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Supplementary appendix

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Supplementary Appendix

In-hospital and 6-month Outcomes of COVID-19 Patients Supported with Extracorporeal Membrane Oxygenation: the EuroECMO-COVID Multicenter Prospective Observational Study

Table of contents

Authors…….3
Contributors……4
Collaborators……...5
Methods……..13
Data Collection……..15
Supplementary Tables……..16
Supplementary Figures……25
STROBE Checklist……..34

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Supplementary Methods

European Extracorporeal Life Support Organization (EuroELSO) and EuroECMO COVID Project

EuroELSO is the European branch of the Extracorporeal Life Support Organization (ELSO). The objective of the organization is to provide support to institutions delivering extracorporeal life support through continuing education, sharing knowledge, updating on original research and publications.

Due to the rapid growth in the number of critically ill COVID-19 patients in Europe, an unexpected high number of severely compromised patients were considered eligible for extracorporeal membrane oxygenation (ECMO) support. For this reason, the Steering Committee of EuroELSO initiated a prospective observational study among European centers and adjacent countries including Belarus, Israel, Norway, Russian Federation and Switzerland with the intention of providing near-real time information on ECMO use in COVID-19.

The study, named European/EuroELSO Survey on COVID Adult Patient (EuroECMO-COVID), represented the first collaborative investigation to assist locally and globally in response to the COVID-19 outbreak. The aim of the study was to evaluate clinical features, severity of pulmonary dysfunction and risk factors in patients with COVID-19 who needed ECMO support, evaluate the technical characteristics, duration and complication of the extracorporeal support and the in-hospital and at 6 months outcomes of patients requiring ECMO. The data collection started on March 15th 2020 and weekly provided an anonymously report through the EuroELSO website (https://www.euroelso.net/covid-19/covid-19-survey/). The EuroELSO publication policy and data access is available in the EuroELSO website (addendum chapter Coronavirus COVID-19).

Definition of first COVID wave

The definition of first COVID wave (March 1st to September 13th, 2020) applied in the current study was based on official statements of the World Health Organization (who.int/director-general/speeches/detail/WHO-director-General's opening remarks at the media briefing on COVID-19-11 March 2020)which declared the outbreak of COVID-19 as a pandemic on 11th March, 2020 and announced the secondary sanitary crisis at the end of September.

The following waves in European countries have a more nuanced temporal characterization due to the differences in stage of infection, the diverse control strategies (i.e. various lockdowns restrictions), the socio-demographic and socio-economic local characteristics, the different levels of air pollution and meteorological factors that may had influenced the spread of COVID-19 waves across the Europe.

Rationale for first wave analysis

The current analysis of the EuroECMO-COVID Study was designed to evaluate singularly the first wave of the COVID-19 pandemic, while future analyses will focus on the subsequent waves. This study design was based on three major factors:

- The first wave was characterized by exceptional working conditions during an unexpected pandemic. This has pushed European centers to adapt their admission criteria based on a utilitarian medical approach with stricter admission criteria for ICU. Contrarily, the subsequent waves were characterized by more knowledge and greater resources, which have significantly influenced the patients' selection criteria.
- The medical approach to the first wave was significantly different compared to the following waves. Indeed, the first medical protocols were based on previous experiences, such as the one with the Middle East Respiratory Syndrome coronavirus infection epidemic in the Arabic peninsula. Furthermore, the unexpected emergency of COVID-19 during the influenza season has led to the extensive use of antibiotics, neuraminidase enzyme inhibitors, chloroquine, antiviral drugs, and hyperimmune plasma and immunoglobulins. Although some of these therapies might ultimately prove to be beneficial, they all had potential serious adverse event and several of them were abandoned during the following waves. For example, a recent Cochrane Analysis[1] on a widely used medication during the first COVID-19 wave, such as chloroquine and idroxichloroquine, showed that the use of these medications was related to a rate of adverse events that was triple compared to placebo. Furthermore, during the initial pandemic phase, the treatment approaches were selected according to the local access to different types of medications, hospital policies, and resource shortage.
- The patient population of the first wave was considered different from the subsequent waves in terms of time of intubation, mechanical ventilatory approach and discovery of different pattern of lung disease. Based on the results of these first experiences, the following COVID-19 waves could benefit from a more structured clinical approach.

For these reasons, the first pandemic wave was considered as a unique and not a comparable event. Future investigations focused on the following COVID-19 waves are warranted to investigate the evolution of this disease and its clinical approach.

Data Collection

The following predefined variables were collected:

- Demographic data: age, sex, race, weight, height and body mass index
- Patients comorbidities: chronic obstructive pulmonary disease, diabetes, cardiovascular disease, renal insufficiency, arterial hypertension, smoking, obesity
- Pre ECMO characteristics:
    1. Timing: time onset symptoms to intensive care admission, time onset symptoms to orotracheal intubation, time intensive care admission to orotracheal intubation, duration of mechanical ventilation, time orotracheal intubation to tracheostomy
    2. Medical therapy: antibiotics, antiviral drugs, steroids, immunomodulators, convalescent plasma, inotropes, vasopressors, second line therapy (prone therapy and number of proning session, neuromuscular blockage, inhaled nitric oxide)
    3. Gas exchange before ECMO implant: pH, PaO2, PaCO2, FiO2
- ECMO Characteristics:
    1. ECMO configuration (veno-venous, veno-arterial, hybrid or other), time orotracheal intubation to ECMO start, implant indication, type of vascular access (femoro-femoral, double lumen cannula in jugular vein, femoro-jugular, jugular-femoral, femoro-axillary, central cannulation, pulmonary artery cannulation), distal perfusion (when indicated), presence and type of left ventricle venting system, adjunctive treatments to ECMO (renal replace therapy, plasmapheresis, Cytosorb, leukophoresis, molecular adsorbent recirculating system), ECMO configuration change, configuration after change, indication for variation of ECMO configuration, time ECMO onset and ECMO new configuration, maximum blood flow on ECMO
    2. Medical therapy on ECMO: anticoagulation and type (heparin, bivalirudin, argatroban or other), antiplatelets (aspirin, clopidogrel or other)
    3. Complications: multi-organ-failure, sepsis, renal failure, bleeding, neurological complications (ischemic stroke, hemorrhagic stroke, intracranial bleeding, seizure, delirium), gastro-intestinal complications (bowel ischemia, gastro-intestinal bleeding, ileus, gastro-intestinal perforation), pulmonary complications (pneumothorax, pulmonary embolism, hemothorax, pulmonary sovra-infection, pulmonary abscess, acquired hospital pneumonia, ventilatory acquired pneumonia)
- Outcome in-hospital: ECMO weaning, time ECMO start to ECMO weaning, overall time in intensive care unit, overall time in hospital, death, reason for death, time ECMO start to death, death after weaning, time ECMO start to death after weaning, reasons for death after weaning
- Outcome 6 months follow-up: alive at 6 months, lung transplant, time ECMO start to lung transplant, heart transplant, time ECMO start to heart transplant, persistent dyspnea o limited effort at 6 months, persistent oxygen requirements at 6 months, ongoing respiratory rehabilitation at 6 months, still on mechanical ventilation at 6 months, in hospital/nursing home at 6 months, cardiac symptoms at 6 months (tachycardia, exhaustion, new cardiac therapy), cognitive problems at 6 months, back to work at 6 months (part time, full time)

