ANTI-COAGULANTS (HEPARIN AND ITS DERIVATIVES)

Fatima Johanna T. Santos-Ocampo, MD and Roxanne C. Hao, MD

Introduction

Common laboratory abnormalities found in patients with COVID-19 not only include lymphopenia, elevation in lactate dehydrogenase, C-reactive protein, and interleukin-6 (IL-6) but also a procoagulant profile[1]. Characteristically, elevated concentrations of D-dimer, fibrin degeneration products and fibrinogen, and modestly low platelet counts are seen[2,3]. This type of profile is consistent with the increasing reports of widespread thromboses and disseminated intravascular coagulopathy in COVID-19 patients[4,5,6,7,8]. Lung involvement has been primarily noted and a strong association between coagulation dysfunction and ARDS was seen and is therefore considered as risk factors for mortality.[9]

Mechanism of Action

Among the anticoagulants that are in standard use and those that are under investigation, heparin is the most widely studied. At present, it is known to have at least four functions based on studies on different clinical conditions.

1. Anti-coagulant

Its anticoagulant properties come indirectly from its binding with antithrombin III (AT) and facilitating the subsequent inhibitory effect of AT on thrombin and activated factor X (factor Xa)[10,11]. It contains a unique pentasaccharide sequence that has an inhibitory action on factor Xa [12,13] recently synthesized for its targeted effect.

Types:

a. Unfractionated (UFH): short acting form, more suitable for patients with renal failure and acute coronary syndromes due to ease of hepatic clearance and better reversibility with protamine sulfate.
b. Low molecular weight Heparin (LMWH): long acting form such as enoxaparin, dalteparin and tinzaparin, with better adverse reaction profile than the UFH, less requirements for monitoring, higher bioavailability, and the potential for outpatient administration[14,15].
c. Fondaparinux: a synthetic analog of the pentasaccharide sequence of heparin necessary for AT binding as a prerequisite for Factor Xa inhibition and does not affect platelet function[16].

2. Anti-inflammatory

Heparin may **indirectly,** decrease inflammation by blocking the production of more fibrin as well as generation of degradation products. These substances can promote development of inflammation by activating neutrophils and monocytes, inducing the secretion of some inflammatory cytokine as seen in

Figure 4.[17,18,19]

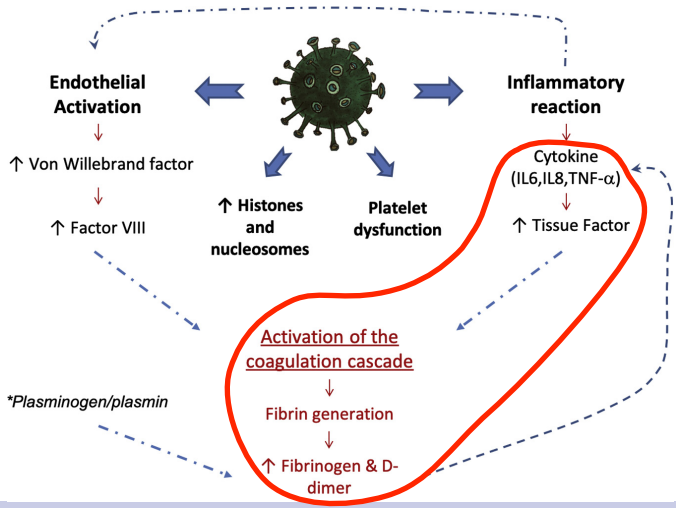

Figure 4. Mechanisms of coagulation impairment in COVID-19 infection. Area encircled shows the relationship between coagulation by-products and augmentation of the inflammatory response. (Adapted from Constanzo, et al)

A possible direct anti-inflammatory action of heparin in COVID-19 is being considered as well. In a systematic review by Mousavi et al in 2015 [20], it was found out that heparin can decrease the level of inflammatory biomarkers. The review mainly involved the following conditions: asthma, inflammatory bowel disease, cardiopulmonary bypass, cataract surgery and acute coronary syndrome.

Heparin's anti-inflammatory effects may be attributed to its ability to bind with inflammatory cytokines[21] , inhibit neutrophil chemotaxis and leucocyte migration[22], sequester acute phase proteins such as P-selectin and L-selectin[23], induce cell apoptosis through tumor necrosis factor a and nuclear factor kβ pathways[21], affect histone methylation[24], affect mitogen-activated protein kinase and nuclear factor kβ signal pathways by inhibiting NF kappa β translocation from cytoplasm to the nucleus [25] and to neutralize complement factor C5a [26].

