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Heparins and 2019-nCoV infection: a narrative review

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Abstract. – OBJECTIVE: Patients with 2019-nCoV infection have a high risk to develop venous thrombotic events. Several guidelines recommend the use of either unfractionated heparin or low molecular weight heparins in preventing thrombotic events in these patients. However, results from clinical studies, so far published, reached controversial conclusions on heparin efficacy in this kind of patients since the incidence of venous thromboembolism remains high despite prophylaxis. This narrative review aims to provide an overview of the antiviral and anti-inflammatory properties of heparins and their efficacy and safety in SARS-CoV-2 medical ward-patients. Moreover, anatomical findings and ongoing trials are also reported. Finally, this narrative review tries to explain why heparins fail to prevent venous thrombosis.

MATERIALS AND METHODS: We searched for the most relevant published studies on heparins and 2019-nCoV infected patients using the MEDLINE electronic database in the period between January and December 2020. Articles were preliminarily defined as eligible if they: a) were in English language, b) enrolled 250 or more medical ward-patients and 100 or more ICU-patients, c) reported results on patients treated with heparins in a percentage of at least 70% and d) performed an objectively confirmed diagnosis of VTE.

RESULTS: Data from medium to large scientific studies show that the incidence of venous thrombotic events in medical ward-patients with SARS-CoV-2 vary between 0% and 8.3%, while this rate is higher, from 6.2% to 49%, in Intensive Care Unit-patients. However, heparins reduce the mortality rate in these patients of about 50%. Histological findings show that thrombosis could affect capillaries, main and small-mid-sized vessels, and it is associated with diffuse alveolar damage.

CONCLUSIONS: Heparins have anti-inflammatory and anti-viral properties, which may be of help in reducing mortality in SARS-CoV-2 patients. Failure of heparins at prophylactic dosages in preventing VTE, especially in ICU-patients, could be due to the severity of the disease. Data on the use of heparins in an early phase of the 2019-nCoV infection are still lacking.

Key Words:
SARS-CoV-2, Venous thromboembolism, Heparins.

Introduction

A novel coronavirus (2019-nCoV), responsible for a severe acute respiratory syndrome (SARS-CoV-2), was identified in December 2019 in patients with pneumonia infection epidemiologically linked to an animal market in Wuhan, China[1]. In less than three months from the discovery, the infection spreaded to at least 114 countries, causing more than 4000 deaths. On March 11, the World Health Organization (WHO) announced the pandemic from 2019-nCoV. Until now, more than 112 million people are infected worldwide, and more than 2.5 million died[2].

Even if the respiratory system is principally involved, it soon became clear that intensive care unit- (ICU) and non-ICU patients with SARS-CoV-2 had a high risk of developing venous and arterial thrombosis. Initial studies reported a very high venous thromboembolism (VTE) percentage of up to 50%[3]. Since then, several guidelines and recommendations[4-7] were published with the aim to provide advice on prophylaxis and treatment of VTE. Low molecular

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weight heparin (LMWH) and unfractionated heparin (UFH) are the drugs more frequently used in SARS-CoV-2 patients but the results from clinical studies so far published reached controversial conclusions on the safety and efficacy of heparins in this kind of patients. This narrative review focuses on the role of heparins in SARS-CoV-2 patients.

Materials and Methods

We searched for the most relevant published studies on heparins and 2019-nCoV infected patients using the MEDLINE (January-December 2020) electronic database. The following search terms (text words and MeSH) were used: "COVID-19", OR "SARS-CoV-2", OR "2019-nCoV infection" AND "deep vein thrombosis", OR "pulmonary embolism" AND "low molecular weight heparin" OR "unfractionated heparin" OR "fondaparinux". Articles were preliminarily defined as eligible if they: a) were in English language, b) enrolled 250 or more medical ward-patients and 100 or more ICU-patients, c) reported results on patients treated with heparins in a percentage of at least 70%, and d) performed an objectively confirmed diagnosis of VTE. Articles extracted from the electronic database were screened by reading both titles and abstracts, and then full texts were reviewed. Attention was paid to exclude duplicate articles.

