STUDY PROTOCOL

Version 1. 25.11.2020

Study Title

EFFICACY AND SAFETY OF HEPARIN THROMBOPROPHYLAXIS IN PATIENTS WITH COVID-19 AND RESPIRATORY FAILURE: AN OPEN LABEL RANDOMIZED STUDY. The RESPECT-COVID (RESPiratory failure and hEparin Clinical Ttrial in patients with COVID-19)

CRU-Unipg 2020-12

EudraCT 2020-005884-29

SYNOPSYS

STUDY TITLE	**EFFICACY AND SAFETY OF HEPARIN THROMBOPROPHYLAXIS IN PATIENTS WITH COVID-19 AND RESPIRATORY FAILURE: AN OPEN LABEL RANDOMIZED STUDY. The RESPECT-COVID (RESPiratory failure and hEparin Clinical Ttrial in patients with COVID-19)**
Sponsor/Promoter	Clinical Research Unit, Department of Medicine and Surgery, University of Perugia, Perugia, Italy
BACKGROUND	A new coronavirus (SARS-CoV-2) has recently been detected able to infect humans and induce pneumonia and severe acute respiratory syndrome (COVID-19), similarly to other coronaviruses. From recent experiences, a disproportionately high incidence of thrombosis in COVID-19 patients despite regular thromboprophylaxis has been reported. Moreover, autoptic data have shown diffuse lung micro-thrombosis in patients who died due to COVID-19. Low molecular weight heparin (LMWH) prophylaxis is the current standard of care for COVID-19 patients admitted to the hospital. However, several cohort studies reported a high incidence of venous thromboembolism (up to 25%) in COVID-19 patients with severe respiratory failure receiving standard thromboprophylaxis. Preliminary data suggested an improvement in clinical outcome of patients with severe COVID-19 receiving heparin treatment. About 20% absolute risk reduction (40% relative risk reduction) in mortality at 28 days was reported in patients with D-dimer above 6-fold the upper limit of normal.
STUDY AIMS	The aims of this study, in patients with confirmed COVID-19 and respiratory failure receiving either standard LMWH or intermediate LMWH thromboprophylaxis, are: Primary a) to assess whether intermediate dose LMWH is more effective than standard LMWH prophylaxis in reducing the composite of all-cause death or invasive mechanical ventilation or venous thromboembolic events; Secondary b) to assess whether intermediate dose LMWH is more effective than standard LMWH prophylaxis in reducing venous thromboembolic events; c) to assess the safety of intermediate dose LMWH in comparison to standard LMWH prophylaxis in the incidence of major or clinically relevant non-major bleedings.
STUDY DESIGN	Multicenter, randomized open-label (1:1)
STUDY PHASE	Study phase III
STUDY POPULATION	All patients with confirmed SARS-CoV-2 infection and respiratory failure observed at the study centers will be evaluated for inclusion in the study.
INCLUSION AND EXCLUSION CRITERIA	Inclusion criteria: - Age ≥ 18 years - Diagnosis of COVID-19 confirmed by RT-PCR with pneumonia detected by chest X-ray or ultrasonography or computed tomography - Severe respiratory failure (PaO2/FiO2 <250) - Informed consent Exclusion criteria: - Need for invasive mechanical ventilation - Any contraindication to LMWH (known hypersensitivity, history of heparin induced thrombocytopenia, stage IV or V renal failure,..) - Platelet count <30 x10^9/L