Supplemental Tables

Supplemental Table 1: Variables and outcomes definitions.

VARIABLE	DEFINITION
Comorbidities	
Arterial Hypertension	Systolic blood pressure > 140 mmHg or diastolic blood pressure > 90 mmHg, or use of antihypertensive agent to maintain normal blood pressure
Diabetes	Chronic metabolic disease characterized by elevated level of blood glucose over a prolonged period of time (any type 1, 2, gestational)
COPD	Diagnosis of chronic obstructive pulmonary disease, any Gold classification
Cardiovascular disease	Disease affecting heart and blood vessel including arrhythmia, valve disease, coronary artery disease, heart failure, peripheral artery disease, congenital heart disease, pericardial disease, deep vein thrombosis
Renal Insufficiency	Reduced kidney function due to renal artery disease
Smoking	Active (smoking during the past 30 days) and more than 100 cigarettes during lifetime
Obesity	Excessive fat accumulation (Body mass index > 30)
ECMO details	
1) Indications	
ARDS	Acute diffuse inflammatory lung injury requiring invasive mechanical ventilation or extracorporeal membrane oxygenation
Pneumonia	Any suspected pulmonary infection treated with antibiotics
Septic shock	Sepsis with vasopressors requirement to maintain MAP> 65 mmHg and serum lactate levels greater than 2 mmol/L in absence of hypovolemia
Myocarditis/Cardiac dysfunction	Inflammation of myocardium with necrosis of cardiac myocites due to infection, cardiotoxin, drugs and systemic disorder or idiopathic/ cardiac dysfunction inability to provide the necessary blood flow for metabolic and functional need of vital organs under normal conditions
Isolated ventricular failure	Altered ventricle function due to direct or indirect damage that reduce the pump ability of the heart
Pulmonary Embolism	Condition due to a sudden blockadge to a lung artery
eCPR	Implantation of veno-arterial ECMO in patient who experienced a sudden pulseless condition due to the cessation of cardiac mechanical function
Postpartum	Cardiac and or lung dysfunction related to infection or sepsis, major bleeding, cardiomyopathy, thrombotic pulmonary embolism, stroke, amniotic fluid embolism
2) Type of ECMO	
Veno-venous (V-V)	Configuration able to maintain gas exchange (oxygenation and removal of CO2) in isolated lung failure and preserved cardiac output
Veno-Arterial (V-A)	Configuration that bypass lungs and heart in order to provide respiratory and circulatory support

Veno-ArterialVenous (V-AV)	Configuration with an extra inflow cannula to provide increased delivering of oxygenated blood to the pulmonary circulation and correct differential hypoxemia
Veno-Venoarterial (VVA)	Configuration with an extra arterial perfusion cannula when patient on VV ECMO have hemodynamic deterioration and require cardio-circulatory support
Oxy-RVAD	Right ventricular assist device with an oxygenator using ECMO with drainage cannulation from the femoral vein and return cannulation to the main pulmonary artery
Other	Various others configuration involving more than three cannula to support dynamic evolution during the ECMO course
3) Indications for changing ECMO configuration	
Left ventricular failure	Dysfunction of the left ventricle resulting insufficient delivery of blood to vital organs either with preserved or reduced ejection fraction
Right ventricular failure	Evidence of right-sided structural and/or functional abnormalities in combination with clinical symptoms and signs of RV failure
Bi-ventricular failure	Biventricular dysfunction accompanied by both signs and symptoms of right-sided and left-sided heart failure
Refractory hypoxemia	Persistent PaO2 less than 60 mmHg for at least 1 hour while receiving FiO2 1.0
Cannulation site bleeding	Blood loss from the access cannulation site with inevitable treatment (by cannula removal and compression or by vascular surgery)
Leg ischemia	Clinical signs of lower extremity ischemia requiring intervention (either by vascular surgery or cannula removal)
Drainage problem	Inability to adequately support a patient on ECMO due to impaired blood aspiration from the cannula due to hypovolemia, kinks in the circuit, cannula malposition or inadequate cannula size
Complications	
Renal failure	Renal function at less than 15% of normal levels, either acute or chronic
Major bleeding	According to the ISTH criteria in non -surgical patients is defined as having a symptomatic presentation and is divided in fatal bleeding and/ or bleeding in a critical area or organ and/or bleeding causing a fall in hemoglobin level of 2 g/dL or more, or leading to transfusion of two or more packed red blood cells
Ischemic stroke	Neurological dysfunction by focal brain with clinical symptoms lasting less more than 24 hours, with or without permanent disability
Intracranial bleeding	Bleeding between the brain tissue and skull or within the brain tissue itself

