
Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Thrombosis Research 195 (2020) 226–227

Contents lists available at ScienceDirect

Thrombosis Research

journal homepage: www.elsevier.com/locate/thromres

Letter to the Editors-in-Chief

# Heparin failure and COVID-19: Should we explore other anticoagulants? An observational report regarding *in-vitro* recovery of anticoagulant action in COVID-19 patients in intensive care

*Dear Editor,*

COVID-19 has been associated with high rates of thrombosis (both venous and arterial) in critically unwell patients in intensive care units (ICU) despite the utilisation of prophylactic low-molecular weight heparin (LMWH), which we and others have previously described [1–3]. Llitjos et al., 2020, found a cumulative incidence for venous thromboembolism of 69% for COVID-19 ICU patients, with 31% on prophylactic LMWH and 69% on therapeutic LMWH [4]. If 'immunothrombosis' is excluded however the venous thromboembolism rates may potentially be lower, and a 5% prevalence rate was reported in a single-centre ICU COVID-19 study [5]. To put this into context, prophylactic LMWH reduces venous thromboembolism risk by around 50% in hospitalised (non COVID-19) medical patients, so there is a significant 'failure' rate even in non-COVID patients [6]. We have recently found that ICU COVID-19 patients on therapeutic doses of either LMWH or unfractionated (UFH) are heparin resistant, demonstrating this *via* either decreased anti-Xa peak after therapeutic LMWH or requiring doses of UFH in excess of 35,000 units per day [7]. The *in-vitro* anti-Xa recovery of ICU COVID-19 patient samples spiked with LMWH was decreased compared to normal pooled plasma. It has been shown previously in (non COVID-19) ICU patients that after 2500 IU of dalteparin the anti-Xa activity is approximately half of the value of that in healthy volunteers [8]. Other anticoagulants, including direct-oral anticoagulants (DOAC), as primary thromboprophylaxis, could therefore merit further consideration in COVID-19 ICU patients. These may have a theoretical advantage due to their independence from antithrombin.

We investigated this by comparing the *in-vitro* recovery of 12 COVID-19 ICU patients plasma spiked separately with LMWH, rivaroxaban and apixaban. This study had institutional approval from the research & development department. An *in-vitro* spiking study was conducted with 12 randomly selected ICU COVID-19 confirmed patients from one day.

The anti-Xa activity was assessed at baseline using the heparin anti-Xa activity assay and then immediately post-spiking with either the heparin anti-Xa activity assay, rivaroxaban assay (an anti-Xa activity assay) or apixaban assay (an anti-Xa activity assay) as appropriate. The inter-assay coefficient of variation for these 3 assays was 3.44%, 5.79% and 3.19% respectively. *In-vitro* recovery of the anti-Xa was determined by spiking a commercial pool of normal plasma (Cryocheck, Precision Biologic, USA) with 0.9 IU/ml dalteparin (Pfizer, UK), or 80 ng/ml rivaroxaban (Bayer) or 110 ng/ml apixaban (Pfizer, UK). Baseline antithrombin (Stago, France), one-stage Factor VIII, Clauss Fibrinogen and anti-Xa (Werfen, UK) were assessed using the ACL TOP 750 (Werfen, UK).

The *in-vitro* absolute recovery for each sample is shown. This was calculated by subtracting the baseline anti-Xa from the anti-Xa after spiking, and where appropriate converting to DOAC concentration. The normal pooled plasma anti-Xa, after spiking, was defined as 100% recovery of anti-Xa activity given it has no baseline anti-Xa activity. To convert this to a percentage expected recovery for the 12 samples, compared to the normal plasma pool, we used the formula: mean *in-vitro* recovery (% expected) = observed increase in anti-Xa activity (or DOAC concentration) of patient sample from baseline/observed increase in anti-Xa activity of normal pooled plasma from baseline (or DOAC concentration) x100.

Baseline anti-Xa was elevated in many patients; patients were managed with twice daily prophylactic LMWH and some may have had additional unfractionated heparin through a renal replacement therapy circuit.