	- Indication for therapeutic anticoagulation - Evidence of active bleeding - Recent stroke in the preceding 4 weeks - Women who are pregnant or breastfeeding
INTERVENTION	Patients will be randomized to receive: - Standard LMWH prophylaxis: LMWH at usual prophylactic dose - Intermediate LMWH prophylaxis: LMWH at 2/3 of the therapeutic dose
OUTCOMES	The primary study outcome is the composite of all-cause death or respiratory failure requiring invasive mechanical ventilation or confirmed venous thromboembolism, whichever comes first, within 28 days from randomization. The secondary study outcomes are: a) confirmed venous thromboembolism within 28 days from randomization; b) major or clinically relevant non-major bleedings within 28 days from randomization. c) confirmed venous thromboembolism or persistent respiratory failure with need for oxygen therapy at 3 months from randomization.
PLANNED NUMBER OF PARTICIPANTS	Assuming a 20% rate of the composite primary outcome in the standard prophylaxis LMWH arm, 326 patients will be required in each arm to have 80% power at a one-sided alpha level of 0.05% to show the superiority of intermediate dose LMWH over standard LMWH in reducing the incidence of the primary outcome by 40%. Patients will be randomized 1:1 to standard LMWH prophylaxis or intermediate dose LMWH.
TREATMENT ALLOCATION	A randomization sequence will be centrally generated at the Coordinating center. Randomization will be performed through a direct contact with the Coordinating center through an over the clock telephone service.
TREATMENT DURATION	From randomization to discharge or 4 weeks, whichever comes first.
FOLLOW UP DURATION	3 months.
STUDY FEASIBILITY	In order to complete patient accrual in 6 months, 15 study centers are required. Centers will be selected based on the volume of COVID 19 patients assisted in October and November 2020 and on expertise in clinical research.

TABLE OF CONTENTS

1. LIST OF ABBREVIATIONS

COVID-19 Novel coronavirus disease 2019

CRF Case Report Form

CT Computed Tomography

DVT Deep Vein Thrombosis

LMWH low molecular weight heparin

PE Pulmonary Embolism

SARS-CoV-2 Severe Acute Respiratory Syndrome Coronavirus 2

VTE Venous Thromboembolism

WHO World Health Organization

2. INTRODUCTION

A novel coronavirus was detected (severe acute respiratory syndrome coronavirus 2 [SARS-CoV-2]) at the end of 2019, capable of infecting humans and inducing pneumonia and severe acute respiratory syndrome, similarly to other coronavirus pneumonia.

The clinical manifestations of the SARS-CoV2 infection (coronavirus associated disease [COVID-19]) are heterogeneous; varying from asymptomatic carriers to acute respiratory disease, pneumonia, multi-organ failure; and can lead to rapid death in a proportion of patients. Evidence regarding the infection is lacking at present and without targeted therapies against the virus, treatment options in those who become unwell are limited. In many cases, only supportive care is feasible with moderate-high mortality rates. COVID-19 can infect anyone however, those with existing co-morbidities are at greater risk of developing complications leading to poorer outcomes.

SARS-CoV-2 infection seems to induce in most critical cases an excessive and aberrant hyper-inflammatory host immune response that is associated with a so-called "cytokine storm", characterized by plasma increase of infection-related biomarkers and of many cytokines and chemokines documented in many observational studies. Pro-thrombotic derangements of the hemostatic system are other common findings in most severe forms of COVID-19 infections, which may be explained by the activation of the coagulation cascade primed by inflammatory stimuli, as also observed in many other forms of sepsis. Indeed, several studies have demonstrated the close interconnection between thrombosis and inflammation, two processes mutually reinforcing each other.

The net effect of the excess of thrombin generation and fibrinolytic shutdown is the production of a profound hypercoagulable state. Further evidence for this includes occlusion and microthrombosis formation in pulmonary small vessels of critical patient with COVID-19 that has been reported. From recent experiences, a disproportionately high incidence of thrombosis (DVT, PE overt and incidental subsegmental, arterial events) in COVID-19 patients despite regular thromboprophylaxis has been observed. Emerging autopsy findings from Italian pathologists reporting that COVID-19 is associated with microthrombosis in the lung, liver and portal vessels, aside overt thrombosis.

