Evaluation of The Hemostatic Ability of The New Device 'Total Thrombus Formation Analysis System' (T-TAS) for Thrombocytopenic Patients. In *vitro* effect of lyophilized human platelets

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INTRODUCTION

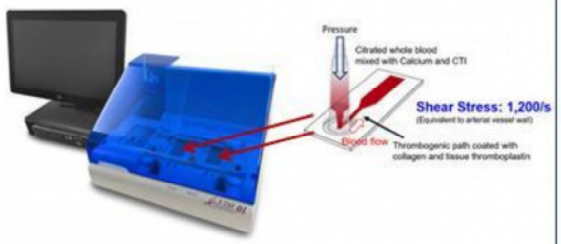
- Bleeding is a frequent complication in thrombocytopenic onco-hematological patients.
- Current laboratory approaches fail to evaluate hemostasis in thrombocytopenia and the efficacy of platelet transfusion.
- The Total Thrombus-Formation Analysis System (T-TAS® 01) is a new approach proposed to explore hemostasis in thrombocytopenia (1).
- T-TAS appears as an interesting device to explore the hemostatic effect of hemoderivatives, such as lyophilized human platelets with potential hemostatic capacity (2).

AIMS

1. **To validate T-TAS as a suitable device to evaluate hemostasis in samples from thrombocytopenic patients before and after platelet transfusion.**
2. **To explore the hemostatic efficiency of *in vitro* addition of Thrombosomes®, a human platelet-derived lyophilized hemostatic agent (pending phase 2 trial for the treatment of bleeding in thrombocytopenia).**

METHODS

- Whole blood samples from onco-hematological patients with thrombocytopenia (platelet count <30x10^3 platelets/µL) were collected before and after platelet transfusion.
- Hemostasis was evaluated in the T-TAS 01, with specific chips (HD) containing microcapillary channels (width of 300 µm; 50-µm-deep) coated with collagen and tissue factor.
- T-TAS parameters:
  - Area under the curve (**AUC**)
  - Occlusion times (**OT**, min)
- In blood samples before (**BT**) and after (**AT**) transfusion.
- Thrombosomes® were spiked ex vivo to each sample (final concentration of 50x10^3/µL).



RESULTS

T-TAS parameters showed significant improvements after platelet transfusion and after *in vitro* addition of Thrombosomes®.

The addition of LHP increased AUC and shortened OT in almost 60% of samples from non-transfused patients and in 80% of samples from transfused patients (Figure 1, Table 1) (p<0.01). Figure 2 shows a representative graphic obtained at the T-TAS before and after adding LHP to non-transfused and transfused samples based on pressure increases with respect to time in minutes.

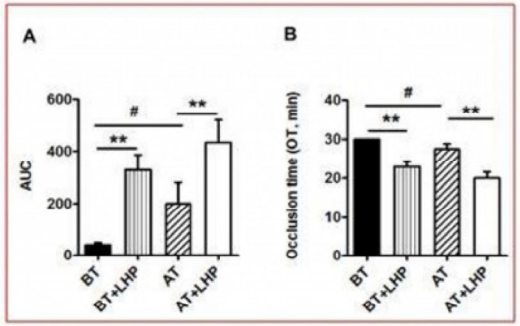


Figure 1. Bar diagrams show A) AUC; B) OT, min in samples before transfusion (BT) and after transfusion (AT) in the absence and in the presence of LHP, as indicated.

LHP= Lyophilized Human Platelets.
n=31 for BT and n=19 for AT
Mean ± SEM, **p<0.01 #p <0.05

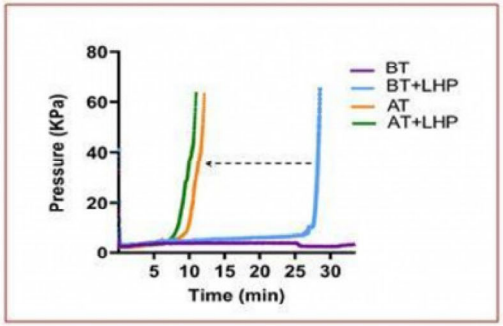


Figure 2. The linear diagram shows representative graphics obtained at the T-TAS, based on pressure per minute, corresponding to a sample from a patient before (BT) and after (AT) transfusion, in the absence and presence of LHP added in vitro.

LHP= lyophilized Human Platelets.

Table 1. T-TAS parameters (AUC and OT; Mean ± SEM) obtained in samples from thrombocytopenic patients before (n=31) and after (n=19) platelet transfusion. In *vitro* effect of LHP. BT= before transfusion, AT= after transfusion, LHP= lyophilized human platelets.

**p<0.01 for the effect of the presence of Thrombosomes® vs. their absence, in each sample
#p<0.05 after transfusion vs. before transfusion (AUC = 28 ± 2) in the n=19 patients

Mean±SEM	BT	BT+LHP	AT	AT+LHP
Area under curve (AUC)	41±8.4	330±57**	200±82#	436±83**
Occlusion time (OT, min)	29.8±0.1	23.1±1.3**	27.4±1.4#	20±1.6**
Hematocrit (HCT%)	26±0.8		27±0.6	
Platelet count (×10^3 Platelet/µL)	9.7±0.8		23.8±1.9**	

CONCLUSIONS

1. **T-TAS, using a modified microchip-based flow chamber, was able to measure the hemostatic ability in samples with thrombocytopenia.**
2. **T-TAS proved useful to evaluate the hemostatic capacity of transfused platelets.**
3. **Thrombosomes®, added *ex vivo* to samples from thrombocytopenic patients, improved their hemostatic performance in the T-TAS.**
4. **T-TAS has the potential to predict the efficiency of transfusional products.**

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