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Balanced haemostasis with both hypo- and hyper-coagulable features in critically ill patients with acute-on-chronic-liver failure

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ABSTRACT

Background: Cirrhotic patients have complex haemostatic abnormalities. Current evidence suggests stable cirrhotic (SC) patients have a "re-balanced" haemostatic state. However, limited data exists in acute decompensated (AD) or acute on chronic liver failure (ACLF) patients.
Methods: We utilised thrombin generation analysis, fibrinolysis assessment, and evaluation of haemostatic parameters to assess haemostasis in liver disease of progressive severity.
Results: The study cohorts were comprised of: SC, $n = 8$; AD $n = 44$; ACLF, $n = 17$; and Healthy Control (HC), $n = 35$. There was a progressive increase across the cohorts in INR ($p = 0.0001$), Factor VIII ($p = 0.0001$) and VWF levels ($p = 0.0001$) and a correspondingly decrease in anti-thrombin ($p = 0.0001$), ADAMTS-13 ($p = 0.01$) and fibrinogen levels ($p = 0.0001$). In the presence of thrombomodulin, thrombin generation was equivalent or significantly higher in all the cohorts compared to HC ($p = 0.0001$). Compared to AD, ACLF had a lower ETP ($p = 0.002$) and thrombin peak ($p = 0.0001$). There was no significant difference across the cohorts in clot lysis time ($p = 0.07$), although compared to HC, AD had a significantly shorter lysis time ($p = 0.001$).
Conclusions: Our cohorts, despite significant differences in haemostatic parameters, displayed intact thrombin generation but progressive hypo-functional clot stability and potentially but not universal hyper-functional haemostasis.

1. Introduction

Hospitalised critically ill cirrhotic patients have significant morbidity and mortality and despite improvements in critical care, this patient group has a hospital mortality persistently reported up to 50% [1-5]. A ubiquitous finding in this patient group is a disturbed haemostatic system, commonly represented by the prolongation of the prothrombin time (PT), the associated international normalized ratio (INR), and thrombocytopenia. These abnormalities have traditionally thought to be the result of a progressive decrease in hepatic synthesis of pro-haemostatic factors over the stages (stable to decompensated to acute on chronic liver failure) of chronic liver disease, and represent an overall pro-haemorrhagic tendency.

Recent studies have highlighted that the coagulation abnormalities in liver disease are more complex than these traditional concepts suggest [6-11]. Clinical and laboratory studies in patients with liver disease have demonstrated that whilst abnormalities exist in primary haemostasis, coagulation and fibrinolysis, the loss of pro-haemostatic drivers is balanced by the loss of anti-haemostatic processes [12-15]. The current paradigm is of a delicate "balanced haemostasis", and that imbalance can potentially produce both pro-haemorrhagic or pro-thrombotic states. This may occur in the setting of such systemic disturbance as seen in acute decompensated (AD) cirrhosis and acute-on-chronic liver failure (ACLF) [10,16].

Abbreviations: SC, Stable Cirrhosis; AD, Acute Decompensated Chronic Liver Disease; ACLF, Acute on Chronic Liver Failure; HC, Healthy Control; TM, Thrombomodulin; ETP, Endogenous Thrombin Potential; ICU, Intensive Care Unit; INR, International Normalized Ratio; PT, Pro-thrombin time; LITU, Liver Intensive Therapy Unit; SOFA, Sequential Organ Failure Assessment scale; CLiF-SOFA, Sequential Organ Failure Assessment scale - Chronic Liver Failure; MELD, Model of End-Stage Liver Disease; UK-MELD, United Kingdom - Model of End-Stage Liver Disease; CP, Child-Pugh Score; APTT, Activated partial prothrombin time; VWF, Von Willebrand Factor; ADAMTS 13, A disintegrin and metalloprotease with thrombospondin type 1 motif, member 13; PAI, Plasminogen Activator Inhibitor – type 1; IQR, Interquartile range; nM, nanoMolar; SD, Standard Deviation.

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Table 1
Patient demographic, clinical, and laboratory data, median (IQR).