Prophylactic dose LMWH is the current standard of care for patients admitted to hospital with COVID-19. Preliminary data have shown that, heparin treatment appeared to be associated with better prognosis in severe COVID-19 patients with a ~20% absolute risk reduction (40% relative risk reduction) in mortality at 28 days where the D-dimer is >6-fold the upper limit of normal. The heparin used was mainly prophylactic to high-prophylactic LMWH which was given in approximately 22% of the Chinese patients. On this background, some data have been reported that despite standard thromboprophylaxis numerous treatment failures are occurring. Based on these findings, the Italian Agency of Drugs suggested a dose higher than standard LMWH prophylaxis in patients with severe respiratory failure and COVID-19.

Thus, we propose a randomized open label trial to compare the impact of standard versus more intensive anticoagulation as thromboprophylaxis in patients admitted to Hospital with COVID-19 and respiratory failure.

3. **AIMS OF THE STUDY**

The aims of this study, in patients with confirmed COVID-19 and respiratory failure receiving standard prophylaxis with LMWH or intermediate LMWH thromboprophylaxis, are:

Primary a) to assess whether intermediate dose LMWH is more effective than standard LMWH prophylaxis in reducing the composite of all-cause death or invasive mechanical ventilation or venous thromboembolic events;

Secondary b) to assess whether intermediate dose LMWH is more effective than standard LMWH prophylaxis in reducing venous thromboembolic events; c) to assess the safety of intermediate dose LMWH in comparison to standard LMWH prophylaxis in the incidence of major or clinically relevant non-major bleedings.

4. METHODS

Patients

Inclusion criteria:

- Age ≥ 18 years

- Diagnosis of COVID-19 confirmed by RT-PCR with pneumonia detected by chest X-ray or ultrasonography or computed tomography

- Severe respiratory failure (PaO2/FiO2 <250)

- Informed consent

Exclusion criteria:

- Need for invasive ventilation and endotracheal intubation

- Any contraindication to LMWH (as known hypersensitivity, history of heparin induced thrombocytopenia, stage IV or V renal failure, acute bacterial endocarditis, major trauma, epidural anaesthesia, haemophilia or other significant haemorrhagic disorders, peptic ulcer, recent cerebral haemorrhage, recent surgery to eye and nervous system, spinal anaesthesia)

- Platelet count <30 $x10^9$/L

- Indication for therapeutic anticoagulation

- Evidence of active relevant bleeding

- Recent stroke in the preceding 4 weeks

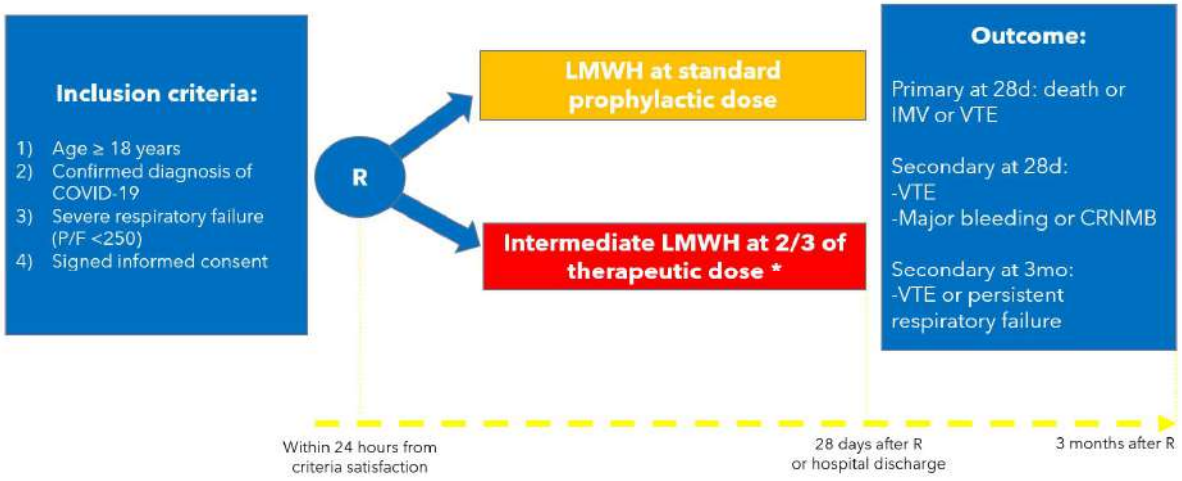
- Women who are pregnant or breastfeeding