Results

Heparins' Overview

UFH was discovered in 1916 by Jean McLean[8]. It is a high sulfated mucopolysaccharide, with a mean molecular weight of about 15.000 Dalton, constituted by a pentasaccharide and approximately 45 saccharide units. The pentasaccharide is the main responsible for the anticoagulant effect that allows UFH to inhibit several coagulation factors as XIIa, XIa, Xa, IXa, and thrombin[9]. However, UFH needs to bind to antithrombin (AT) as a cofactor for its anticoagulant activity. This binding induces a conformational change of AT which turns from a slow to a rapid inhibitor of the coagulation cascade[10,11]. Factor Xa and thrombin are more sensitive to UFH-AT complex inhibition than the other coagulation factors but thrombin is the most responsive. UFH should be administered subcutaneously or by continuous intravenous infusion since it is not absorbed by mouth. In the bloodstream, the binding to several plasma proteins, endothelial cells, and macrophages reduces its anticoagulant effect which, therefore, should be monitored by the activated Partial Thromboplastin Time (aPTT)[12,13]. The aPTT ratio should be maintained between 1.5 and 2.5 times the basal value to reduce both the risk of venous thromboembolic recurrences and bleeding[14]. UFH has a dose-dependent short half-life, and it is cleared by two different mechanisms: the first is a rapid saturable phase, mainly due to its endothelial and macrophages binding, that cleared a large proportion of the molecule by means of its internalization and depolymerization. The second one is a slow non-saturable mechanism of renal clearance[15,16].

LMWH, discovered in 1976 by Johnson et al[17], is the result of chemical or enzymatic depolymerization of UFH. Several LMWHs are available with a molecular weight ranging from 2000 to 9000 Dalton. Like UFH, LMWHs are indirect inhibitors of the coagulation cascade since they need to bind to AT for their anticoagulant activity[18]. The small structure of LMWHs allows the inhibition of factor Xa and thrombin in a proportion of either 2:1 or 4:1, depending on their chain length. However, this short structure confers to LMWHs a more favorable profile since these drugs show a reduced binding to proteins, endothelial cells, and macrophages, explaining their longer half-life and a more predictable dose-response relationship[9]. In fact, LMWHs do not need laboratory monitoring by aPTT. After subcutaneous administration, the half-life of LMWHs is of up 3-6 hours, and they are cleared by the kidneys.

In 1983, Choay et al[19] isolated the heparin pentasaccharide showing that it was able to form a complex with AT so enhancing the inhibition of factor Xa. This synthetic analogue of the heparin pentasaccharide, Fondaparinux, is characterized not only by its increased AT bond but also by a longer half-life that is of about 17 hours. The advantage of Fondaparinux is the lack of binding to proteins, endothelial cells, and macrophages, so its bioavailability after subcutaneous administration is good, and it should be administered once a day in a fixed dose without laboratory monitoring[20]. Like LMWHs, Fondaparinux is contraindicated in patients with severe renal failure since it is excreted unchanged by the kidneys.

Heparins' Anti-Inflammatory and Anti-Viral Properties

Several studies show that heparins have anti-inflammatory and anti-viral properties.

The anti-inflammatory properties of both UFH and LMWHs were studied in clinical pathological conditions such as asthma, cardiopulmonary bypass, and superficial vein thrombosis. In patients with exercise- or allergen-induced asthma, inhaled heparins are able to reduce histamine and leukotrienes compared to placebo, mitigate the response to adenosine 5-monophosphate, and decrease eosinophils and lymphocytes count in the bronchoalveolar lavage showing an important anti-inflammatory effect in these patients[21-23]. Cardiopulmonary bypass circuit can induce a systemic inflammatory syndrome due to blood contact with this artificial surface. UFH not only prevents circuit's clot formation but is able to decrease the plasma level of several cytokines such as interleukin (IL)-6, IL-8, tumor necrosis factor (TNF)-α, elastase, and complement[24-26].