The neutralizing effect of heparin on C5a may also reduce its prothrombotic effect of upregulating tissue factor and PAI-1 expression by endothelial cells and monocytes.[27,28]

Other mechanisms for heparin's anti-inflammatory and anticoagulant effects have been previously studied in obstetric antiphospholipid antibody syndrome. Since a few case reports on COVID19 patients revealed significant levels of antiphospholipid antibodies[29,30] it would be worth investigating if heparin's therapeutic effects in such patients may be similar mechanistically to what is seen in patients with antiphospholipid antibody syndrome. To prove the theory, more high-quality evidence coming from RCT's are needed.

3. Endothelial protection

In rats, heparin has been shown to antagonize histones which, once released from damaged cells can injure endothelial cells. [31,32]

4. Anti-viral

In vitro studies have shown that heparan sulfate, an ubiquitous glycosaminoglycan on cell surfaces has been seen to interact with the SARS-Cov-2 spike protein and facilitate viral entry [33,34] It cleaves the S1 and S2 subunit of the S protein which exposes the S2 subunit, allowing it to bind with the ACE2 receptor. Heparin can bind to SARS-COV-2 and competitively inhibit [35] its attachment to the cell surface heparan sulfate. This property was seen in unfractionated heparin and was not appreciated in low molecular weight heparin[36].

Clinical Studies

According to a search done at clinicaltrials.gov site, only 3 (2 observational, 1 interventional, no RCT's) studies on heparin and its derivatives have been completed as of this writing but results have not been reported yet. There are at least 9 other studies that are in various stages of development.

Recommended Dose

The International Society of Thrombosis and Haemostasis (ISTH) has also endorsed the guidelines of the Journal of American College of Cardiology which recommends that hospitalized COVID19 patients with respiratory failure, comorbid conditions (cancer, heart failure), bedridden and receiving intensive care should receive pharmacological venous thromboembolism prophylaxis unless with contraindications. No specific doses were given.[37]

In the Philippines, the Philippine Society of Vascular Medicine (PSVM) has suggested the initiation of anticoagulation prophylaxis if any the following are present in COVID 19 patients:[38]

a) ISTH criteria [39,40]: D-dimer > 2 micrograms/ml ± prolonged protime ± platelet <100x $10^{9/}$L
b) Bleeding risk by Padua Prediction score for venous thromboembolism ≥ 4 [41]
c) Sepsis-induced coagulopathy (SIC) >4[42]
d) Critically ill (admission to ICU requiring mechanical ventilation or FiO2 of 60% or higher) [41]

PSVM recommended doses:

		Enoxaparin	Unfractionated Heparin
Patient's Weight	<80 kg 80-120 kg >120 kg	40 mg SC OD 60 mg SC OD 80 mg SC OD	5,000 u SC Q8H or Q12H (for all weight categories)
Creatinine Clearance (CrCl)	≥ 30 ml/min 15-29 ml/min <15 ml/min	As above (according to patient's weight 20 mg SC Not indicated	5,000 u SC Q8H or Q12H 5,000 u SC Q8H or Q12H 5,000 u SC Q12H

Contraindications for heparin prophylaxis include:[39]
(1) Platelet <25 x $10^{9/}$L
(2) Active bleeding

The PSVM also advises that anticoagulation should be discontinued when:
(1) Platelet count ≤ 20 x 10^9/L without bleeding[39]
(2) Platelet count ≤ 50 x 10^9/L with PT ratio ≥ 1.5[39]
(3) General ISTH bleeding criteria:
   a. Fatal bleeding, and/or
   b. Bleeding in a critical area or organ (i.e., intracranial, intraspinal, intraocular, retroperitoneal, intra-articular or pericardial, or intramuscular with compartment syndrome and/or
   c. Bleeding causing a fall in hemoglobin level of 2g/dl (1.24 mmol/L) or more or leading to transfusion of two or more units of whole blood or red cells

Adverse Effects

There is a 10-15% risk of significant bleeding in heparin use. [43,44] Risk factors for bleeding in the general population is older age, worse illness severity, longer hospital stay, decreased white blood cell and platelet counts which is commonly seen in COVID 19 patients. Another rare complication is heparin induced thrombocytopenia due to the development of antibodies to protein platelet factor 4.[45] However, this is not seen in the use of fondaparinux.

Conclusion

Although heparin has many immunomodulatory effects, its exact mechanism in improving outcomes for COVID-19 patients has yet to be elucidated. However, its potential in providing prophylaxis of thromboembolism as a consequence of COVID-19 should be considered.

