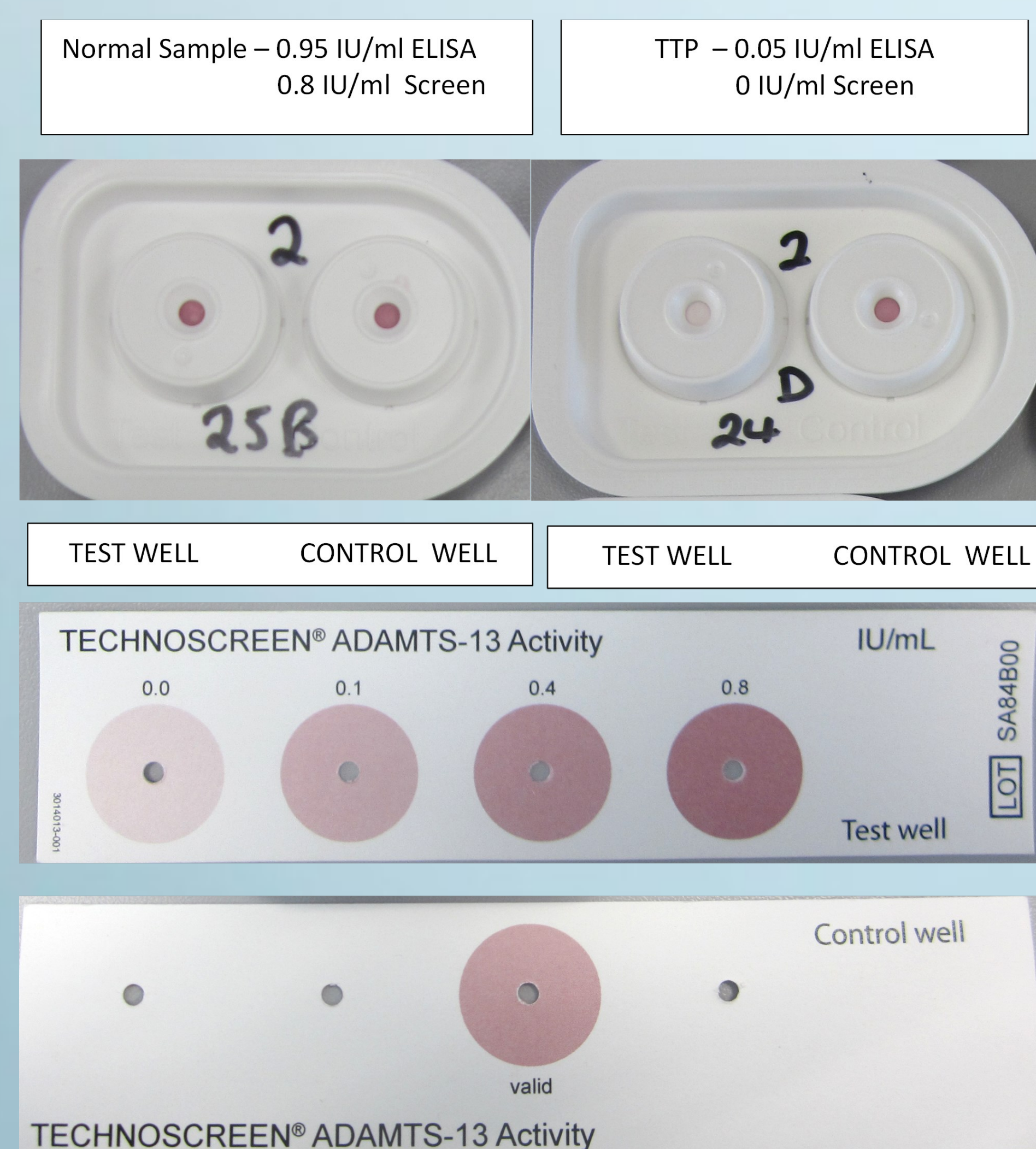


Fig 1. Examples of normal & TTP sample in TECHNOSCREEN® ADAMTS13 Activity Test

At least 3 different readers were used to interpret the results. There was excellent agreement between the different readers. The assay showed good repeatability and reproducibility. Lot to lot consistency was determined showing excellent correlation between the lots and the ELISA results.

*The samples with differing classification were all borderline samples.

For example: sample 1. SCREEN 0.1 IU/mL — ELISA 0 IU/mL

sample 2. SCREEN 0 IU/mL — ELISA 0.11 IU/mL

		TECHNOZYM ELISA	
		POSITIVE ≥0.1 IU/ml	NEGATIVE ≤0.1 IU/ml
TECHNOSCREEN Kit Lot 1	POSITIVE 0.1, 0.4 or 0.8 IU/ml	27	4*
	NEGATIVE 0 IU/ml	0	8
Positive Predictive Value		87.1%	
Negative Predictive Value		100%	
Sensitivity		100%	
Specificity		66.7%	
Accuracy		89.7%	

		TECHNOZYM ELISA	
		POSITIVE ≥0.1 IU/ml	NEGATIVE ≤0.1 IU/ml
TECHNOSCREEN Kit Lot 2	POSITIVE 0.1, 0.4 or 0.8 IU/ml	26	3*
	NEGATIVE 0 IU/ml	1*	9
Positive Predictive Value		89.7%	
Negative Predictive Value		90%	
Sensitivity		96.3%	
Specificity		75%	
Accuracy		89.7%	

CONCLUSIONS

We have demonstrated that this screening test is an excellent tool for determining deficient levels of ADAMTS13 activity with a sensitivity >96%.

- The TECHNOSCREEN ADAMTS13 Activity is:**
- Designed for testing single patient samples within 30 min.
 - Versatile as can be performed without specialized laboratory equipment
 - Ideal as a cost effective screening tool for TTP