Design

This is a multicenter, randomized, open-label trial comparing two different doses of LMWH in patients with confirmed COVID-19 and respiratory failure. Patients will be recruited by their attending medical team. Patients will enter the study as soon as severe respiratory failure (PaO2/FiO2 <250) is diagnosed.

After randomization, study treatment will be administered up to hospital discharge or for a maximum of 4 weeks, whichever comes first. In case of discharge with continuing oxygen therapy further continuation of LMWH prophylaxis will be at the discretion of the attending medical team.
Patients will be followed up to 3 months from randomization.

Patients can withdraw from the study at any time and will revert to standard of care thromboprophylaxis / anticoagulation if needed. The participation in the study will not affect their routine clinical care.



Lower limbs US: at hospital discharge or at 28 days after randomization

* Half dose if creatinine clearance < 30 ml/min or platelet count <50000/mmc

CRNMB=clinically relevant non-major bleeding; IMV=invasive mechanical ventilation; LMWH=low molecular weight heparin; P/F=PaO2/FiO2; US=ultrasonography; VTE=venous thromboembolism

Data collection

The following data will be collected at hospital admission in included patients: clinical, laboratory and radiological findings, results of the RT-PCR for SARS-CoV-2 detection.
The following data will be collected during hospital stay: LMWH treatment allocation, other treatments administered, need for mechanical or non-invasive ventilation, need for ICU, venous thromboembolic events, date of hospital discharge or death. In case of death the cause will be registered.
The following data will be collected at study follow-up: persistent need for oxygen treatment, need for invasive or non-invasive mechanical ventilation, need for ICU, venous thromboembolic events or death. In case of death the cause will be registered.
See case report forms (CRFs) for details.

5. STUDY OUTCOMES

The primary study outcome is the composite of all-cause death or respiratory failure requiring upgrading to invasive mechanical ventilation or confirmed symptomatic or asymptomatic DVT or PE, whichever comes first, within 28 days from randomization.

The secondary study outcomes are:

a) confirmed symptomatic or asymptomatic DVT or PE within 28 days from randomization;
b) major or clinically relevant non-major bleedings within 28 days from randomization;
c) confirmed symptomatic or asymptomatic DVT or PE or persistent respiratory failure with need for oxygen therapy at 3 months from randomization.

Major bleeding events is classified according to the ISTH definition (clinically overt bleeding associated with a decrease in haemoglobin of 2 g/dL or more, bleeding requiring transfusion of two or more units of blood, bleeding occurring in a critical site, or fatal bleeding).

Clinically relevant non-major bleeding (CRNMB) events is defined as acute clinically overt bleeding that did not meet the criteria for major bleeding, but requiring non-surgical, medical intervention by a healthcare professional, leading to hospitalization or increased level of care, or prompting evaluation.

Venous thromboembolism will be defined as a newly diagnosed, objectively confirmed:

a) symptomatic or asymptomatic, proximal or distal lower-limb DVT or
b) symptomatic PE or
c) symptomatic PE in a segmental or more proximal pulmonary artery.

In case of clinical suspicion of a symptomatic deep vein thrombosis or pulmonary embolism, an objective diagnosis should be performed at any time during the study period at discretion of the attending physician.

In order to detect asymptomatic DVT, a lower leg ultrasonography will be performed at hospital discharge or at 28 days from randomization, whichever comes first.

The diagnosis of DVT requires the evidence of one or more filling defects at compression ultrasonography, venography, CT venography or MR venography involving at least the popliteal vein or more proximal veins or peroneal or tibial veins.