Patients with superficial vein thrombosis always manifest pain and erythema of their arm or leg. It is daily clinical practice experience of doctors managing this kind of thrombosis that patients get to the better a couple of days after starting heparins. Moreover, several studies[27,28] showed that LMWHs are similar to non-steroidal anti-inflammatory drugs in relieving these symptoms.

In SARS-CoV-2 patients, high levels of chemokines and cytokines are produced[29]. Recently, it has been demonstrated that the severity of 2019-nCoV infection correlates with the serum level of cytokines such as IL-6, IL-10, and TNF-α[30]. Chemokines and cytokines regulate different biological processes, including extravasation of leukocytes from the bloodstream to the adjacent tissues. UFH and LMWHs can be of help in 2019-nCoV infected patients since these anticoagulants are able to inhibit the chemotaxis and the adhesion of the neutrophils to the endothelial cells and to reduce the secretion of TNF-α, TNF-γ, IL-6, and IL-8[31,32]. Moreover, 2019-nCoV induces a cytopathic effect with cellular death, thus facilitating the release of histones that contribute to inflammation, necrosis, apoptosis, and neutrophils extracellular traps (NETs) formation. *In vitro*, heparins are able to neutralize the histones' cytotoxic effects, thus preserving organs from severe damage[33].

The antimicrobial capacities of heparins are suggested by several aspects. First, mast cells, principally located along capillaries and venules, store heparin in granules and release it to the bloodstream. Second, mast cells are widely present in organs such as lung and gut more exposed to the action of external pathogens. Third, a wide variety of vertebrates and invertebrates produce heparins even though they do not come with a hemostatic systemic. Taken together, the evidence suggests that heparins could have further effects other than the anticoagulant properties[34,35].

In humans, anti-viral action of heparins has been studied in several *in vitro* experiments.

In physiological conditions, endothelial cells are linked on their luminal plasma membrane to a network of macromolecules named glycocalyx. The endothelial glycocalyx is mainly formed by heparan sulphate (HS), which along with other glycoproteins and proteoglycans, constitute a luminal mesh allowing endothelial cells to bind soluble proteins[36]. Many viruses such as human immunodeficiency virus, dengue virus and rabies virus utilize HS to enter their target cells[37-39]. Human coronaviruses, including 2019-nCoV, bind to HS for increasing the virus density on cells surface, thus facilitating their interaction with angiotensin-converting enzyme 2 (ACE2) receptors[40]. Recently, it has been demonstrated that the spike glycoprotein's receptor-binding domain firstly interacts with HS favoring a conformational change of the protein in an open form, thus facilitating the binding to the ACE2 receptor. UFH may compete with 2019-nCoV for the binding to HS, thus inhibiting the virus attachment to the cell surface and the viral entry. Authors also demonstrated that, in vitro, 0.3-0.7 U/ml of UFH are able to reduce the percentage of SARS-CoV-2 infected cells[41].

To our knowledge, there are no studies in humans about Fondaparinux and either its anti-viral or anti-inflammatory properties. The only study[42] in the scientific literature reports about the ability of Fondaparinux to reduce chemokines, cytokines, monocyte-chemoattractant protein-1 and granulocyte-macrophage colony-stimulating factor in baboons with sepsis-induced by *Escherichia coli*.

Heparins and Venous Thromboembolism in SARS-CoV-2

Undoubtedly, thrombotic events complicate the management of SARS-CoV-2 patients since pulmonary embolism or pulmonary thrombosis (PE-PT) worsens respiratory failure, and anticoagulant therapy becomes necessary[43,44]. Theo-

