

Diagnostica di laboratorio in emostasi e trombosi

WHOLE-BLOOD THROMBIN GENERATION ASSAY REVEALS HYPERCOAGULABLE STATE IN SICKLE CELL DISEASE IN STEADY STATE AND CORRELATES WITH CLINICAL EVENTS

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Background and Aims. Sickle cell disease (SCD) is a systemic condition characterized by vaso-occlusive crises (VOC), which underlie acute complications and chronic organ damage. The pathophysiology of VOC is multifactorial and includes chronic hypercoagulability. Whole blood thrombin generation (WB-TG) with and without thrombomodulin (TM) enables global and dynamic assessment of coagulation, integrating both plasma factors and cellular components. This study aimed (i) to compare WB-TG parameters in SCD patients in steady state with healthy controls, and (ii) to explore associations between TM-modified WB-TG parameters and laboratory variables and clinical outcomes.

Methods. In this single-center observational study, SCD patients (≥10 years) were enrolled in steady state (no vaso-occlusive crisis, hospitalization, or blood transfusion/exchange within 4 weeks). WB-TG was performed within 4 hours from blood sampling, using TF 1 pM, without added phospholipids, in the absence/presence of TM (20 nM, selected to achieve ~50% inhibition in controls). TG parameters included lag time, time-to-peak, peak thrombin, endogenous thrombin potential (ETP), velocity index, ETP ratio (ETP+TM/ETP), and ETP inhibition (%). Clinical history and standard laboratory data were collected at baseline. Patients were followed for 12 months. Statistical analysis was performed using Mann–Whitney tests for comparisons and exploratory univariate logistic regression for associations with clinical outcomes.

Results. WB-TG was analyzed in 28 SCD patients (median age 19 years) and 21 healthy controls. In steady state, SCD patients showed accelerated thrombin generation kinetics compared to controls: shorter lag time 2.5 [2.4–2.8] vs 3.8 [3.2–4.3] min and time-to-peak 6.3 [5.8–7.1] vs 9.7 [8.3–10.0] min (both p<0.001), higher velocity index 54.4 [44.3–64.8] vs 36.4 [29.3–39.7] nM/min (p<0.001), while ETP and peak thrombin were not significantly different. In presence of TM, SCD patients maintained faster kinetics: shorter lag time+TM 4.2 [3.8–5.4] vs 5.9 [5.2–6.5] min and time-to-peak+TM 7.4 [6.8–7.7] vs 10.2 [9.5–11.5] min (both p<0.001), higher velocity index+TM 49.5 [39.9-60.4] vs 30.6 [24.5-37.6] and higher peak+TM 152.3 [130.9-180.0] vs 117.3 [109.6-132.7] (p=0.034). TM-mediated inhibition was reduced in SCD, indicating TM resistance, as shown by lower ETP inhibition 27.8% [17.6–34.8] vs 46.6% [33.4–52.5] (p=0.018). Lower ETP inhibition was associated with acute clinical events in the previous 12 months: vaso-occlusive crises (p=0.015, OR 0.93, 95% CI 0.87–0.99), acute chest syndromes (p=0.003, OR 0.86, 95% CI 0.76–0.97), and hospitalizations (p=0.015, OR 0.94, 95% CI 0.88–1.00) (Table 1). No significant associations were observed between WB-TG parameters and events over the 12-months follow-up.

Conclusions. In steady state, SCD patients exhibit accelerated thrombin generation kinetics and reduced TM-mediated inhibition, supporting a persistent hypercoagulable phenotype likely related with impaired protein C pathway modulation. The association between TM-modified WB-TG parameters, particularly ETP inhibition, and prior acute clinical events suggests a potential role of this assay in thrombo-inflammatory risk stratification in SCD. Larger prospective studies with longer follow-up are needed to validate these findings and confirm the predictive clinical utility of WB-TG.

Table 1. ETP inhibition according to clinical events in the prior 12 months

Event	Present	Absent	ETP inhibition (%) Event present*	ETP inhibition (%) Event absent*	p value†	OR (95% CI)‡
VOC	18	10	21.3 (IQR 12.9–29.9)	33.6 (IQR 31.7–37.9)	0.015	0.93 (0.87–0.99)
ACS	7	21	11.9 (IQR 2.4–16.6)	30.5 (IQR 24.7–35.2)	0.003	0.86 (0.76–0.97)
Hospitalizations	16	12	21.3 (IQR 10.8–28.6)	33.6 (IQR 29.0–36.1)	0.015	0.94 (0.88–1.00)

* Values are reported as median (interquartile range, IQR).
† p value obtained using Mann–Whitney U test.
‡ Odds ratio from univariable logistic regression (exploratory analysis due to small sample size).
Abbreviations: VOC, vaso-occlusive crisis; ACS, acute chest syndrome; ETP, endogenous thrombin potential.