Seizures	Burst of uncontrolled electrical activity between brain cells that causes temporary abnormalities in muscle tone or movements, behaviors, sensations or states of awareness
Delirium	Disturbance in mental abilities that results in confused thinking and reduced awareness of the environment
Bowel ischemia	Intestinal ischemia with elevated lactate levels requiring abdominal surgical intervention
GI bleeding	any type of bleeding that starts in the digestive tract, due to a disease or a condition itself
Ileus requiring medications	Ileus is an occlusion or paralysis of the bowel preventing the passage of intestinal content, causing their accumulation proximal to the site of blockage due to mechanical or functional conditions
GI perforation	any part of the gastrointestinal tract can present (acutely or in an indolent manner) a perforation with consequent releasing of gastric or intestinal content into the peritoneal space
Pneumothorax/Pneumomediastinum	Abnormal collection of air in the pleural space between the lung and the chest wall/abnormal collection of air in the mediastinum
Lung bleeding	Extravasion of blood into airways and/or lung parenchyma
Hemothorax	Collection of blood in the space between the chest wall and the pleural cavity
Pulmonary sovrainfection	secondary infection that occurs during an existing infection, or immediately following a previous infection, particularly caused by microorganisms that are resistant or have become resistant to antibiotics used earlier
Pulmonary abscess	Microbial infection of the lung that results in necrosis of pulmonary parenchima
HAP/VAP	HAP or nosocomial pneumonia refers to any pneumonia contracted by a patient in hospital at least 48-72 hours after being admitted/VAP is a hospital acquired pneumonia that occurs more than 48 hours after having been intubated and received mechanical ventilation
Outcomes	
Respiratory failure	reversible pulmonary disease which cannot anymore be managed by conventional mechanical ventilation and/or ECMO despite pharmacological intervention
cardiac arrest	Abrupt loss of heart function despite acute and simple interventions such as pacing and defibrillation
Neurological injury	Non- traumatic brain injuries caused by lack of oxygen, toxin exposure or pressure from an hemorrhagic or ischemic lesion
Sepsis	Life-threating organ dysfunction caused by a dysregulated host response to an infection

MOF	Hypometabolic state with involvement of more than one organ as established by biochemical and/or radiological analysis
Follow up 6 months	
Persistent Dyspnea	shortness of breath lasting longer than one month, according to ATS chronic dyspnea is a subjective experience of breathing discomfort that consist of qualitatively distinct sensations that vary in intensity. When shortness of breath is greater than expected for a given level of exertion is considered pathologic and a symptom of disease
Persistent requirements Oxygen	LTOT is a treatment to improve survival in patients with chronic severe daytime hypoxemia, can be defined as oxygen used for at least 15 h per day in chronically hypoxaemic patients ($PaO_2 < 7.3$ kPa)
Cardiac symptoms	Temporary or lasting damage due to hypoxic injury, myocarditis, ischemic injury, stress cardiomyopathy, cytokine storm related to COVID-19 infection characterized by arrhythmias, dizziness, chest discomfort and shortness of breath
Cognitive problems	Signs and symptoms that develop during or after COVID-19 infection, continue for more than 12 weeks and not explained by an alternative diagnosis. According to MoCA score: cognitive deficit in memory, executive function and language

Supplemental Table 2- Patients' Characteristics after Multiple Inputations for Cox Regression

	Full Cohort (n=1215)		In-hospital Survivors (n=613)		In-hospital Non Survivors (n=602)		p-value
Age - years	54	(46-61)	51,0	(43-58)	58,0	(50-62)	<0,001
Age - categories							
< 59 years old	840	(69,1%)	489	(79,8%)	351	(58,4%)	
60-69 years old	325	(26,7%)	117	(19,1%)	208	(34,4%)	
≥ 70 years old	50	(4,1%)	7	(1,1%)	43	(7,2%)	
Sex- n, %							
Male	946	(77,9%)	467	(76,2%)	479	(79,5%)	
Female	269	(22,1%)	146	(23,8%)	123	(20,5%)	
Race- n, %							
White (non-hispanic)	880	(72,4%)	417	(68%)	463	(77,0%)	
Asian	162	(13,3%)	99	(16,2%)	63	(10,3%)	
Black	95	(7,8%)	52	(8,5%)	43	(7,2%)	
Hispanic	78	(6,4%)	45	(7,3%)	33	(5,5%)	
BMI	29,2	(26,0-32,9)	29,3	(26,0-33,0)	29,0	(26.0-32.8)	0,593
Co-existing condition- n, %							
BMI class							
BMI ≤ 29.9 Kg/m2	679	(55,9%)	342	(55,8%)	337	(55,9%)	
Obesity class I	324	(26,7%)	163	(26,6%)	161	(26,8%)	
Obesity class II	121	(10%)	55	(9%)	66	(11,0%)	
Obesity class III	91	(7,5%)	53	(8,6%)	38	(6,3%)	
Pre-existing Comorbidities- n,%							
Diabetes	288	(23,7%)	127	(20,7%)	161	(26,8%)	
Arterial Hypertension	507	(41,7%)	220	(35,9%)	287	(47,6%)	
Cardiovascular Disease	149	(12,3%)	55	(9%)	94	(15,5%)	
Renal Failure	55	(4,5%)	15	(2,4%)	40	(6,7%)	
Chronic Obstructive Pulmonary Disease	91	(7,5%)	27	(4,4%)	64	(10,5%)	
COVID-19 Medical therapy- n;%							
Antiviral Drugs	626	(51,5%)	291	(47,5%)	335	(55,6%)	
Pre-ECLS Support n;%							
Prone Therapy before ECLS	1003	(82,6%)	407	(82,9%)	495	(73,7%)	
Inotropes	432	(35,6%)	172	(28,1%)	260	(43,1%)	
Vasopressors	958	(78,8%)	454	(74,1%)	504	(83,9%)	
Time from Intubation to ECLS cannulation - days	4	(2-8)	4	(1-7)	5	(2-9)	0,008
Indication for ECLS - n, %							
ARDS	972	(80%)	495	(80,8%)	477		
Pneumonia	177	(14,6%)	83	(13,5%)	94		
Septic Shock	15	(1,2%)	9	(1,5%)	6		
Myocarditis/ Cardiac Dysfunction	28	(2,3%)	16	(2,6%)	12		
Isolated Ventricular Failure	6	(0,5%)	3	(0,5%)	3		
Pulmonary Embolism	10	(0,8%)	6	(1%)	4		
Cardiac arrest	7	(0,6%)	1	(0,2%)	6		
Type of ECLS- n,%							
VV ECLS	1105	(90,9%)	561	(91,5%)	544		
VA ECLS	89	(7,3%)	42	(6,9%)	47		
V-AV ECLS	10	(0,8%)	7	(1,1%)	3		

