PB0573

ANALYTICAL PERFORMANCE VALIDATION OF THE T-TAS 01 PL ASSAY

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INTRODUCTION

The T-TAS® 01 PL assay is a novel in vitro diagnostic system that passes whole blood through a collagen-coated microcapillary bed at arterial shear stress to measure the platelet thrombus formation process. The test is used to assess overall primary hemostatic function. Results are reported as the area under the pressure-time curve (AUC).

AIMS

To validate the analytical performance of the T-TAS 01 PL assay by evaluating precision, reproducibility, and analytical specificity.

METHODS

All blood samples were collected using the BAPA tube for T-TAS 01 and measured using the PL Chip for T-TAS 01.

- 1 normal donor and 2 donors taking aspirin were used for the precision study.
- 1 normal donor and 3 donors taking aspirin were used for the reproducibility study.
- 1 normal donor and 1 donor taking aspirin were used for the analytical specificity study.

Precision and reproducibility was evaluated according to CLSI EP05-A3E.

- Due to limited sample stability, the precision study was performed on a single day using three operators that tested three lots of reagents using two instruments, for a total of 12 measurements per source of variability (operator, manufacturing lot, and instrument), and 36 measurements per sample.
- The reproducibility study was performed using a 3x5x5 design (3 sites, 5 replicates per sample, 5 days).

Analytical specificity testing was performed according to CLSI EP07-A3E by preparing stock solutions for each of the compounds to be tested in either aqueous solutions or organic solvents, depending on solubility. Samples were split and received either the compound or vehicle (control). The highest level of compound that did not produce interference was determined. Compounds were determined to have no effect if there was no statistically significant difference in AUC results compared to the control sample.

RESULTS

Precision and Reproducibility:

All precision testing met the specification of CV ≤ 15% or SD ≤ 39. All three sites passed reproducibility testing on all days and for all four samples.

Sample	N	Mean	Repeatability Within-Run (SD, %CV)	Between-Operator (SD, %CV)	Between-Lot (SD, %CV)	Between-Instrument (SD, %CV)	Total (SD, %CV)
High	36	428.1	10.7, 2.5	2.0, 0.5	4.7, 1.1	1.6, 0.4	11.9, 2.8
Middle	36	237.3	31.7, 13.4	6.4, 2.7	10.5, 4.4	0.0, 0.0	34.0, 14.3
Low	36	130.7	18.4, 14.1	11.8, 9.0	13.5, 10.3	0.0, 0.0	25.7, 19.6

Reproducibility Site 1

Donor	Day 1	Day 2	Day 3	Day 4	Day 5
1	PASS	PASS	PASS	PASS	PASS
2	PASS	PASS	PASS	PASS	PASS
3	PASS	PASS	PASS	PASS	PASS
4	PASS	PASS	PASS	PASS	PASS

Reproducibility Site 2

Donor	Day 1	Day 2	Day 3	Day 4	Day 5
1	PASS	PASS	PASS	PASS	PASS
2	PASS	PASS	PASS	PASS	PASS
3	PASS	PASS	PASS	PASS	PASS
4	PASS	PASS	PASS	PASS	PASS

Reproducibility Site 3

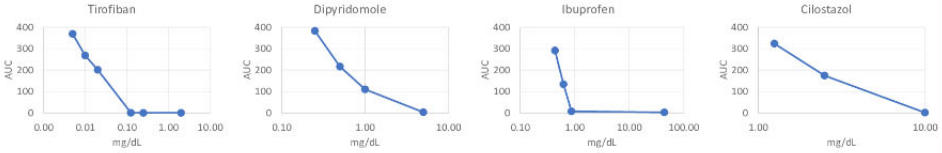
Donor	Day 1	Day 2	Day 3	Day 4	Day 5
1	PASS	PASS	PASS	PASS	PASS
2	PASS	PASS	PASS	PASS	PASS
3	PASS	PASS	PASS	PASS	PASS
4	PASS	PASS	PASS	PASS	PASS

Analytical Specificity:

Concentrations listed are the highest concentration that did not affect PL AUC results.

Compound	Class	Concentration	Compound	Class	Concentration
Acetaminophen	Analgesic	7.8 mg/dL	Heparin	Anticoagulant	525 U/mL
Bilirubin	Blood component	40 mg/dL	L-Thyroxine	Hormone	0.0858 mg/dL
Caffeine	Stimulant	21.6 mg/dL	Metformin	Antihyperglycemic	2.4 mg/dL
Captopril	ACE inhibitor	0.528 mg/dL	Omeprazole	Proton pump inhibitor	1.68 mg/dL
Catechin	Flavinol/antioxidant	5 mg/dL	Pravastatin	Statin	0.414 mg/dL
Cilostazol	Vasodilator/antiplatelet	1.25 mg/dL	Propranolol	Beta-blocker	0.202 mg/dL
Dabigatran	Anticoagulant	0.047 mg/dL	Rivaroxaban	Anticoagulant	0.044 mg/dL
Dextran 40	Plasma expander	2400 mg/dL	Streptokinase	Fibrinolytic	50,000 U/dL
Diltiazem	Calcium channel blocker	0.18 mg/dL	Theophylline	Bronchodilator	6 mg/dL
Dipyridamole	Vasodilator/antiplatelet	0.25 mg/dL	Tirofiban	Antiplatelet	N/A
Fish Oil	Dietary supplement	25.6 mg/dL	Triglycerides	Blood component	750 mg/dL
Ibuprofen	NSAID	0.438 mg/dL	Warfarin	Anticoagulant	7.5 mg/dL

Compounds known to affect platelet function produced a predictable dose-dependent decrease in PL AUC results.



CONCLUSIONS

The T-TAS 01 PL assay is highly specific for the measurement of primary hemostatic function.

- The results of precision testing demonstrate that the test meets the specification of CV ≤ 15% or SD ≤ 39 across the measurement range.
- The results of reproducibility testing demonstrate that the test performance is consistent between different testing facilities.
- The results of reproducibility testing demonstrate that the test is not affected by therapeutic levels of various common compounds, and is highly specific for primary hemostatic function
  - Dose-dependent effect of antiplatelet agents
  - No effect of compounds causing secondary hemostasis defects.

PL assay measurements may be useful for the assessment of overall primary hemostatic function in patients with bleeding tendencies, defects in primary hemostatic function, or prior to clinical procedures involving bleeding risk.

REFERENCES

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