REFERENCES:

1. Libby P, Simon DI. Inflammation and thrombosis: the clot thickens. Circulation 2001;103:1718-20.
2. Marik, PE, Andrews L, Maini B. The incidence of deep venous thrombosis in ICU patients. Chest 1997;**111**, 661–664
3. Song J, Wang, G, Zhang W. et al. Chinese expert consensus on diagnosis and treatment of coagulation dysfunction in COVID-19. Military Med Res 2020; **7,** 19. Available from https://doi.org/10.1186/s40779-020-00247-7
4. Merrill, JT, Erkan, D, Winakur, J. *et al.* Emerging evidence of a COVID-19 thrombotic syndrome has treatment implications. Nat Rev Rheumatol 2020. Available from: https://doi.org/10.1038/s41584-020-0474-5
5. Tang N, Li D, Wang X, Sun Z. Abnormal coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia. J Thromb Haemost. 2020;18:844–847. Available from: https://doi.org/10.1111/jth.14768
6. KlokFA,KruipMJHA,vanderMeerNJM,etal.IncidenceofthromboticcomplicationsincriticallyillICU patients with COVID-19. Thromb Res. 2020. doi:10.1016/j.thromres.2020.04.013
7. Llitjos JF, Leclerc M, Chochois C, etal. High incidence of venous thromboembolic events in anticoagulated severe COVID-19 patients. J Thromb Haemost. Published online April 22, 2020. Available from: doi:10.1111/jth.14869
8. Nahum J, Morichau-Beauchant T, Daviaud F, et al. Venous Thrombosis Among Critically Ill Patients With Coronavirus Disease 2019 (COVID-19). JAMA Netw Open. 2020;3(5):e2010478. Available from: doi:10.1001/jamanetworkopen.2020.10478
9. Wu C, Chen X, Cai Y, et al. Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China. JAMA Intern Med. 2020. Available from: https:// doi.org/10.1001/jamainternmed.2020.0994.
10. Brinkhous K, Smith H, Warner E, Seegers W. The inhibition of blood clotting: An unidentified substance which acts in conjunction with heparin to prevent the conversion of prothrombin into thrombin. Am. J. Physiol 1939;125:683–687.
11. Lindahl U, Bäckström G, Höök M et al. Structure of the antithrombin-binding site in heparin. Proc. Natl. Acad. Sci. USA. 1979;76:3198–3202. Available from: doi: 10.1073/pnas.76.7.3198.
12. Lane DA, Denton J, Flynn AM et al. Anticoagulant activities of heparin oligosaccharides and their neutralization by platelet Ffactor 4. Biochem. J. 1984;218:725–732. Available from: doi: 10.1042/bj2180725.
13. Oosta GM, Gardner WT, Beeler DL et al. Multiple functional domains of the heparin molecule. Proc. Natl. Acad. Sci. USA. 1981;78:829–833. Avaialble from: doi: 10.1073/pnas.78.2.829.
14. Gray E, Mulloy B, Barrowcliffe TW. Heparin and low-molecular-height Heparin. Thromb. Haemost. 2008;99:807–818. Available from: doi: 10.1160/TH08-01-0032.
15. Shaughnessy SG, Young E, Deschamps P, Hirsh J. The effects of low molecular weight and standard heparin on calcium loss from fetal rat calvaria. Blood. 1995;86:1368–1373.
16. Oduah EI, Linhardt RJ, Sharfstein ST. Heparin: Past, present, and future. Pharmaceuticals (Basel). 2016;9(3):38. Availlble from: doi:10.3390/ph9030038
17. Li J, Hara H, Wang Y, Esmon C, Cooper DK, Iwase H. Evidence for the important role of in"ammation in xeno- transplantation. J Infamm 2019;16:10.
18. Robson SC, Shephard EG, Kirsch RE. Fibrin degradation product D-dimer induces the synthesis and release of biologically active IL-1 beta, IL-6 and plasminogen activator inhibitors from monocytes in vitro. Br J Haematol 1994;86: 322-6.
19. Shorr AF, Thomas SJ, Alkins SA, Fitzpatrick TM, Ling GS. D-dimer correlates with proinfammatory cytokine levels and outcomes in critically ill patients. Chest 2002;121:1262-8.
20. Mousavi S, Moradi M, Khorshidahmad T, Motamedi M. Anti-inflammatory effects of heparin and its derivatives: A systematic review. Adv Pharmacol Sci. 2015;2015:507151. Available from: doi:10.1155/2015/507151