The diagnosis of PE requires one or more among:
- an intraluminal filling defect at CT pulmonary angiography;
- an intraluminal filling defect, or a new sudden cut-off of vessels more than 2.5 mm in diameter at pulmonary angiogram;
- a perfusion defect of at least 75% of a segment with a local normal ventilation result (high probability) on ventilation/perfusion lung scan (VQ scan);
- in asymptomatic PE, there must be one or more filling defect in segmental or more-proximal arteries at chest CT pulmonary angiography.

All study outcomes will be centrally adjudicated by independent members unaware of study treatment allocation.

6. STUDY PROCEDURES

All patients with confirmed SARS-CoV-2 infection and respiratory failure observed at the study centers will be evaluated for inclusion in the study.

Informed consent will be obtained prior the participant undergoes any study procedure. This includes collection of identifiable participant data.

Informed consent and randomization should be completed as soon as and within 24 hours from diagnosis of severe respiratory failure (PaO2/FiO2 <250).

Randomization will be in a 1:1 ratio. A randomization sequence will be centrally generated at the Coordinating center. Randomization will be performed through a direct contact with the Coordinating center through an over the clock telephone service.

Patients will be randomized to receive:

- LMWH prophylaxis at standard dose: LMWH at usual prophylactic dose by s.c. injection

- LMWH prophylaxis at intermediate dose: LMWH at 2/3 of the therapeutic dose by s.c. injection

The intervention is any licensed subcutaneously injected LMWH administered at a standard or intermediate dose. LMWH for thromboprophylaxis will be administered subcutaneously.

Nonsteroidal anti-inflammatory drugs (NSAIDs) are discouraged, but will be recorded if used.

Single drug antiplatelet therapy is allowed at a dose of 75 mg daily for clopidogrel, 100 mg/day for acetylsalicylic acid, if indicated for other indications than prevention of VTE according to the discretion of the treating physician.

The use of any LMWH approved for prevention or treatment of VTE and available at the study center is allowed for the study at the following doses:

Treatment Arm	Enoxaparin	Parnaparin	Bemiparin	Nadroparin	Reviparin	Dalteparin
STANDARD	4000 units OD	4250 units OD	3500 units OD	2850 units OD	4200 units OD	5000 units OD
Intermediate	70 U/kg BID	45 U/kg BID	80 U/kg OD	60 U/kg BID	70 U/kg BID	130 U/kg OD
	Max 7000 units BID	Max 9000 units OD	Max 8000 units OD	Max 6000 units BID	Max 7000 units BID	Max 13000 units OD

The LMWHs used will be part of routine hospital pharmacy stock and be maintained within standard hospital pharmacy procedures and practice.

During study treatment a weekly blood sample is advised (hemoglobin, platelet, liver and renal function). In case of creatinine clearance <30 ml/min or platelet count <50000/mmc the dose of study treatment should be halved. Study treatment should be interrupted in case of: creatinine clearance < 15 ml/min or need for dialysis or platelet count <30000/mmc or transaminase >4 times the upper limit or in the presence of major bleeding.

In the need for invasive procedure or surgery, it should be scheduled at least 12 hours since the last LMWH administration. LMWH will be resumed at least 12 hours from achievement of effective hemostasis and at discretion of the attending physician, also according to the associated bleeding risk (see Table).