Statistical analysis was performed using Microsoft Excel 2010 (Microsoft, Redmond, WA, USA) using the paired *t*-test and a *p*-value of 0.01 was considered significant. *In-vitro* absolute recovery is shown in the Table 1. The mean recovery of LMWH was 74%, rivaroxaban 119% and apixaban 100%. A statistically significant difference between LMWH recovery and both rivaroxaban and apixaban, was observed ($p < .01$ in both cases). This demonstrates that Xa-inhibiting DOAC have a more predictable response in COVID-19 patients in this *in-vitro* model, and LMWH appeared to have a somewhat blunted response. We have previously speculated reasons for this could include antithrombin deficiency (not seen in these samples) or absorption of heparin onto plasma proteins [7]. In the future work should be performed to examine this in non COVID-19 patients.

Apart from anti-coagulant effects, heparin has other biological effects (antiviral, anti-inflammatory and endothelial protection) which direct oral anticoagulants do not have and thus may be at a disadvantage compared to heparin [9]. In summary, other (non-heparin) anticoagulants used as primary thromboprophylaxis, which have more predictable (and less blunted) effects than heparin could warrant exploration in well conducted clinical trials. Previously however, when DOAC has been studied as thromboprophylaxis in medical patients, it has not proved successful, with the exception of betrixaban [10–12]. Another limitation is that patients in ICU are generally not able to take tablets (limiting DOAC utility) and may have renal and hepatic impairment. Anti-virals can also cause an increase in plasma DOAC level, which may be of concern due to bleeding risk [13]. Whether any anticoagulant therapy can influence the micro-thrombosis seen with COVID-19, as this may be the end point of extreme inflammation (the so called 'immunothrombosis'), requires further investigation. Further research into thromboprophylaxis failure is required.

https://doi.org/10.1016/j.thromres.2020.08.010
Received 19 May 2020; Received in revised form 24 July 2020; Accepted 5 August 2020
Available online 09 August 2020

**Table 1**
Laboratory analysis of the recovery of dalteparin, rivaroxaban and apixaban levels in 12 patients from ICU with COVID-19. All patients received prophylactic LMWH however due to differing sampling times (and possible heparin resistance) not all patients had detectable baseline anti-Xa activity. Abbreviation: LMWH – low-molecular weight heparin.

| Patient number | Antithrombin activity (U/dL) | Factor VIII (IU/dL) | Clauss fibrinogen (g/l) | Baseline anti-Xa (IU/ml) | *In-vitro* recovery of anti-Xa IU/ml | *In-vitro* recovery of Rivaroxaban ng/ml | *In-vitro* recovery of Apixaban ng/ml |
|---|---|---|---|---|---|---|---|
| Normal pooled plasma | 109.5 | 101.6 | 2.67 | 0 | 0.87 | 80 | 109 |
| 1 | 111.3 | 209.7 | 3.64 | 0.03 | 0.68 | 65 | 106 |
| 2 | 94 | 277.9 | 4.2 | 0.1 | 0.66 | 115 | 127 |
| 3 | 110.1 | 322.4 | 5.62 | 0.27 | 0.69 | 104 | 112 |
| 4 | 137.8 | 294.2 | 4.56 | 0.44 | 0.65 | 68 | 105 |
| 5 | 118.4 | 142.4 | 4.38 | 0.09 | 0.65 | 107 | 112 |
| 6 | 114 | 311.5 | 4.76 | 0.35 | 0.70 | 95 | 99 |
| 7 | 68.4 | 294.2 | 5.77 | 0.02 | 0.66 | 96 | 97 |
| 8 | 86.2 | 219.3 | 5.62 | 0 | 0.57 | 95 | 112 |
| 9 | 99.1 | 251 | 4.38 | 0.54 | 0.59 | 85 | 97 |
| 10 | 106.9 | 396.8 | 6.83 | 0.23 | 0.48 | 102 | 114 |
| 11 | 115.3 | 237.2 | 3.83 | 0 | 0.74 | 96 | 130 |
| 12 | 72.5 | 365.9 | 6.63 | 0 | 0.63 | 110 | 100 |

## CRediT authorship contribution statement

WT, SM & MB – designed the experiment. DW & SC-M undertook laboratory work. All authors wrote the manuscript and approved the final draft.