retically, several drugs could be administered to these patients but each of them has several limitations in this context. In the medical ward, both vitamin K antagonists (VKAs) and direct oral anticoagulants (DOACs) have the advantage to be administered orally. However, monitoring the INR for VKAs and managing the pharmacological interferences among these two anticoagulants and the anti-viral drugs could be very difficult[45]. On the other hand, in ICU-patients a different route of administration other than oral could be preferable due to the critical clinical conditions of these patients. UFH should be monitored by means of the aPTT but, in patients who require full dosages, it could be complicated to reach and maintain therapeutic plasma concentrations. Sudden changes of both the patient's clinical conditions and plasma reactive proteins' levels that can bind to UFH, seizing it from the bloodstream, could expose patients to either a high thrombotic or hemorrhagic risk. However, UFH is an efficacy and safety anticoagulant in SARS-CoV-2 patients with severe renal failure[46]. LMWHs appear the easiest therapeutic approach since it does not require a laboratory monitoring and can be administered subcutaneously after considering the patient's renal function and the body weight. LMWHs, especially enoxaparin, are the most frequent anticoagulant used in SARS-CoV-2 patients. However, LMWHs can provoke heparin-induced thrombocytopenia (HIT), which may lead to venous and/or arterial thrombosis[47]. HIT has also been described in SARS-CoV-2 patients, as some studies showed[48-49].

Finally, Fondaparinux, a synthetic heparin used in the prevention and treatment of both venous thromboembolism (VTE) and superficial vein thrombosis, has several potential advantages. Its pharmacokinetic and pharmacodynamic properties allow a single daily administration of both prophylactic and therapeutic doses, it is easy to perform in ICU- and non-ICU-patients and guarantee a longer anticoagulant effect. Moreover, Fondaparinux is one of the drugs suggested in case of HIT[50], but, at least up to now, it is not widely used in SARS-CoV-2 patients.

VTE has been largely described[51-54] in patients with SARS-CoV-2 as a result of several risk factors such as the viral cytopathic effect and the patient's comorbidities. In general, the rate of VTE in SARS-CoV-2 patients varies widely in the scientific literature, going from 0% in a study on 388 medical ward Italian patients to up 85% in 48 Chinese critically ill patients[55-56]. These large percentages of VTE could be due to the enrollment of patients with varying degrees of severity of the disease, difficulties in performing appropriate diagnostic procedures, the use or non-use of anticoagulants, and the small sample size considered in some studies.

However, several studies do not report type and dosage of the drugs used, how the evaluation of the thromboembolic events was performed (screening or clinical suspicion), and results are often from medical ward- and ICU-patients together, not allowing to draw definitive conclusions. It is also noteworthy that SARS-CoV-2 patients are mostly elderly, and the incidence rate of asymptomatic deep vein thrombosis (DVT) increases with increasing age[57], so if these patients are not systematically screened, the actual incidence rate of VET could be underestimated regardless of its severity.

When we examined the studies (Table I), which considered a medium-large number of patients (from 256 to 2505), the percentages of thrombotic events have become narrower[55,58-64]. In all these studies, patients were admitted to a medical

Table I. Studies on VTE and 2019-nCoV infected medical ward-patients treated with heparins.

Studies	Country	Setting	Patients n	Design	VTE assessment	Heparins treatment	VTE %
Al-Samkari[58]	USA	MW	256	R	CD	89% SD, 9% ID	4.8%
Bilaloglu[59]	USA	MW	2505	R	CD	100% SD	6.2%
Cattaneo[55]	Italy	MW	388	R	CD	100% SD	0%
Fauvel[60]	France	MW	1240	R	CD	63% SD, 8.4% ID	8.3%
Lodigiani[61]	Italy	MW	388	R	CD	45% SD, 22% ID, 20% FD	6.6%
Pesavento[62]	Italy	MW	324	R	CD	74% SD, 26% ID	2.7%
Trimaille[63]	France	MW	289	R	CD	59% SD, 11% ID, 20 FD	17%
Whyte[64]	UK	MW	1255	R	CD	100% SD	5.4%

Legend: MW = medical ward, R = retrospective, CD = clinical diagnosis, SD = standard dose, ID = intermediate dose, FD = full dose.

Table II. Studies on VTE and 2019-nCoV infected ICU-patients treated with heparins.