Demographics & clinical characteristics	Healthy controls, $n = 35$	Stable cirrhosis, $n = 8$	Acute decompensated cirrhosis, $n = 44$	Acute on chronic liver failure, $n = 17$	*P* values
Age	37 (31–41)	59 (45–73)	56 (51–63)	57 (43–61)	0.0001
Male n, (%)	16 (46%)	2 (25%)	30 (68%)	11 (65%)	0.170
CLIF-SOFA score	N/A	5 [3-10]	2 [1-4]	12 (6–16)	0.0001
MELD	N/A	10 (3–19)	9 (6–14)	29 (21–36)	0.0001
UK-MELD	N/A	49 (44–55)	53 (50–58)	65 (59–67)	0.0001
Child-Pugh score	N/A	6 (5–10)	8 (7–10)	11 (11 − 12)	0.0001
Laboratory parameters					
Haemoglobin (g/L)	N/A	99 (89–132)	111 (99–127)	95 (85–105)	0.005
WCC ($\times 10^9$/L)	N/A	4.8 (3.6–9.7)	4.8 (2.9–6.3)	10.7 (5.1–18.6)	0.003
Sodium (mmol/L)	N/A	139 (137–142)	137 (132–140)	136 (132–139)	0.167
Urea (μmol/L)	N/A	13.0 (5.2–22.4)	5.2 (3.4–7.2)	13.3 (7.5–17.7)	0.002
Creatinine (μmol/L)	N/A	94 (57–115)	65 (53–93)	131 (92–230)	0.0001
Bilirubin (μmol/L)	N/A	20 (6–57)	38 (24–70)	362 (100–476)	0.001
Gamma-glutamyl transpeptidase (IU/L)	N/A	108 (45–176)	83 (45–191)	58 (32 − 101)	0.192
Alakaline phosphatase (IU/L)	N/A	104 (94–130)	157 (115–195)	136 (112 − 201)	0.036
Aspartate transaminase (IU/L)	N/A	31 (27–43)	51 (35–74)	78 (50–85)	0.002
Albumin (g/L)	N/A	43 (41–46)	32 (30–37)	27 (24–33)	0.0001
Ammonia (μmol/L)	N/A	44 (22–95)	58 (41–79)	76 (35–100)	0.369
Lactate (mmol/L)	N/A	1.2 (1.1–1.2)	1.4 (1.1–2.1)	1.8 (1.5–2.9)	0.01

CLIF-SOFA = Chronic Liver Failure-Sequential Organ Failure Assessment, MELD = model for end-stage liver disease, UK = United Kingdom model for end-stage liver disease, WCC = white cell count,

Table 2
Patient coagulation assays, median (IQR).

	Healthy controls, $n = 35$	Stable cirrhosis, $n = 8$	Acute decompensated cirrhosis, $n = 44$	Acute on chronic liver failure, $n = 17$	*P* values
Primary haemostasis					
Platelets ($\times 10^9$/L)	N/A	108 (81–141)	73 (53–122)	76 (48–110)	0.341
VWF (%)	72 (47–89)	298 (140–502)	363 (302–475)	695 (385–1047)	0.0001
ADAMTS 13 (%)	90 (79–104)	90 (31–109)	70 (46–91)	33 (17–51)	0.01
Fibrinogen (g/L)	2.7 (2.5–3.0)	3.4 (2.2–4.3)	2.3 (1.6–3.1)	1.35 (1.2–1.5)	0.0001
Coagulation assays					
INR	1.0 (0.98–1.07)	1.2 (1.16–1.4)	1.4 (1.2–1.7)	1.9 (1.6–2.3)	0.0001
Prothrombin time (sec)	11 (10.6–11.6)	13 (12.6–15.3)	15 (13–19)	21 (17–25)	0.0001
AT (%)	112 (102–117)	87 (49–106)	48 (30–68)	27 (14–34)	0.0001
FVIII (%)	95 (75–106)	147 (116–141)	145 (127–167)	168 (131–205)	0.0001
Fibrinolysis					
PAI-1 (ng/ml)	0.60 (0.27–1.31)	0.96 (0.60–2.2)	1.65 (0.80–2.90)	8.60 (2.12–11.40)	0.0001
Clot lysis time (min)	64 (59–68)	59 (55–67)	55 (47–61)	67 (49–109)	0.07

VWF = von Willebrand Factor, ADAMTS 13 = a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13, INR – international normalized ratio, AT = Antithrombin, FVIII = factor VIII [8]. PAI = Plasminogen Activator inhibitor type-1,

Concurrently, evidence and opinion indicates that the commonly utilised standard coagulation assays are insufficient to reflect the complexity of this coagulation imbalance and that more sophisticated measures of global haemostasis may be required [7,11,16,17]. Thrombin generation assays have been increasing utilised in research of the coagulation system in patients with liver disease. Thrombin generation is a critical step in the formation of the fibrin clot, and thrombin generation assays are able to quantify the amount of thrombin generated within a sample. The addition of thrombomodulin to the assay, and subsequent activation of the anti-coagulant protein C pathway, enables a more accurate representation of the in vivo haemostatic characteristics. Studies utilising thrombin generation assays in patients with acute liver failure and those with stable cirrhosis have demonstrated that despite abnormal coagulation assays, these patients are able to, in the presence of adequate platelet counts, generate adequate thrombin for clot formation, with a tendency to a pro-thrombotic state [11,18]. However, in acutely decompensated and acute-on-chronic liver failure (ACLF) patients, there exists very limited data as to what haemostatic changes occur, and in which direction the coagulation balance is shifted.