## Declaration of competing interest

WT – speakers fees from Pfizer, Bayer, Takeda and advisory boards for Daiichi-Sankyo, Sanofi and Ablynx. SM – support to attend educational meetings from Sysmex, Werfen and Stago. MB – speakers fee from Stago, and advisory boards for Novartis, Cosmopharma, Werfen, Agios and a grant from Mitsubishi. DW & SC-M – none to declare.

## References


[1] W. Thomas, J. Varley, A. Johnston, et al., Thrombotic complications of patients admitted to intensive care with COVID-19 at a teaching hospital in the United Kingdom, Thromb. Res. 191 (2020) 76–77.

[2] S. Middeldorp, M. Coppens M, T.F. van Haaps, et al., Incidence of venous thromboembolism in hospitalized patients with COVID-19, J. Thromb. Haemost. (2020), https://doi.org/10.1111/jth.14888 Accepted Author Manuscript.

[3] F.A. Klok, M.J.H.A. Kruip, N.J.M. van der Meer, et al., Incidence of thrombotic complications in critically ill ICU patients with COVID-19 [published online ahead of print, 2020 Apr 10], Thromb. Res. (2020), https://doi.org/10.1016/j.thromres.2020.04.013 S0049-3848 (20)30120-1.

[4] J. Llitjos, M. Leclerc, C. Chochois, et al., High incidence of venous thromboembolic events in anticoagulated severe COVID-19 patients, J. Thromb. Haemost. (2020), https://doi.org/10.1111/jth.14869 Accepted Author Manuscript.

[5] M.J.R. Desborough, A.J. Doyle, A. Griffiths, et al., Image-proven thromboembolism in patients with severe COVID-19 in a tertiary critical care unit in the United Kingdom, Thromb. Res. 193 (2020) 1–4.

[6] L. Wein, S. Wein, S.J. Haas, et al., Pharmacological venous thromboembolism prophylaxis in hospitalized medical patients: a meta-analysis of randomized controlled trials, Arch. Intern. Med. 167 (2007) 1476–1486.

[7] D. White, S. MacDonald, T. Bull, et al., Heparin resistance in COVID-19 patients in the intensive care unit, J. Thromb. Thrombolysis 50 (2020) 287–291.

[8] M.K. Rommers, N. Van der Lely, T.C. Egberts, et al., Anti-Xa activity after subcutaneous administration of dalteparin in ICU patients with and without subcutaneous oedema: a pilot study, Crit. Care 10 (2006) R93.

[9] J. Thachil, The versatile heparin in COVID-19, J. Thromb. Haemost. 18 (2020) 1020–1022.

[10] A.T. Cohen, R.A. Harrington, S.Z. Goldhaber, et al., Extended Thromboprophylaxis with betrixaban in acutely ill medical patients, N. Engl. J. Med. 375 (2016) 534–544.

[11] A.C. Spyropoulos, W. Ageno, G.W. Albers, et al., Rivaroxaban for thromboprophylaxis after hospitalization for medical illness, N. Engl. J. Med. 379 (2018) 1118–1127.

[12] S.Z. Goldhaber, A. Leizorovicz, A.K. Kakkar, et al., Apixaban versus enoxaparin for thromboprophylaxis in medically ill patients, N. Engl. J. Med. 365 (2011) 2167–2177.

[13] S. Testa, P. Prandoni, O. Paoletti, et al., Direct oral anticoagulant plasma levels' striking increase in severe COVID-19 respiratory syndrome patients treated with antiviral agents: the Cremona experience, J. Thromb. Haemost. 18 (2020) 1320–1323.



W. Thomas*, D. White, S. Cox-Morton, S. MacDonald, M. Besser
*Department of Haematology, Cambridge University Hospitals NHS Foundation Trust, Cambridge, United Kingdom*
*E-mail address:* w.thomas1@nhs.net (W. Thomas).

* Corresponding author.