Procedural bleeding risk stratification

Minimal Risk	Low Risk	High Risk
Minor dermatologic procedures (basal cell carcinoma/squamous cell carcinoma excisions, excision of actinic keratoses, excision of premalignant or cancerous nevi)	Arthroscopy	Any major surgery >45 min in duration
Cataract surgery	Cutaneous biopsies	Any surgery involving neuraxial anesthesia
Minor dental procedures (dental extractions, restorations, prosthetics, endodontics), dental cleanings, fillings	Lymph node biopsies	Major surgery with planned extensive tissue injury
Pacemaker or ICD implantation	Foot/hand surgery	Cancer surgery, especially solid tumor resection
Arthrocentesis or joint injection	Coronary angiography[a]	Major orthopedic surgery, including shoulder replacement surgery
	Gastrointestinal endoscopy ± biopsy	Reconstructive plastic surgery
	Colonoscopy ± biopsy	Urologic surgery (including TURP, TURBT, or tumor ablation, nephrectomy, kidney biopsy)
	Abdominal hysterectomy	Gastrointestinal surgery, especially involving bowel anastomoses (including bowel resection)
	Laparoscopic cholecystectomy	Colonic polyp resection (if unknown at the time of periprocedural planning, colonoscopies should be considered high risk)
	Abdominal hernia repair	PEG placement, ERCP
	Hemorrhoidal surgery	Surgery/biopsies involving highly vascular organs (kidneys, liver, spleen)
	Bronchoscopy ± biopsy	Cardiac surgery
	Epidural injections	Neurosurgery
		Spinal surgery

Study treatment will end at hospital discharge or at 28 days, whichever comes first. In case of persistent need for oxygen therapy at discharge, continuation of LMWH prophylaxis will be at the discretion of the attending medical team.

A follow up phone call will occur at 3 months after randomization.

	Study inclusion	Treatment Phase	Day 28 from randomization or hospital discharge	3 months post randomization
Consent	X			
Inclusion/exclusion criteria and eligibility assessment	X			
Randomization	X			
Allocation to treatment group	X	X		
Data Collection	X	X	X	X
Ultrasonography			X	
Follow Up Call				X

7. STATISTICAL CONSIDERATION

The primary efficacy data set will consist of all randomized subjects who received at least one dose of study drug (modified intention-to treat population). The safety data set (as-treated) will consist of all treated subjects (randomized subjects who received at least one dose of study drug).

Secondary efficacy data sets will consist of all randomized subjects (intent-to treat population, ITT) and the per-protocol population (PP) as defined in the predefined statistical analysis plan. Briefly, the per protocol population will consist of all randomized patients who complete the study fully compliant with the protocol and without any major violation or deviation.

The intention to treat population will be considered for primary analysis. The time to first event of the primary outcome during the 28-days study period will be analyzed using a Cox's proportional hazard model including treatment group. The evaluation of the primary objective will be done by considering the time from randomization to the first VTE or to the need for invasive mechanical ventilation or death.

A descriptive statistical analysis will be carried out to summarize every relevant variable, categorical data will be summarized by using counts and percentages, whilst continuous variables will be presented using the number of patients, mean, standard deviation. Baseline comparisons will be performed by using Chi-squared test or T-test. Comparisons involving time to event data will be displayed by using Kaplan-Maier survival curves and summarized by Hazard Ratios (HRs). All measures of association will be presented with their 95% confidence interval.

The rate of major bleeding (the primary safety outcome) and clinically relevant non major bleeding in patients treated with intermediate or standard LMWH will be compared by using the safety data set.

Assuming a 20% rate of the composite primary outcome with standard LMWH prophylaxis, 326 patients will be required in each treatment arm to show 80% power at a one-sided alpha level of 0.05% to show the superiority of intermediate dose LMWH over standard LMWH in reducing the incidence of the primary outcome by 40%.

8. FEASIBILITY

In order to complete patient accrual in 6 months, 15 study centers are required. Centers will be selected based on the volume of COVID 19 patients assisted in October and November 2020 and on expertise in clinical research.