We therefore performed a prospective observational study to investigate the coagulation abnormalities in patients with stable cirrhosis, acute decompensated cirrhosis and ACLF treated at our institution, in the outpatient setting, on the ordinary wards and the Liver Intensive Therapy Unit (LITU). Specifically, we sought to describe the functional status of the haemostatic system by thrombin generation tests and clot lysis assays. In addition, we performed detailed analyses of routine diagnostic tests of haemostasis and assessed selected haemostatic proteins.

2. Methods

2.1. Study design

The study was performed at King's College Hospital, a 950-bed tertiary hospital in London, United Kingdom, between August 2013 and August 2015. The study was approved by NRES Committee London – Westminster, Study Number 12/LO/1417. Informed consent or assent was obtained from participants or their personal consultees.

2.2. Study participants

2.2.1. Inclusion criteria

Patients were recruited sequentially from outpatients, upon admission to the ward or Intensive Care Unit during the study period. Cirrhosis was defined by the presence of 2 or more of: i) histological evidence of cirrhosis on liver biopsy, ii) Laboratory abnormalities consistent with cirrhosis or iii) Radiological findings consistent with cirrhosis and portal hypertension. Patients were stratified into those with stable cirrhosis (SC), acute decompensation (AD) and acute-on-chronic liver failure (ACLF). Acute decompensation of chronic liver disease and ACLF were defined and graded according to number of organ failures in concordance criteria reported in the CANONIC study [19]. Patients were excluded if they were aged <18-years, or >75-years, pregnant, had serological evidence of blood-borne viral infection (specifically hepatitis B, hepatitis C, and the Human Immunodeficiency Virus), or had evidence of disseminated malignancy. Healthy controls (HC) aged >18-years were enrolled for the comparative cohort, and excluded those

Table 3
Thrombin generation parameters (mean, SD).

	Healthy controls, $N = 35$	Stable cirrhosis, $n = 8$	Acute decompensated cirrhosis, $n = 44$	Acute on chronic liver failure, $n = 17$	P value
Thrombin generation parameters (thrombomodulin −)					
ETP (nM ∗ min)	1133	935	1006	774	0.0001
	(145)	(184)	(194)	(306)	
Lag time (min)	1.86	2.68	1.79	2.42	0.0001
	(0.31)	(1.56)	(0.43)	(1.03)	
Peak (nM)	212	187	197	112	0.0001
	(42)	(41)	(44)	(46)	
Velocity index (nM/min)	73	75	89	44	0.0001
	(27)	(15)	(26)	(24)	
Thrombin generation parameters (thrombomodulin +)					
ETP (nM ∗ min)	459	607	791	597	0.0001
	(165)	(123)	(217)	(292)	
Lag time (min)	1.81	2.78	1.92	2.50	0.0001
	(0.22)	(1.54)	(0.42)	(1.14)	
Peak (nM)	111	133	161	93	0.0001
	(40)	(22)	(41)	(43)	
Velocity index (nM/min)	49	59	75	38	0.0001
	(21)	(15)	(25)	(20)	

ETP = Endogenous Thrombin Potential, nM = nanoMolar, min = minutes,

with a personal history of thrombotic, or liver disease, un-treated medical conditions, or currently using anti-coagulants or the oral contraceptive pill.

2.3. Data collection

We collected baseline data on patient demographics, co-morbidities, biochemistry and illness severity scores. Specifically, the illness severity scores of interest were the sequential organ failure assessment scale (SOFA) and its variant for chronic liver failure (CLiF-SOFA), the model of end-stage Liver disease (MELD), and UK-specific version UK-MELD, and Child-Pugh (CP) Scoring systems. Sepsis syndrome was defined according to SEPSIS-3 guidelines [20], and a bleeding episode defined as one requiring therapeutic intervention or the administration of greater than two packed red blood cells.

2.4. Blood Sampling and plasma preparation

Blood samples were drawn into 9NC coagulation sodium citrate 3.2% vacutainer blood collection tubes (Greiner Bio-one). These were obtained from an arterial line or central venous catheter or standard peripheral venous phlebotomy. Blood from healthy controls was obtained by standard peripheral venous phlebotomy. Samples were obtained prior to the administration of blood products, or the prescription of anti-coagulation or anti-platelet agents, on day 1 on admission. Once samples were obtained, they were processed to platelet-poor plasma by double centrifugation at 2000 and 10,000 g, respectively for 10 min. Platelet poor plasma was aliquoted into CryoVials and immediately stored at −80 °C until use.