Studies	Country	Setting	Patients n	Design	VTE assessment	Heparins treatment	VTE %
Al-Samkari[58]	USA	ICU	144	R	CD	89% SD, 9% ID	7.6%
Bilaloglu[59]	USA	ICU	829	R	CD	100% SD	6.2%
Helms[65]	France	ICU	150	P	CD	78% SD, 22% FD	16.7%
Klok[66]	Netherlands	ICU	184	P	CD	50% SD, 50% ID	49%
Moll[67]	USA	ICU	102	R	CD	100% SD	8.8%
Poissy[68]	France	ICU	107	R	CD	100% SD	20.6%
Whyte[64]	UK	ICU	222	R	CD	100% SD	16.2%

Legend: ICU = intensive care unit, R = retrospective, P = prospective, CD = clinical diagnosis, SD = standard dose, ID = intermediate dose, FD = full dose.

ward for a moderate-severe disease, and at least 70% of them received a daily thromboprophylaxis with LMWHs (enoxaparin was the most used) at standard or intermediate dosages. They had an objectively confirmed diagnosis of VTE performed either systematically or to indicate a clinical suspicion. In these studies, the incidence rate of VTE varied between 0% and 8.3%. Only in the study by Fauvel et al[60], therapeutic (OR=0.87, 95% CI 0.82-0.92) and prophylactic doses (OR=0.83, 95% CI 0.79-0.85) of anticoagulation were protective against PE-PT.

As expected, the worsening of SARS-CoV-2 has led to an increased rate of VTE in ICU-patients who requires mechanical ventilation. Several studies (Table II) showed that the VTE rate in ICU-patients with 2019-nCoV pneumonia was higher than both ICU-patients without SARS-CoV2 and medical ward-patients[58,59,64,65-68]. Prophylactic and intermediate dosages of UFH or LMWHs were administered to all the patients enrolled in these studies. The incidence of VTE ranges widely from 6.2% to 49%.

In a recent meta-analysis on 48 studies[69], considering 18.093 SARS-CoV-2 patients, mean age between 52 and 71 years, the use or non-use of heparin does not appear to have had a significant impact on the incidence of VTE. In patients who did not receive a thromboprophylaxis VTE occurred in 21.0% (95% CI: 2.8-48.9) of them, while VTE happened in 18.2% (12.6-24.5) and 19.4% (10.5-30.2) of the patients treated with prophylactic or intermediate-full dose of heparin, respectively. However, the incidence rate of VTE was higher when considering only studies that performed a systematic screening for venous thrombotic events (33%), in ICU-patients (27%), and in prospective studies (25.5%).

Regarding the safety of heparins in treating SARS-CoV-2 patients, five studies[58,62,65,70,71] (Table III) reported an incidence rate of major bleeding ranging from 2.3% to 21%. The highest incidence of any bleeding was reported in patients receiving intermediate or full dose of anticoagulants (21%).

Table III. Studies on hemorrhages in 2019-nCoV infected patients treated with heparins.

Studies	Country	Setting	Patients n	Design	Bleeding assessment	Heparins treatment	Major Bleeding %
Al-Samkari[58]	USA	ICU	144	R	Yes, WHO gs	2% none, 89% SD, 9% ID	2.3%
Helms[65]	France	ICU	150	P	Yes, NR	70% SD, 30% FD	2.7%
Patell[70]	USA	NR	399	R	Yes, ISTH cr	7% none, 67% SD, 22% ID, 38% FD	21%
Pesavento[62]	Italy	MW	324	R	Yes, ISTH cr	74% SD, 26% ID	HR = 3.89 95% CI 1.9-8
Xu[71]	China	ICU = 15 MW = 123 MW= 123	138	R	Yes, NR	70% none, 30% SD	6.7%

Legend: ICU = intensive care unit, MW = medical ward, NR = not reported, R = retrospective, P = prospective, WHO gs = World Health Organization grading system, ISTH cr = International Society of Thrombosis and Haemostasis criteria, HR (95% CI) = hazard ratio and 95% Confidence Intervals.

However, this finding was significantly lower in the only published prospective study[65] which reported a bleeding rate of 2.7%.