VV-A ECLS	7	(0,6%)	1	(0,2%)	6
OxyRVAD	4	(0,3%)	0	(0%)	0
Other ECLS	0	(0%)	2	(0,3%)	2
ECLS Configuration Change- n,%	128	(10,5%)	45	(7,3%)	83
Renal Failure					
No Renal Failure	506	(41,6%)	329	(53,7%)	177
Renal Failure without RRT	445	(36,6%)	171	(27,9%)	274
Renal Failure with RRT	264	(21,7%)	113	(18,4%)	151
Pneumothorax- n,%	236	(19,4%)	91	(14,8%)	145
Ischemic Stroke- n,%	60	(4,9%)	21	(3,4%)	39
Hemorragic Stroke- n,%	73	(6,0%)	19	(3,1%)	54
Intracranial Bleeding- n,%	134	(11%)	47	(7,7%)	87
Gastro-intestinal Bleeding- n,%	139	(11,4%)	60	(9,8%)	79
Bowel Ischemia- n,%	86	(7,1%)	19	(3,1%)	67
Lung Bleeding- n,%	168	(13,8%)	64	(10,4%)	104
Pulmonary Embolism- n,%	172	(14,2%)	87	(14,2%)	85
Pulmonary Sovrainfection- n,%	485	(39,9%)	229	(37,4%)	256

Data are reported as n (% of available data) or median (IQR, interquartile range)Ys: years. BMI, Body Mass Index. ICU: Intensive Care Unit. ECLS: Extracorporeal Membrane Oxygenation: ARDS: Acute Respiratory, Distress Syndrome; ARF: Acute Respiratory Failure. V-V:veno-venous.V-A: veno-arterial.OxyRVAD: Oxygenator in right ventricular assist device. MARS: Molecular Adsorbent Recirculating System.

Supplemental Table 3- Numbers and rates of collected and missing 6-month data.

	Valid (n)	Missing (n)	Valid (%)	Missing (%)
Alive 6 months	577	36	94.13	5.87
Persistent Dyspnea 6 months	523	90	85.32	14.68
Persistent Oxygen 6 months	521	92	84.99	15.01
Respiratory Rehab at 6 months	522	91	85.15	14.85
Still on Mechanical Ventilation	528	85	86.13	13.87
In hospital nursing home	523	90	85.32	14.68
Cardiac symptom 6 months	514	99	83.85	16.15
Cognitive problem	512	101	83.52	16.48
Back to work part time	428	185	69.82	30.18
Back to work full time	431	182	70.31	29.69

Supplemental table 4 - Contributing centers and geographical distribution**.**

	Centers n, %	
Number of Patients per Contributing Center		
0-9	91	(68.4%)
10-19	29	(21.8%)
20-39	9	(6.8%)
40-60	4	(3.0%)
Country		
Austria	5	(3.8%)
Belgium	15	(11.3%)
Czech Republic	2	(1.5%)
Denmark	2	(1.5%)
Estonia	2	(1.5%)
France	9	(6.8%)
Germany	13	(9.8%)
Ireland	1	(0.8%)
Israel	8	(6.0%)
Italy	18	(13.5%)
Lithuania	1	(0.8%)
Poland	1	(0.8%)
Portugal	4	(3.0%)
Russia	5	(3.8%)
Slovenia	1	(0.8%)
Spain	22	(16.5%)
Sweden	4	(3.0%)
Switzerland	4	(3.0%)
The Netherlands	6	(4.5%)
Turkey	4	(3.0%)
United Kingdom	6	(4.5%)

Supplemental Table 5 - Number of patients based on geographical distribution**.**

	Patients n, %	
Country		
Austria	33	(2.7%)
Belgium	98	(8.1%)
Czech Republic	10	(0.8%)
Denmark	26	(2.1%)
Estonia	4	(0.3%)
France	114	(9.4%)
Germany	180	(14.8%)
Ireland	5	(0.4%)
Israel	54	(4.4%)
Italy	134	(11.0%)
Lithuania	5	(0.4%)
Poland	11	(0.9%)
Portugal	47	(3.9%)
Russia	44	(3.6%)
Slovenia	3	(0.2%)
Spain	158	(13.0%)
Sweden	38	(3.1%)
Switzerland	22	(1.8%)
The Netherlands	20	(1.6%)
Turkey	9	(0.7%)
United Kingdom	200	(16.5%)

Supplementary Figure

Figure S1: ROC curve with best predicting cutoff at different time points (0,30, 60, 90, 120, 150, 180 months)