2.5. Haemostatic assays

Primary haemostasis was assessed by measurement of platelet count, levels of von Willebrand factor (VWF) and its metalloproteinase inhibitor, ADAMTS 13, as previously described [21]. Coagulation was assessed by measurement of traditional markers of coagulation, prothrombin time (PT), international normalized ratio (INR), activated partial thromboplastin time (APTT), and specific markers including antithrombin, fibrinogen, and factor VIII levels using a an automated coagulation analyzer (ACL 300 TOP) with reagents and protocols from the manufacturer (Werfen, Breda, The Netherlands) as described previously [22]. Thrombin generation testing was performed in absence and presence of thrombomodulin using protocols and reagents from Thrombinoscope (Maastricht, The Netherlands) as described previously [23]. Thrombin generation curves (thrombin generation vs time) allowed calculation of endogenous thrombin potential (ETP) (nM ∗ min) as area under the curve, thrombin lag-time (min) as time to commencement of thrombin generation, thrombin velocity index (nM/min) as the rate of thrombin generation, and thrombin peak (nM) as peak value of thrombin generation. Fibrinolysis was assessed by a plasma-based clot lysis time as described previously [23] and measurement of plasminogen activation-inhibitor type 1 (PAI-1).

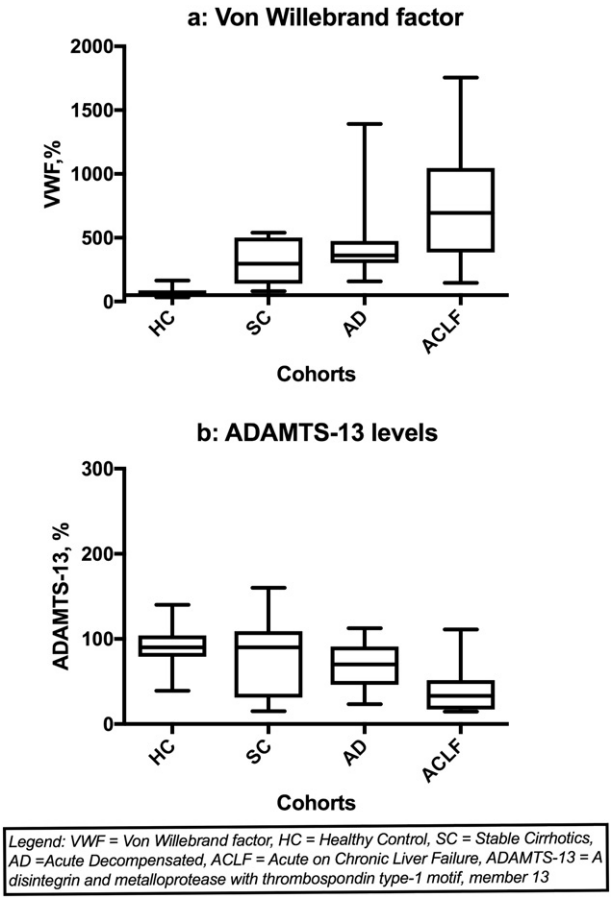

Fig. 1. a–b. Measures of primary haemostasis.

2.6. Statistical analysis

All data analysis was performed by SPSS (IBM SPSS Statistics for Macintosh, Version 22.0. Armonk, NY: IBM Corp). Variables were assessed for normality. Group comparisons were made using chi-squared tests for equal proportions with results presented as numbers (percentages). Normally presented variables were compared with students *t*-test or one-way analysis of variance and presented as means and standard deviations (SD). Post Hoc analysis was compared by Scheffe's method. Non-parametric data were compared by Kruskal-Wallis and Wilcoxon rank- sum tests and presented as medians and inter-quartile ranges (IQR). Non-parametric post-hoc comparisons were performed with Dunn's Test. A two-sided *p*-value of 0.05 was considered statistically significant.

3. Results

3.1. Patient characteristics

Sixty-nine patients met the inclusion criteria; eight patients with stable cirrhosis, 44 with decompensated cirrhosis and 17 with acute on chronic liver failure. In addition, 35 healthy controls were recruited. Two patients were classed as ACLF grade 1, nine patients grade 2, six patients grade 3. Patient demographics, clinical and laboratory data are presented in Table 1. Twenty-five patients underwent sampling as outpatients, and 44 patients whilst admitted to hospital. Of the patients admitted, 23 patients were electively admitted (18 pre-transplant assessment, three for peri-procedural prophylactic blood product administration, two for elective endoscopic procedures) and 21 patients as emergent admissions. Details of the clinical and laboratory features of the different groups are shown in Supplementary Tables 1 and 2.

Table 1. Patient demographic, clinical and laboratory data.

3.2. Haemostatic parameters

The Haemostatic parameters are presented in Tables 2 & 3.

3.2.1. Primary haemostasis

Table 2. Patient haemostatic assays.

Although the overall liver disease cohort was thrombocytopenic with median platelet count of 77×10^9/L (54–112), there existed no significant differences in platelet counts between the patient cohorts. VWF levels increased significantly the across the cohorts ($p = 0.0001$), most markedly in ACLF patients, where levels were significantly higher than the AD cohort, 363% (302–475) vs 695% (385–1047), ($p = 0.048$). Reciprocally, median ADAMTS 13 levels decreased significantly in all cohorts ($p = 0.01$), in particular AD vs ACLF patients, 70 (46–91) vs 33 (17–52), ($p = 0.002$) (Fig. 1).