Histopathological Findings

The post-mortem examination of 2019-nCoV infected patients was not widely performed at the start of the pandemic because of the concern of contagion.

Initial knowledge of microscopic lesions came from lung biopsy, and histological evaluation carried out in a small number of patients. Lung injuries were characterized by the presence of diffuse alveolar damage with intra-alveolar fibrinous exudates, hyaline membranes that covered the alveolar wall, and desquamation of alveolar epithelial cells type 2, while the alveolar septa showed serious inflammatory infiltrations and widespread capillary microthrombosis[72,73]. These morphological lesions were represented as a clinical picture of a distress respiratory syndrome (ARDS).

Autopsies became more frequent when the Center for Disease Control (CDC) and the World Health Organization (WHO) published their guidelines in May 2020[74,75].

Even if the lung injuries were more frequent, other organs such as heart, kidneys, small bowel, liver, brain, and skin could be involved, and thrombosis appears as the result of inflammation, complement activation, coagulopathy, and the severity of the host response to viral entry[76,77].

Autopsies better defined the vasculature lesions showing acute capillarities associated to complement activation and NETs with fibrin deposition in the pulmonary vessels. Asymptomatic deep vein thrombosis was reported by Wichmann et al[78] in 58% of the patients who underwent the autopsy, and pulmonary embolism was the cause of death in 33% of them.

Not only capillaries but also the main and small-mid-sized pulmonary arteries could be affected by fibrin deposition. Thrombosis has also been detected in patients who received prophylactic dosage of heparins suggesting their failure in preventing both venous and arterial thrombosis[79].

Heparins and Mortality rate in SARS-CoV-2

There are no randomized clinical trials on mortality rate and the use of heparin in SARS-CoV-2 patients. The largest published studies are retrospective (n=2), observational (n=2) and cohort studies (n=1). Results show that anticoagulation in general, and heparin use in particular, reduces mortality rate in SARS-CoV-2 patients of up 52%. However, some authors did not report neither the heparin dosage used in these patients nor the severity of the disease.

In the retrospective study by Ayerbe et al[80], mortality rate has been evaluated in 2075 SARS-CoV-2 patients admitted to 17 Spanish hospitals. Of those patients, 1734 have been treated with heparins. No information is available regard to dosages and type of heparins used. The median follow-up time was 8 days. The results showed that heparins were associated with 45% reduction in mortality (OR=0.55, 95% CI 0.37-0.82). The results did not change when adjusted for age and gender (OR=0.55, 95% CI 0.37-0.82), oxygen saturation, and hyperpyrexia (OR=0.54, 95% CI 0.36-0.82), and concomitant use of other drugs (OR=0.42, 95% CI 0.26-0.66).

In the retrospective study by Nadkarni et al[81] 4.389 patients with SARS-CoV-2 have been examined. A total of 2430 (65%) patients were on anticoagulant therapy, while 1530 (35%) were not. The anticoagulants used were direct oral anticoagulants (DOACs), UFH, and LMWHs in prophylactic and therapeutic dosages. In general, compared to no anticoagulation, therapeutic (n=900; 20.5%) and prophylactic anticoagulation (n=1.959; 44.6%) were associated with lower in-hospital mortality (aHR=0.53; 95% CI 0.45-0.62 and aHR=0.50, 95% CI 0.45-0.57, respectively).

Paranjpe et al[82] also showed an improved median survival in anticoagulated SARS-CoV-2 patients when compared to patients who did not (21 vs 14 days, respectively). Interestingly, this result did not change when patients who require mechanical ventilation were considered. In this subgroup, the median survival was 21 days in anticoagulated patients versus 9 days in those who did not receive any anticoagulant treatment. Moreover, in the study by Albani et al[83] enoxaparin at dosage of 40 mg daily both reduced the in-hospital mortality of 47% and the risk of intensive care admission of up 52% when compared to no enoxaparin treatment.