21. Young E. The Anti-Inflammatory Effects of Heparin and Related Compounds. Thromb. Res. 2008;122:743–752.Available from: doi: 10.1016/j.thromres.2006.10.026.
22. Li JP, Vlodavsky I. Heparin, heparan sulfate and heparanase in inflammatory reactions. Thromb Haemost. 2009;102(5):823-828
23. Nelson RM., Cecconi O, Roberts WG, et al. Heparin oligosaccharides bind L- and P-Selectin and inhibit acute inflammation. Blood. 1993;82:3253–3258.
24. Ma J, Bai J. Protective effects of heparin on endothelial cells in sepsis. Int J Clin Exp Med 2015;8:5547-52.
25. Thourani VH, Brar SS, Kennedy TP, et al. Nonanticoagulant heparin inhibits NF-κB activation and attenuates myocardial reperfusion injury. Am. J. Physiol. Heart Circ. Physiol. 2000;278:H2084–H2093.
26. Esmon CT. Targeting factor Xa and thrombin: impact on coagulation and beyond. Thromb Haemost. 2014;111(4):625-633
27. Ritis K, et al. A novel C5a receptor-tissue factor cross-talk in neutrophils links innate immunity to coagulation pathways. JImmunol. 2006;177(7):4794–4802.
28. Foley JH, Conway EM. Cross talk pathways between coagulation and inflammation. Circ Res. 2016;118(9):1392–1408.
29. Yan Zhang, Meng Xiao, Shulan Zhang et al. Coagulopathy and antiphospholipid antibodies in patients with Covid-19 Case report. N Engl J Med 2020; 382:e38
30. (31) Uthman IW, Gharavi AE. Viral infections and antiphospholipid antibodies. Semin Arthritis Rheum 2002;31(4):256-263
31. Iba T, Hashiguchi N, Nagaoka I, et al. Heparins attenuated histone-mediated cytotoxicity in vitro and improved the survival in a rat model of histone- induced organ dysfunction. Intensive Care Med Exp 2015;3:36.
32. Zhu C, Liang Y, Li X et al. Unfractionated heparin attenuates histone-mediated cytotoxicity in vitro and prevents intestinal microcirculatory dysfunction in histone- infused rats. J Trauma Acute Care Surg 2019;87:614-22.
33. Milewska A, Nowak P, Owczarek K, et al. Entry of human coronavirus NL63 into the cell. J Virol 2018. 92: e01933-17. Avialble from: doi:10.1128/JVI.01933-17.
34. Lang J, Yang N, Deng J, et al. Inhibition of SARS pseudovirus cell entry by lactoferrin binding to heparan sulfate proteoglycans. PLoS One 6 2011: e23710. Available from: doi:10.1371/journal.pone. 0023710.
35. Hippensteel JA, LaRiviere WB, Colbert JF et al.. Heparin as a therapy for COVID-19: current evidence and future possibilities. Am J Physiol Lung Cell Mol Physiol. 2020;319(2):L211-L217. Avaialble from: doi:10.1152/ajplung.00199.2020
36. Kim SY, Jin W, Sood A, et al. Glycosaminoglycan binding motif at S1/S2 proteolytic cleavage site on spike glycoprotein may facilitate novel coronavirus (SARS-CoV-2) host cell entry. (Preprint). *bioRxiv* 2020. Available from: doi:10.1101/2020.04.14.041459.
37. Bikdeli, et al., COVID-19 and thrombotic or thromboembolic disease: Implications for prevention, antithrombotic therapy, and follow-Up. J Am Coll Cardiol. 2020 Jun, 75 (23) 2950-2973.
38. Philippine Society of Vascular Medicine. PSVM Interim Guidelines on Vascular Procedures and Treatment Interventions During COVID-19 Pandemic. https://www.pcp.org.ph. August 2020
39. Thachill J, Tang N, Gando S et al. International Society of Thrombosis and Haemostasis (ISTH) interim guidance on recognition and management of coagulopathy in COVID-19. J Thromb Haemost 2020. 18: 1023-1026. Available from doi:10.1111/jth.14810
40. Yao Y, Cao J, Wang Q et al. D-dimer as a biomarker for disease severity and mortality in COVID-19 patients: a case control study. J intensive care 2020. **8,** 49 Available from: https://doi.org/10.1186/s40560-020-00466-z
41. Jin-fu Xu, Lan Wang, Lan Zhao et al. Risk assessment of venous thromboembolism and bleeding in COVID-19 patients. PREPRINT (Version 1) (24 March 2020) Available from Research Square https://doi.org/10.21203/rs.3.rs-18340/v1
42. Tang N, Bai H, Chen X et al. Anticoagulant treatment is associated with decreased mortality in severe coronavirus disease 2019 patients with coagulopathy. J Thromb Haemost. 2020;18(5):1094-1099. Available from http://doi:10.1111/jth.14817

43. Cossette B, Pelletier MÈ, Carrier N et al. Evaluation of bleeding risk in patients exposed to therapeutic unfractionated or low-molecular-weight heparin: a cohort study in the context of quality improvement initiative. Ann Pharmacother 2010. 44: 994–1002. Avaialble from: doi:10.1345/aph.1M615.
44. Nieuwenhuis HK, Albada J, Banga JD, Sixma JJ. Identification of risk factors for bleeding during treatment of acute venous thromboembolism with heparin or low molecular weight heparin. Blood 1991. 78: 2337–2343. Avaialble from: doi:10.1182/blood.V78.9.2337.2337.
45. Arepally GM. Heparin-induced thrombocytopenia. Blood 2017. 129: 2864– 2872. Available from: doi:10.1182/blood-2016-11-709873.