3.2.2. Secondary haemostasis

Table 2 reveals the significant differences in the general laboratory assays of coagulation within the study cohort. The AT levels decreased significantly across the cohorts, particularly between AD and ACLF, 48% (30–68) vs 27% [17-25], $p = 0.0001$ and FVIII levels significantly increased across all cohorts, but with a non-significant difference seen between ACLF and AD, 145 (127–167) vs 168 (131–205), $p = 0.469$. Fibrinogen levels varied over the cohorts with the lowest seen in

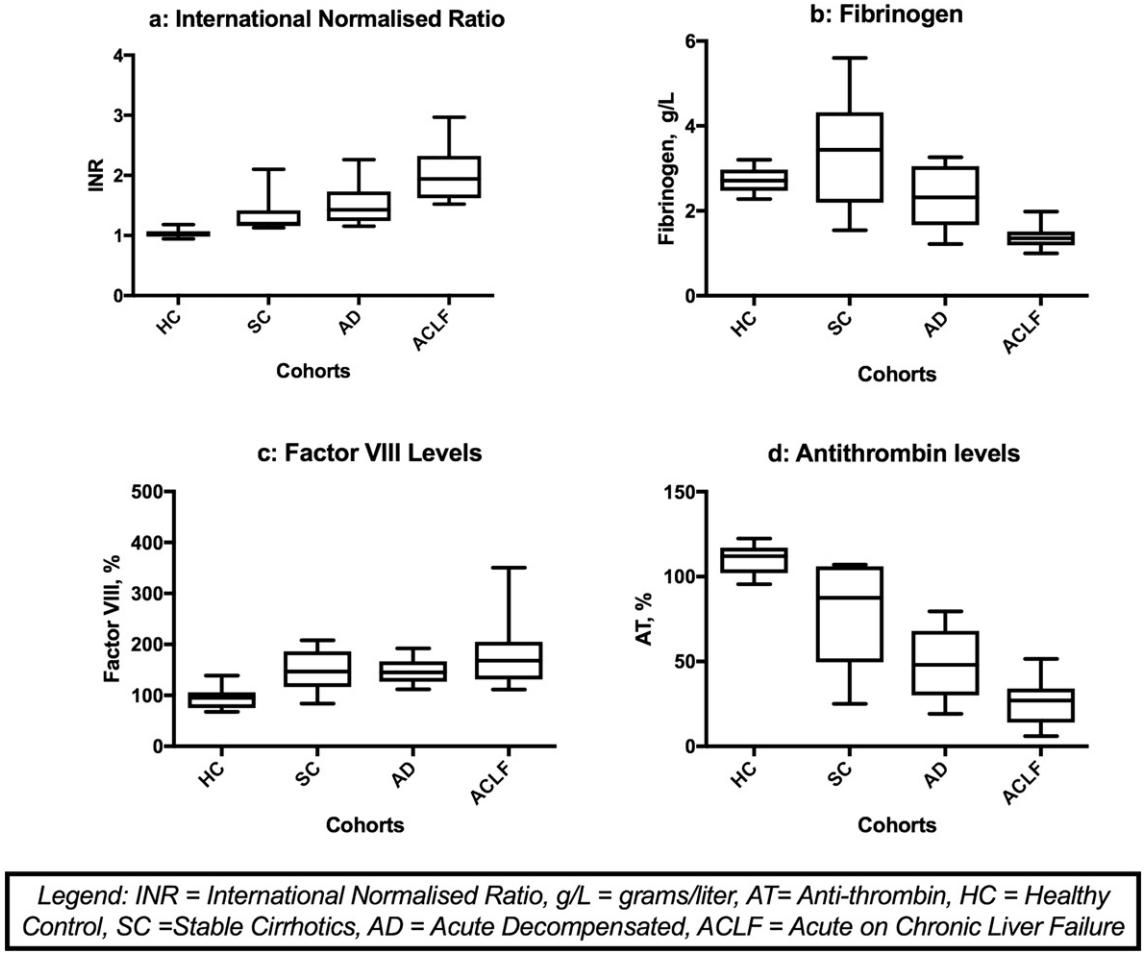

Fig. 2. a–d. Common measures of coagulation.

ACLF, median 1.4 g/L (1.2–1.5). Of note, SC had significantly higher fibrinogen levels compared to HC 3.4 g/L vs 2.7 g, $p = 0.0001$ (Fig. 2).

3.2.3. Estimation of thrombin generation

Table 3. Thrombin generation parameters – in the presence and absence of thrombomodulin.