Finally, in the cohort study by Billett et al[84] a significant decrease in mortality with prophylactic use of apixaban (OR=0.46, p=0.001) and enoxaparin (OR=0.49, p=0.001) was showed in 3625 patients with 2019-nCoV infection when compared to not anticoagulated patients. This study also showed that anticoagulation could be of more benefit in patients who showed a D-Dimer plasma level >10 μg/ml.

Ongoing Randomized Clinical Trials

Several randomized clinical trials are now in progress to evaluate the efficacy and safety of UFH or LMWH in 2019-nCoV infected patients. Most of them are designed to assess the best dosages of heparins. No randomized clinical trials were planned to evaluate the efficacy and safety of Fondaparinux in these patients since the drug, surprisingly, reached a lower priority than both UFH and LMWH by an experts' Consensus[85,86].

Therapeutic or intermediate dose of anticoagulation will be compared with prophylactic dose of intravenous UFH or subcutaneous LMWH in five studies (NCT04401293, NCT04345848, NCT04505774, NCT04406389, NCT04406389, and NCT04408235). It is estimated that, in these studies, a sample of patients between 186 and 2000 will be enrolled to evaluate the incidence of arterial and/or venous thromboembolism, the 30-day mortality, a possible disseminated intravascular coagulation, and the need for ventilatory or vasopressor support at 21 days. These studies should be ended between March-April and December 2021. A French study (NCT04373707) is comparing weight adjusted prophylactic to low prophylactic dosage of LMWH in 602 patients with the aim to evaluate the incidence of VTE and VTE-related death after 28 weeks. The results are attended by November 2021.

Moreover, an Egyptian-Argentine research (NCT04584580) has compared the therapeutic dosage of LMWH to LMWH adjusted for D-Dimer levels and weight to evaluate the incidence of arterial or venous thrombotic events and mortality. The study should have been ended in December 2020. Some other investigations (NCT04485429, NCT04528888 and NCT04600141) will assess the efficacy of heparins when administered with immunosuppressant drugs as tocilizumab or methylprednisolone with the aim to evaluate the need for mechanical ventilation, all-cause mortality at 28 days, and the proportion of patients with clinical improvement at 30 days. Results are expected by December 2021.

Finally, the ETHIC trial (NCT04492254) is the only one that will assess the efficacy of heparins in patients with 2019-nCoV infection but not yet admitted to a hospital. The treatment arm will group patients on enoxaparin 40 mg a day if the bodyweight is <100 Kg or 40 mg twice a day if the bodyweight is >100 kg. The comparator arm will comprise patients on standard care without enoxaparin. The primary endpoint is the incidence of hospital admission and death at 21, 50, and 90 days. The study will end in July 2021.

Some trials will also evaluate the efficacy and safety of nebulized UFH that has been studied in patients with lung injury, showing a reduction in fibrin deposition, microvascular thrombosis, and the need for mechanical ventilation[87].

Up to now, seven clinical trials are recruiting patients: three in USA (NCT04723563, NCT04545541, NCT04397510) and one in Argentina (NCT04530578), Egypt-Argentina (NCT04635241), Brazil (NCT04743011), and Ireland (NCT04511923), respectively. The number of patients estimated to enroll varies between 40 and 712. Nebulized UFH at a dose of 25000 UI 4 times a day will be compared to either placebo or standard care in six studies, while in the Argentine study, 15000 UI a day of nebulized UFH plus subcutaneous enoxaparin 40-60 mg a day, adjusted for body weight, will be compared to subcutaneous enoxaparin 40-60 mg a day. The primary outcomes are the percentage of patients who will require an invasive mechanical ventilation, the changes in D-Dimer levels, aPTT and viral load, the incidence of HIT, the safety of inhaled heparin and mortality. Results are expected by June 2022.

Why Did Heparins Fail to Prevent VTE in 2019-nCoV Infected Patients?

The heparins' failure in preventing VTE events in 2019-nCoV infected patients deserves some thought.