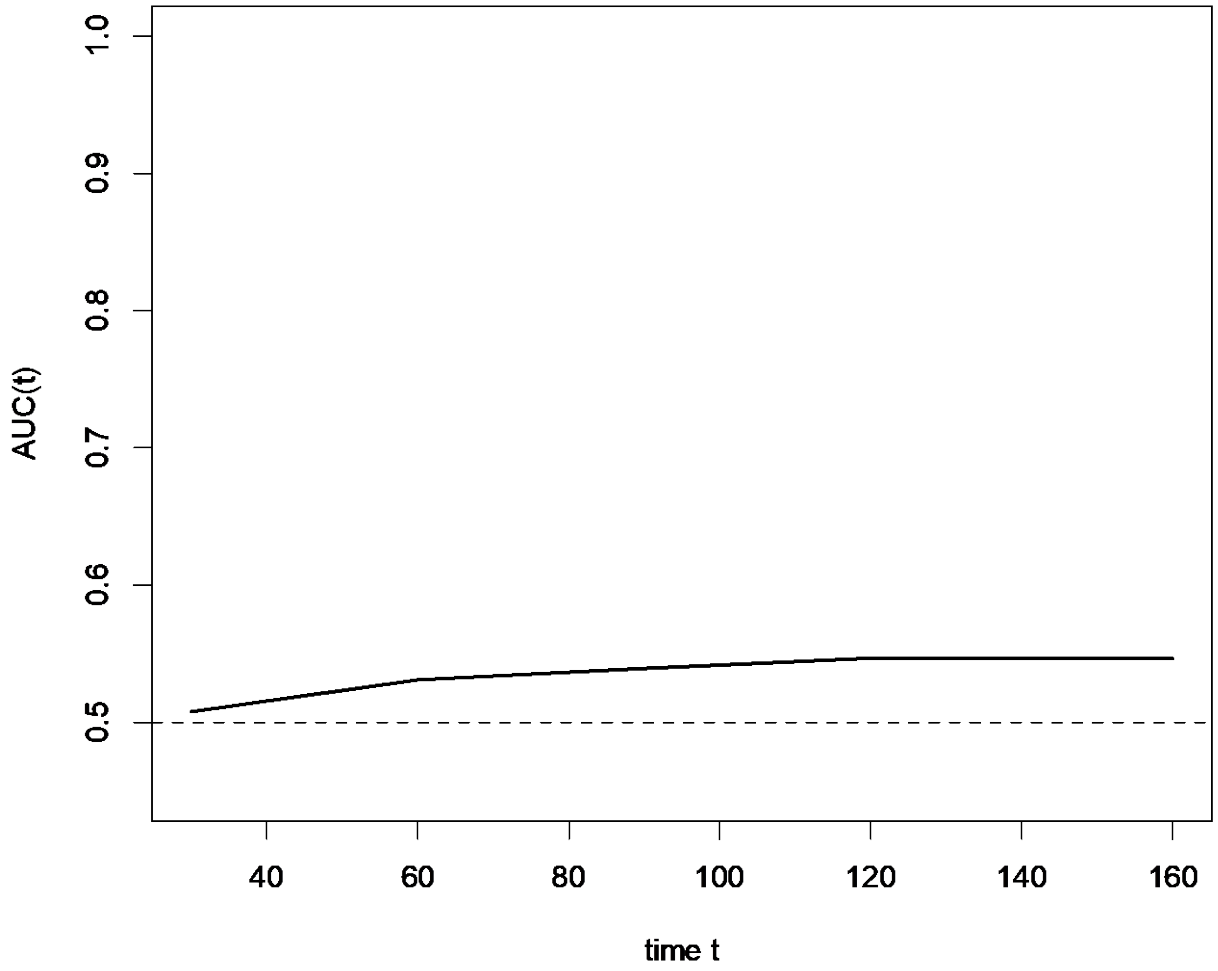


Figure S2: Calibration Plot Model 1

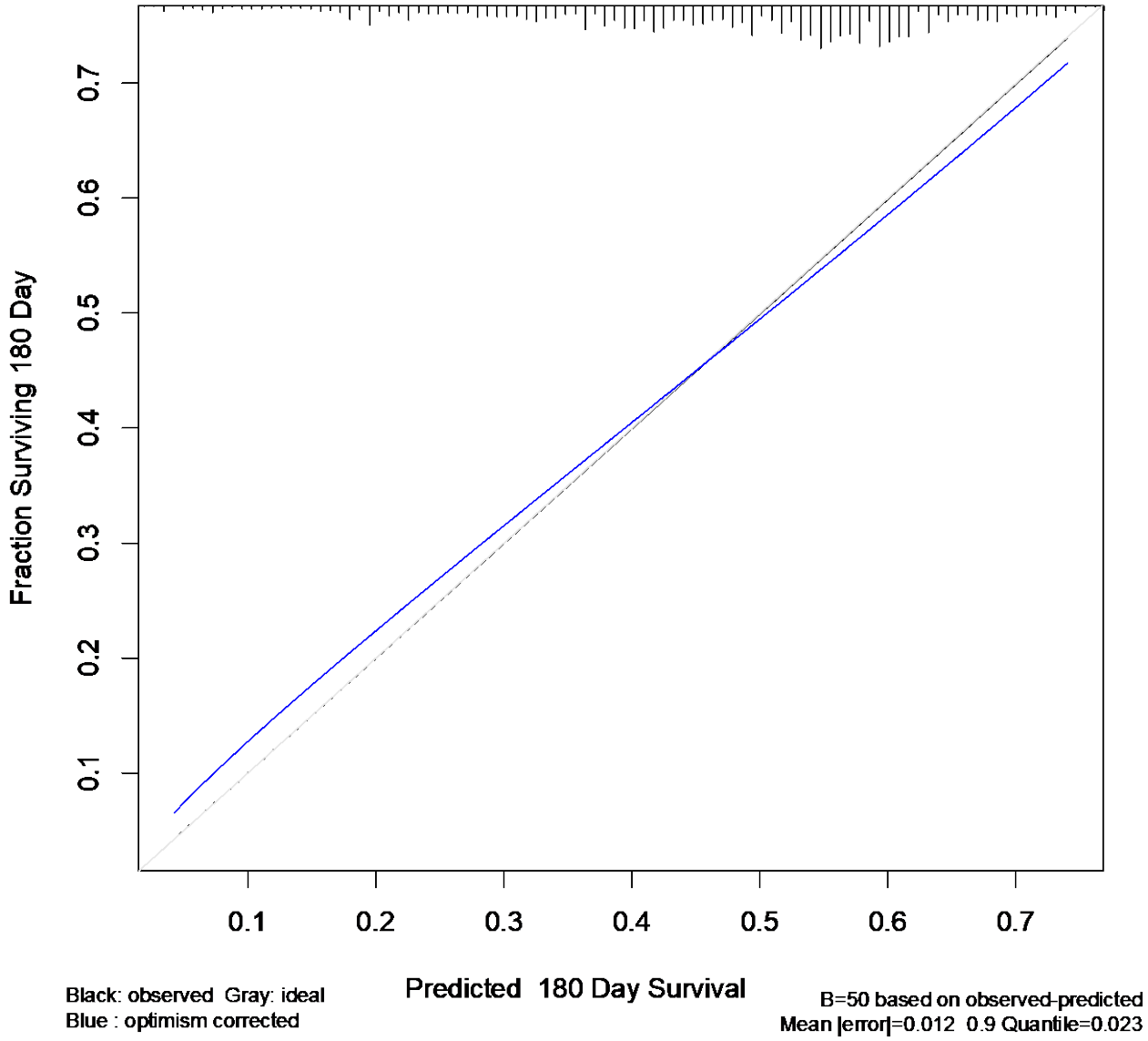
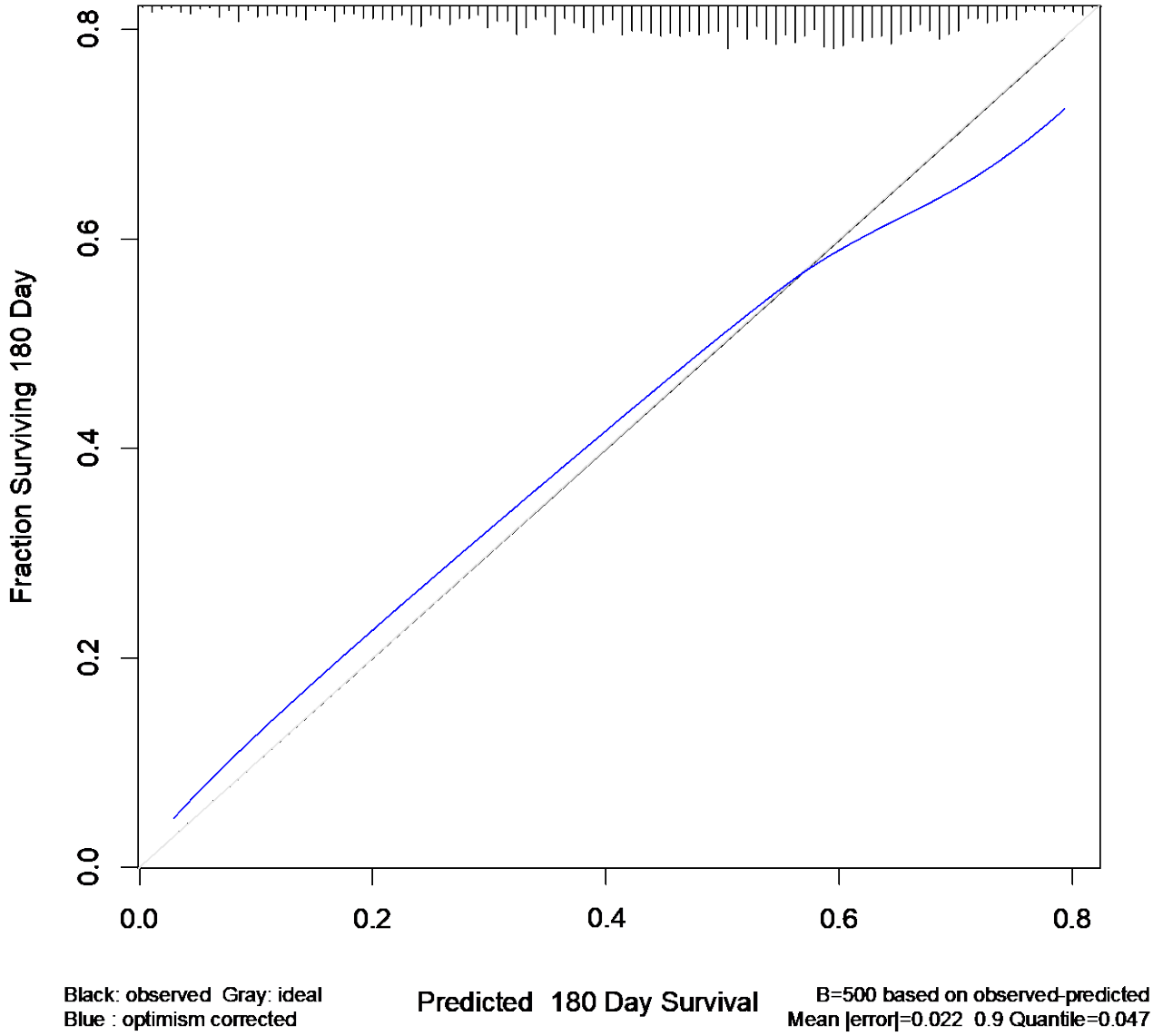


Figure S3: Calibration Plot Model 2



S4: Kaplan Maier curves according to variables significantly associated with in-hospital mortality (Model 1)