In the absence of thrombomodulin, the AD and ACLF cohorts, compared to HC, had significantly lower mean ETP 1006 nM ∗ min (SD 194) vs 1133 nM ∗ min (SD 145), $p = 0.03$ and 774 (SD 306) vs 1133 (SD 145), $p = 0.001$ respectively. Thrombin generation peak and velocity were also significantly lower in ACLF compared to HC, 112 nM (SD 47) vs 212 nM (SD 42), $p = 0.01$ and 44 nM/min (SD 24) vs nM/min 73 (SD 28), $p = 0.0001$, respectively. However, peak thrombin generation was not significantly different between the HC and SC cohorts, 212 min (SD 42) vs 187 min (SD 41) vs, $p = 0.138$, or AD cohort, 212 min (SD 42) vs 197 min (SD 44), $p = 0.131$.

In contrast to the findings above, thrombin generation in the presence of thrombomodulin appeared preserved in our cohort and exaggerated in AD but not ACLF. Compared to HC, neither the SC or ACLF cohorts had statistically significant different ETP, ($p = 1$) and ($p = 0.125$), thrombin generation peak ($p = 0.501$) and ($p = 0.444$) or velocities ($p = 0.742$) and ($p = 0.285$), respectively. The AD cohort, compared to HC, had a significantly higher ETP, mean 791 nM min (SD 217) vs 459 nM min (SD 165) $p = 0.0001$, thrombin generation peak, mean 161 nM (SD 41) vs 111 nM (SD 40), $p = 0.0001$, and velocity, mean 75 nM/min (SD 25) vs 49 nM/min (SD 21), $p = 0.0001$.

The ACLF cohort compared to AD cohort displayed significant differences in the thrombin generation parameters. In the absence of TM, the ACLF cohort compared to AD demonstrated lower mean ETP, 774 (306) vs 1006 (194) nM min, $p = 0.0001$, a longer mean lag time, 2.4 (1.0) vs 1.8 (0.4) min, $p = 0.001$, and lower mean peak thrombin generation, 197 (44) vs 112 (47) min, p 0.0001. This was also evident in the presence of TM, with ACLF cohort having a lower ETP of 597 (292) vs 791 (217) nM min, $p = 0.002$, longer lag time 2.5 (1.1) vs 1.9 (0.4) min, $p = 0.003$, and lower mean peak thrombin generation 93 (43) min vs 112 (47), p 0.0001 (Fig. 3).

3.2.4. Fibrinolysis

Clot lysis was variable over the cohorts. There was no difference in median clot lysis time across the cohorts, $p = 0.07$, however the AD cohort was hyper-fibrinolytic compared to HC, 55 min (47–61) vs 64 min (59–68), $p = 0.001$, but clot lysis time was not significantly different between AD and ACLF. Plasminogen Activator Inhibitor–1 levels differed between the cohorts. This difference was most pronounced but not significant between AD and ACLF patients, 1.7 ng/ml (0.8–2.9) vs 8.6 ng/ml (2.1–11.4) $p = 0.113$, although confidence ranges were wide (Fig. 4).