Prophylactic dosages of heparins were those most frequently used, but the inflammatory burden and the severity of the disease in patients with 2019-nCoV infection can highly vary among subjects[88]. In particular, heparins were administered to patients with moderate-severe disease in whom both the degree of inflammation and the clotting potential seem to be more important than that in patients with non-2019-nCoV pneumonia. Therefore, it is plausible that a greater increase in acute phase proteins is present in these patients, thus enhancing the share of heparins that are sequestered by the bloodstream. Not only UFH but also LMWHs, although to a lesser extent, bind to acute phase proteins, endothelial cells, and macrophages, thus potentially reducing the expected anticoagulant effect of the standard dose administered[12,13,89]. Especially in ICU-patients in whom the incidence of VTE is higher than that of patients in medical ward, the reduced efficacy of heparin therapy can be a direct consequence of the cytokines storm, an expression of severe

inflammation and therefore of the high concentrations of acute phase proteins. Therefore, it is reasonable to think that starting heparins at intermediate or therapeutic dosages when the disease is mild or moderate, and thrombosis has not yet occurred, can help in reducing the thrombotic and inflammatory burden in these patients. The clinical effect of such dosages may be similar to that of patients on chronic anticoagulation who seem to have a better clinical course of 2019-nCoV infection, as recently stated by Harenberg et al[90]. On the other hand, atrial fibrillation and VTE, that require an anticoagulant therapy, are not mortality risk factors in SARS-CoV-2 patients such as older age, hypertension, diabetes, cancer and chronic kidney disease[54]. Moreover, several studies have showed that all-case mortality are reduced in chronic anticoagulated patients with 2019-nCoV pneumonia[91,92].

Finally, these patients on either LMWH or Fondaparinux may benefit from a dosage of the anti-factor Xa activity as it happens in other particular clinical conditions such as pregnancy and extreme body weights[93].

Another crucial point is related to AT, a natural anticoagulant, that plays an important role in the heparins' anticoagulant activity. Several studies reported a decrease of the AT plasma levels (<70%) in SARS-CoV-2 patients suggesting that this reduction could be a possible cause of heparin failure[94]. Nevertheless, randomized clinical trials establishing the cut-off at which AT should be supplemented and its efficacy in preventing VTE are necessary.

Another point to be considered is the short half-life of heparins that could have an important impact on VTE occurrence. We speculate that 3-6 hours of anticoagulation a day may not be enough to counterbalance a hypercoagulable state such as that induced by 2019-nCoV infection. A longer half-life of heparins could be required. Fondaparinux could overcome this problem but this is only a pharmacological consideration since clinical trials are still lacking on this topic.

Finally, the question of why heparins fail to prevent VTE but is able to reduce the mortality rate of about 50% in these patients deserves, in our opinion, some considerations. Recently, three clinical trials [Randomized, Embedded, Multi-factorial Adaptive Platform Trial for Community-Acquired Pneumonia (REMAP-CAP), Therapeutic Anticoagulation; Accelerating COVID-19 Therapeutic Interventions and Vaccines-4 (ACTIV-4) Antithrombotics Inpatient and Antithrombotic Therapy to Ameliorate Complications of COVID-19 (ATTACC)] have stopped patient's enrollment since full dose was superior to prophylactic dosage of heparins in hospitalized patients with regard to the need for ventilation or other organs support[95]. These results suggest that higher dosages of heparins may be necessary in SARS-CoV-2 patients, and perhaps their anti-inflammatory and anti-viral properties significantly contribute to reducing mortality.

Conclusions

This narrative review focuses on heparin use in patients with SARS-CoV-2. During the course of the infection, VTE is a common complication which can worse the outcomes of the disease. Literature data show that heparins at prophylactic dosages were not able to reduce the rate of VTE, especially in ICU-patients. However, recently, three randomized clinical trials stopped patients' enrollment since therapeutic dosages of heparins allow to decrease the need of mechanical ventilation and organ support in medical ward-patients.

Heparins are also able to reduce mortality in SARS-CoV-2 patients probably because of their anti-inflammatory and anti-viral properties. Data on the use of these anticoagulants in an early phase of the 2019-nCoV infection are lacking.

Conflict of Interest

The Authors declare that they have no conflict of interests.

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