- Internal, Vascular and Emergency Medicine – Stroke Unit, Santa Maria della Misericordia Hospital, Perugia, Italy (coordinating center, Prof.ssa Cecilia Becattini)

- Pulmonology, Santa Maria della Misericordia Hospital, Perugia, Italy (Dr. Stefano Baglioni)

- Internal Medicine, Santa Maria Hospital, Terni, Italy (Prof. Gaetano Vaudo)

- Infectious Diseases, Santa Maria Hospital, Terni, Italy (Dr. Michele Palumbo)

- Emergency Medicine, ASST Sette Laghi, Varese, Italy (Prof. Walter Ageno)

- Internal Medicine, Media Valle del Tevere Hospital, Marsciano, Italy (Dr. Ugo Paliani)

- Internal Medicine, Gubbio and Gualdo Tadino Hospital, Branca, Italy (Dr. Stefano Radicchia)

- Internal Medicine, San Matteo degli Infermi Hospital, Spoleto, Italy (Dr.ssa Anna Laura Spinelli)

- Internal Medicine, Città di Castello Hospital, Città di Castello, Italy (Dr. Laura Martinelli)

- Internal Medicine, Michele e Pietro Ferrero Hospital, Verduno, Italy (Dr. Fulvio Pomero)

- Internal Medicine, Misericordia Hospital, Grosseto, Italy (Dr. Andrea Montagnani)

- Pulmonology, Misericordia Hospital, Grosseto, Italy (Dr. Bruno Sposato)

- Internal Medicine, San Giuseppe Hospital, Empoli, Italy (Dr. Luca Masotti)

- Internal Medicine, San Giuseppe Hospital, Empoli, Italy (Dr. Simone Vanni)

- Emergency Medicine and Surgery, S. Anna and S. Sebastiano Hospital, Caserta, Italy (Dr.ssa Iolanda Enea)

REFERENCES

1. Guan WJ, Ni ZY, Hu Y, et al. Clinical Characteristics of Coronavirus Disease 2019 in China. N Engl J Med. 2020;382(18):1708-1720. doi:10.1056/NEJMoa2002032
2. Wu Z, McGoogan JM. Characteristics of and Important Lessons From the Coronavirus Disease 2019 (COVID-19) Outbreak in China: Summary of a Report of 72 314 Cases From the Chinese Center for Disease Control and Prevention [published online ahead of print, 2020 Feb 24]. JAMA. 2020;10.1001/jama.2020.2648. doi:10.1001/jama.2020.2648
3. Jackson SP, Darbousset R,Schoenwaelder SM, et al. Thromboinflammation: challenges of therapeutically targeting coagulation and other host defense mechanisms. Blood. 2019; 133:906-918.
4. Iba T, Levy JH. Inflammation and thrombosis: roles of neutrophils, platelets and endothelial cells and their interactions in thrombus formation during sepsis. J Thromb Haemost 2018; 16: 231–41.
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 Feb 19.
6. Tang N, Bai H, Chen X, Gong J, Li D, Sun Z. Anticoagulant treatment is associated with decreased mortality in severe coronavirus disease 2019 patients with coagulopathy. J Thromb Haemost. 2020 Mar 27. doi: 10.1111/jth.14817.
7. Tcachil J, Tang N, Gando S et al. ISTH interim guidance on recognition and management of coagulopathy in COVID-19. Journal Thomb Haemost first published: 25 March 2020 doi: 10.1111/jth.14810
8. Marietta M, Ageno W, Artoni A et al. COVID-19 and haemostasis: a position paper from Italian Society on Thrombosis and Haemostasis (SISET). Blood Transfus. 2020 Apr 8. doi: 10.2450/2020.0083-20
9. Wein L, Wein S, Haas SJ et al. Pharmacological Venous Thromboembolism Prophylaxis in Hospitalized Medical Patients. A Meta-analysis of Randomized Controlled Trials. Arch Intern Med. 2007; 167:1476-1486.
10. Giustozzi M, Vedovati MC, Agnelli G. Venous thromboembolism and COVID-19: Mind the gap between clinical epidemiology and patient management. Eur J Intern Med. 2020 Oct 22:S0953-6205(20)30404-0. doi: 10.1016/j.ejim.2020.10.018. Epub ahead of print. PMID: 33127217; PMCID: PMC7580525.
11. https://www.aifa.gov.it/documents/20142/1123276/Eparine_update_01_24.11.2020.pdf/e6188538-9478-9625-55e3-8dcc5dae02c6