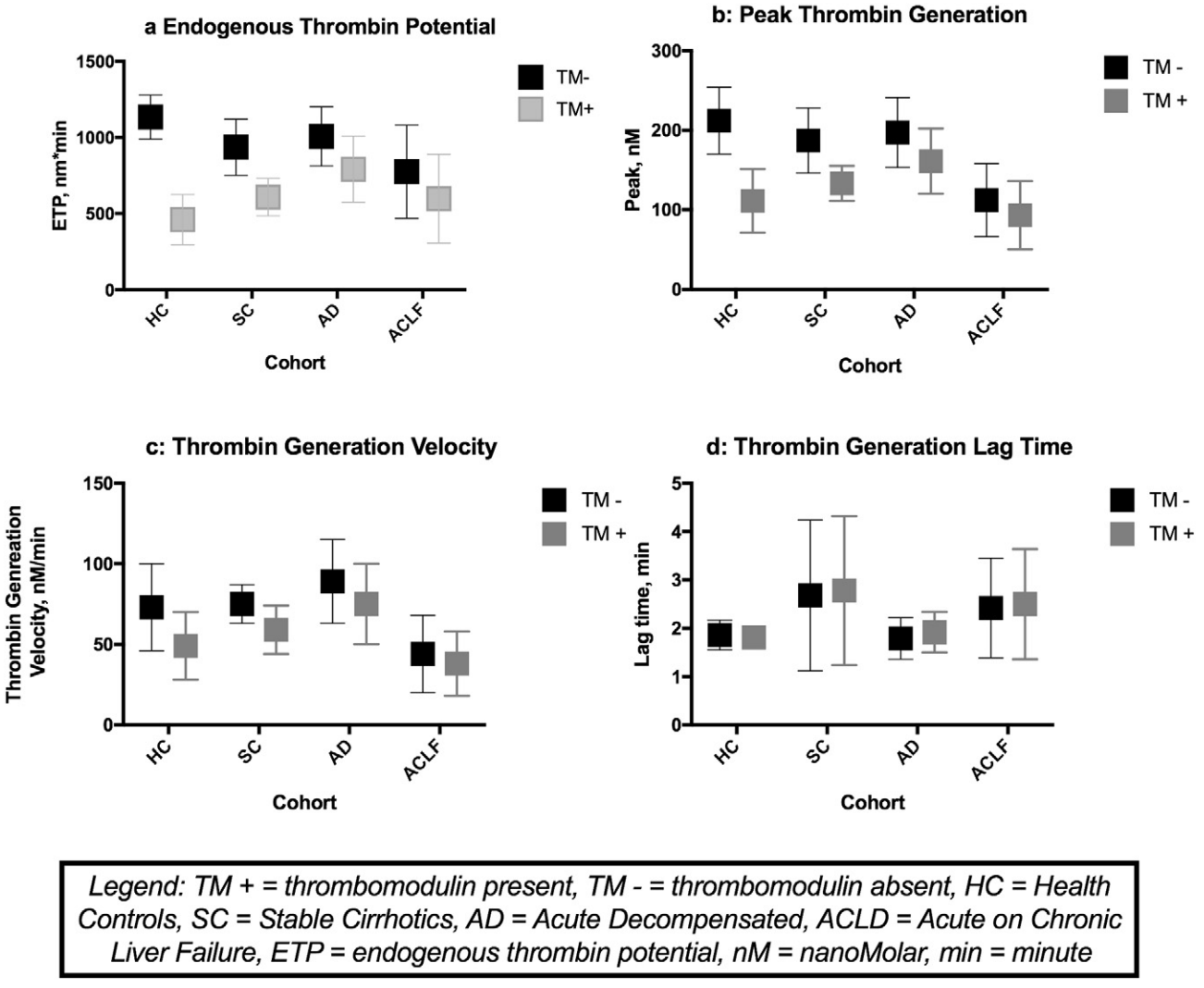

Fig. 3. a–d. Measures of thrombin generation.

4. Discussion

Our study is the first to demonstrate the progression of haemostatic abnormalities that exist in the full spectrum of chronic liver disease of increasing severity. Our patient cohort demonstrated a significant loss of hepatically synthesised pro- and anti-coagulant factors compared to healthy controls. Unsurprisingly, these findings differed according to the stage of liver disease, with more pronounced abnormalities present in acute on chronic liver failure patients as compared to stable and acute decompensated liver disease. Importantly, our study adds to the evidence regarding the complexity of coagulation in patients with liver disease. In presence of thrombomodulin and better reflecting in vivo haemostasis, thrombin generation was equivalent or higher in all cohorts compared to HC, indicating a preserved clotting state. Further, our findings of clear differences in haemostatic parameters between patients with AD and ACLF add further evidence to the concept that ACLF is distinct phenotypic syndrome.

There is increasing recognition of the complexity of the haemostatic changes in liver disease [7,16,17,24]. Studies suggest that primary haemostasis is influenced by the thrombocytopenia of liver disease [12,25] and may be balanced by the increase in the non-hepatic synthesised VWF [12]. The degree of thrombocytopenia appears a crucial determinant in the ability of the haemostatic system to support thrombin formation, with studies suggesting that a platelet count of approximately 60×10^9 is required for thrombin generation [25]. Our patient cohort demonstrated a progressive thrombocytopenia though each stage of liver disease but with a balancing increase in VWF, which has been shown to compensate for the thrombocytopenia of stable cirrhosis [12]. The complex alterations in fibrinolytic potential in our cohort demonstrates that there is not universal hyper-fibrinolysis in cirrhosis [26], but that the fibrinolytic status of cirrhosis is variable and may contribute to either thrombosis or bleeding risk in individual patients. In our cohort, only AD displayed evidence of hyper-fibrinolysis, although the ALCF cohort displayed a very wide range of levels. It is likely that in the haemorrhaging ACLF patient, the combination of hypofibrinogenaemia and potential for hyper-fibrinolysis, the early administration of fibrinogen will be an important therapeutic intervention.