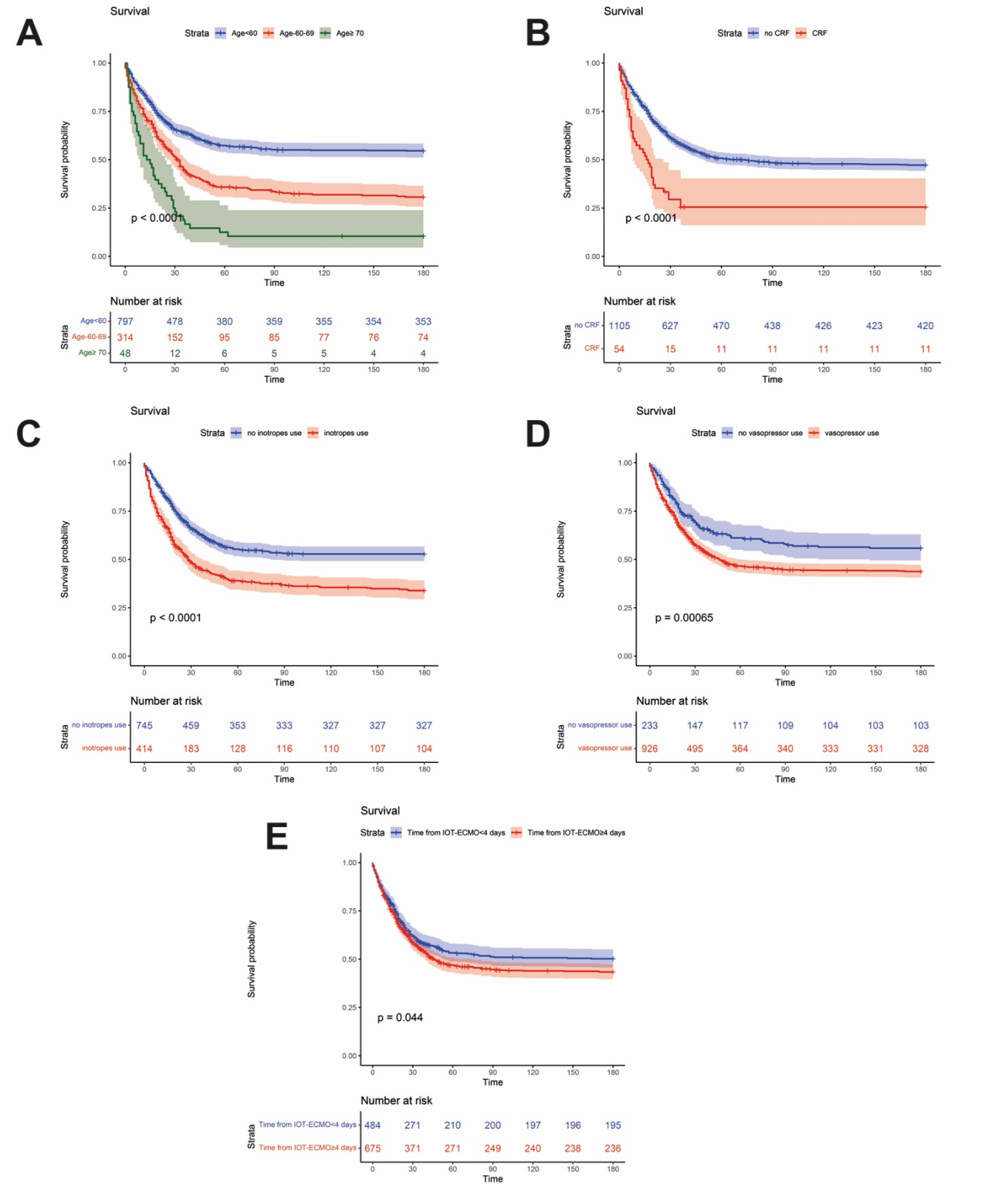


Figure S5: Kaplan Maier curves according to variables significantly associated with in-hospital mortality (Model 2)

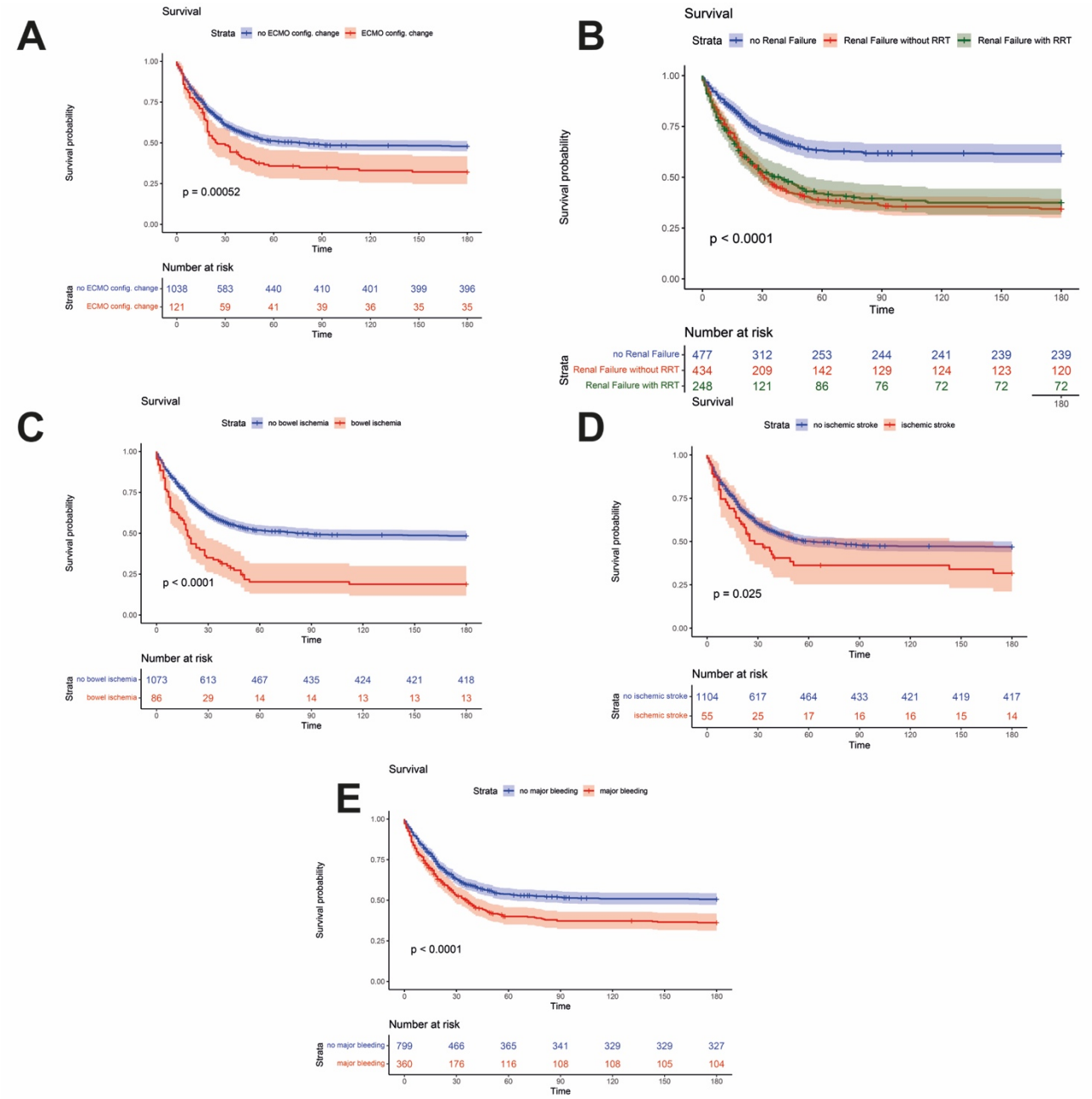
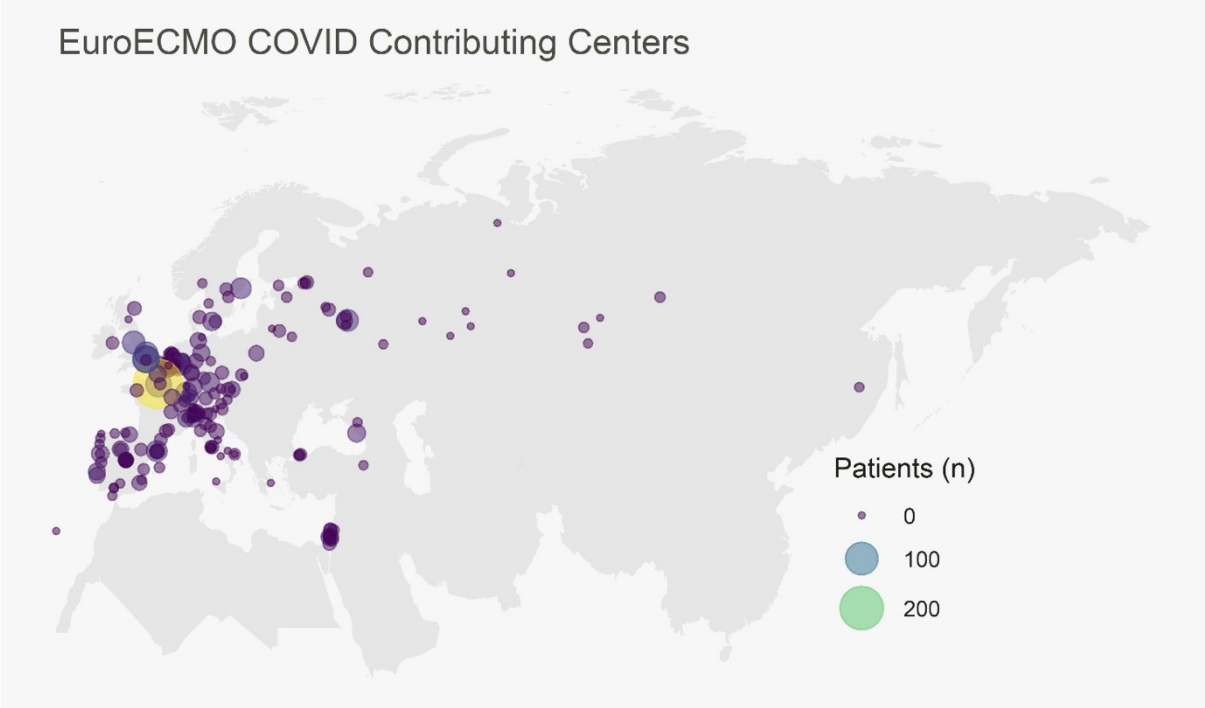