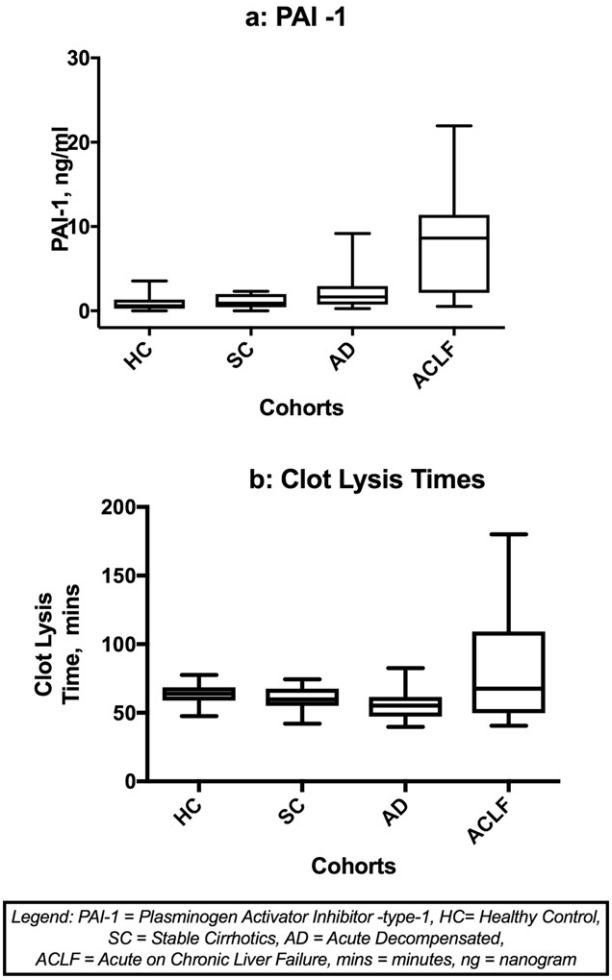

Fig. 4. a–b. Measures of fibrinolysis.

Our findings support previous observations that in many patients with liver disease, thrombin generation is intact despite abnormal coagulation parameters, and many patients may even possess a pro-thrombotic state, as evident by the greater thrombin burst seen in the AD cohort. However, our novel observation is that patients with ACLF appear to have a distinct thrombin generation phenotype that does not represent a simple progression and exaggeration of that seen in SC and AD. As compared to patients with acute decompensation, those with ACLF had similar ETP but longer lag times, lower peak thrombin generation and velocity index. A possible explanation of these findings is that the longer lag time and velocity index are representative of equally decreased pro and anti-coagulant systems resulting in a net normal ETP. As a consequence, this pattern in ACLF represents a balanced but fragile coagulation state. However, although ETP and peak levels are usually well correlated, the peak may be a more sensitive predictor in assessing haemorrhage risk and therefore this may still represent a patient group with increased bleeding risk [27], although bleeding complications were rare in our cohort.

The causes of this distinct ACLF haemostatic phenotype are unclear from our study. It is currently believed that sepsis is a major determinant ACLF in up to 40% of cases [28,29]. The effects on sepsis on coagulation are complex and the interaction between sepsis, systemic inflammation and the complex haemostasis in patients ACLF is still being actively elucidated [30-35]. In our study, a low number of patients had a proven septic determinant and we detected no difference in our haemostasis analysis. Future studies are required to further explore the changes that might occur in this patient cohort.

Our findings continue to challenge the notion that the prophylactic administration of blood products, largely erroneously based on abnormal traditional tests of coagulation, will decrease the risk of bleeding in cirrhotic patients. The increasing accuracy and availability of haemostatic tests that better assess global haemostasis has the potential to positively impact patient management in this regard and recent research suggests they may be beneficial and safe, although uncertainties still exist in regard to the ideal assessment tool [36,37]. Conversely, given the high frequency of venous thrombo-embolic disease in hospitalised cirrhotic patients (VTE) [38], our findings suggests that proactive consideration of thrombo-prophylaxis is required and that clinicians should be wary of relying on "auto-anticoagulation" based on the prothrombin and INR times. By contrast, in those patients who have progressed to ACLF, VTE approaches may need to be different as a possible pro-thrombotic tendency cannot be assumed. Ideally, VTE prophylaxis and haemostatic support should be individualised based upon the results of functional testing.

Our study has a number of strengths. The cohorts were stratified and analysed along clear diagnostic and prognostic groups, prior to the administration of any potentially confounding blood products or anti-haemostatic medications. The samples underwent timely detailed thrombin generation and haemostatic protein analysis. However, our study has the potential limitation of small sample size, particularly in the SC group.

5. Conclusions

Our study demonstrates that haemostasis in liver disease displays a mixed phenotype, with significant differences in primary haemostasis, coagulation and fibrinolysis between stable cirrhosis, acute

decompensated cirrhosis, and acute on chronic liver failure. Additionally, it appears that acute on chronic liver failure appears to have a separate distinct phenotype. Despite these changes, thrombin generation appears preserved in cirrhotic patients, and potentially exaggerated in patients with acute decompensation. Our results highlight the limitations of traditional tests of coagulation and the need for more sophisticated assessments of coagulation and individualisation of blood product replacement in critically ill cirrhotic patients.

Authors statement of claims and financial statement

Debbie Shawcross has served as a speaker, consultant and an advisory board member for Norgine. She has also been the recipient of an investigator initiated study grant from Norgine (RIFSYS Trial).

Vishal Patel has served as a speaker for Norgine.

William Bernal has served as a consultant for Vital Therapies.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.jcrc.2017.07.053.

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