Figure S6: Geographical distribution of EuroECMO-COVID Contributing Centers.



STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation
Title and abstract	1	Indicate the study's design with a commonly used term in the title or the abstract Multi-center observational study as stated in the Abstract pag 3 and Methods pag 4-5
		Provide in the abstract an informative and balanced summary of what was done and what was found Provided in Abstract pag 3
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported Included in Introduction pag 4
Objectives	3	State specific objectives, including any prespecified hypotheses Included in Introduction pag 4
Methods		
Study design	4	Present key elements of study design early in the paper Included in Methods pag 4-5
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection Included in Methods pag 5
Participants	6	*Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up Included in Methods pag 4-5 *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case Not applicable
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable Included in Methods pag 5
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group Included in Methods pag 4-5
Bias	9	Describe any efforts to address potential sources of bias Included in Limitations pag 11-12
Study size	10	Explain how the study size was arrived at Included in Methods pag 5
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why Included in Methods pag 4-5
Statistical methods	12	Describe all statistical methods, including those used to control for confounding Included in Statistical analysis pag 6-7
		Describe any methods used to examine subgroups and interactions Included in Statistical analysis pag 6-7
		Explain how missing data were addressed Included in Statistical analysis pag 6
		Cohort study—If applicable, explain how loss to follow-up was addressed

		Included in Statistical analysis pag 6 and in Limitations pag 12 *Case-control study*—If applicable, explain how matching of cases and controls was addressed *Cross-sectional study*—If applicable, describe analytical methods taking account of sampling strategy
		(*e*) Describe any sensitivity analyses
Results		
Participants	13*	Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed Included in Results pag 7-8
		Give reasons for non-participation at each stage Not applicable
		Consider use of a flow diagram Include in Supplementary material Supplementary Figure 1
Descriptive data	14*	Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders Included in Results pag 7-8
		Indicate number of participants with missing data for each variable of interest Included in Results pag 7-8
		Cohort study—Summarise follow-up time (eg, average and total amount) Included in Results pag 8
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time Included in Results pag 7-8 and Tables 1 and 2
		Case-control study—Report numbers in each exposure category, or summary measures of exposure
		Cross-sectional study—Report numbers of outcome events or summary measures
Main results	16	Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included Included in Results page 7-8 and Tables 1, 2 and 3
		Report category boundaries when continuous variables were categorized Included in Results page 7-8 and Tables 1,2 and 3
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses Included in Statistical analysis pag 7
Discussion		
Key results	18	Summarise key results with reference to study objectives Included in Discussion pag 9-11
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias Included in Limitations pag 11-12
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence Included in Discussion pag 9, 10,11 and Limitations pag 11, 12
Generalisability	21	Discuss the generalisability (external validity) of the study results Included in Discussion pag 9-10
Other information		
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based Included in role of funding pag 7

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.

Supplementary References

1. Singh B, Ryan H, Kredo T, Chaplin M, Fletcher T. Chloroquine or hydroxychloroquine for prevention and treatment of COVID-19. *Cochrane Database Syst Rev* 2021; **2**(2): Cd013587.