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Intravenous methylprednisolone pulses in hospitalised patients with severe COVID-19 pneumonia, A double-blind, randomised, placebo-controlled trial

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Intravenous methylprednisolone pulses in hospitalized patients with severe COVID-19 pneumonia,

A double-blind, randomized, placebo-controlled trial

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Abstract

Rationale. Pulse glucocorticoid therapy is used in hyperinflammation related to coronavirus 2019 (COVID-19). We evaluated the efficacy and safety of pulse intravenous methylprednisolone in addition to standard treatment in COVID-19 pneumonia.

Methods. In this multicenter, randomized, double-blind, placebo-controlled trial, 304 hospitalized patients with Covid-19 pneumonia were randomized to receive 1 g of methylprednisolone intravenously for 3 consecutive days or placebo in addition to standard dexamethasone. The primary outcome was the duration of the patient hospitalization, calculated as the time interval between randomization and hospital discharge without the need of supplementary oxygen. The key secondary outcomes were survival free from invasive ventilation with orotracheal intubation and overall survival.

Results. Overall, 112 of 151 (75.4%) patients in the pulse methylprednisolone arm and 111 of 150 (75.2%) in the placebo arm were discharged from hospital without oxygen within 30 days from randomization. Median time to discharge was similar in both groups [15 days (95% confidence interval (CI), 13.0 to 17.0) and 16 days (95%CI, 13.8 to 18.2); hazard ratio (HR), 0.92; 95% CI 0.71-1.20; p = 0.528]. No significant differences between pulse methylprednisolone and placebo arms were observed in terms of admission to Intensive Care Unit with orotracheal intubation or death (20.0% versus 16.1%; HR, 1.26; 95%CI, 0.74-2.16; p = 0.176), or overall mortality (10.0% versus 12.2%; HR, 0.83; 95%CI, 0.42-1.64; p = 0.584). Serious adverse events occurred with similar frequency in the two groups.

Conclusions. Methylprenisolone pulse therapy added to dexamethasone was not of benefit in patients with COVID-19 pneumonia.

Message of the study: Pulse glucocorticoid therapy is used for severe and/or life threatening immuno-inflammatory diseases. The addition of pulse glucocorticoid therapy to the standard low dose of dexamethasone scheme was not of benefit in patients with COVID-19 pneumonia.

Pharmacologic effective treatments for coronavirus disease 2019 (COVID-19) are needed (1). Glucocorticoids for their broad and rapid anti-inflammatory effects are ideal candidates for the treatment of hyperinflammation in COVID-19 (2-4). Interestingly, Fauci and coworkers in the 70s showed that glucocorticoids block inflammation by inhibiting the efflux of neutrophils and monocytes to the inflammatory sites (5). Both cell types play an important role in COVID-19-induced lesions, both in the peripheral blood and lungs (3, 6). The Randomized Evaluation of COVID-19 Therapy (RECOVERY) trial showed that low-dose dexamethasone reduced mortality in hospitalized patients with COVID-19 requiring respiratory support (7). This observation was confirmed by a prospective meta-analysis of 7 randomized trials in severe COVID-19 patients which showed that the administration of systemic glucocorticoids, compared with usual care or placebo, was associated with lower 28-day all-cause mortality and that it was also safe (8).

Pulse glucocorticoid therapy (> 250 mg of prednisone equivalent per day for 1 or a few days) has the most relevant genomic and nongenomic actions, that are responsible for the anti-inflammatory and rapid effects of glucocorticoids, therefore this treatment modality is used for particularly severe and/or life threatening immuno-inflammatory diseases as initial therapy (9-12). In COVID-19 the cytokine storm represents the acme of the inflammatory process, suggesting the need for a prompt and strong anti-inflammatory effect. Therefore, the addition of pulse glucocorticoid therapy to the standard low dose of dexamethasone scheme can suppresses the hyperinflammatory processes in COVID-19 more effectively than dexamethasone alone and may represent a potential treatment option for patients with severe and critical COVID-19, where pulse glucocorticoid therapy can provide better alternative than non-pulse treatment.

No double-blind randomized trials have been conducted to assess the efficacy of the addition of steroid pulse therapy to the available usual care for COVID-19

pneumonia. To evaluate the efficacy and safety of pulse intravenous methylprednisolone therapy in hospitalized patients with COVID-19 pneumonia, we conducted a multicenter, randomized, double-blind, placebo-controlled trial.

Methods

Study design and participants

This is a multicenter, randomized, double-blind, placebo-controlled trial coordinated by the Azienda Unità Sanitaria Locale-IRCCS of Reggio Emilia, Italy. Nineteen Italian centres participated to this trial.

The trial was submitted and approved on November 20, 2020, by the Italian Medicines Agency (AIFA) (code 130662) and on November 25, 2020, by the COVID-19 Ethics Committee established at the National Institute for Infectious Diseases, Lazzaro Spallanzani, Rome (code 217). The trial was conducted in accordance with the principles of the Good Clinical Practice guidelines of the International Conference on Harmonization. The trial was overseen by an independent data safety and monitoring committee. Written informed consent was obtained from each patient or from the patient's legally authorized representative if the patient was unable to provide consent. Full details of the trial design, conduct, oversight, and analyses can be found in the protocol, statistical analysis plan, and supplementary appendix.

The study population included patients hospitalized for recent-onset COVID-19 pneumonia documented by radiological imaging, with more than five days since initial symptoms of infection, requiring supplemental oxygen in any delivery mode, except invasive mechanical ventilation, with PaO_2/FiO_2 between 100 and 300, and a C-reactive protein (CRP) greater than 5 mg/dL. This CRP value was selected because this cut-off on admission is an indicator of COVID-19 disease severity associated with increased mortality in hospitalized patients (13).

Cases of COVID-19 were confirmed by polymerase chain reaction method with nasopharyngeal swab.

Patients were excluded if they required invasive mechanical ventilation or in the presence of shock or concomitant organ failure requiring an Intensive Care Unit (ICU) admittance. Other exclusion criteria were pregnancy or breastfeeding, severe cardiac or renal failure, diabetes not in good metabolic control based on physician's clinical judgment and other clinical conditions which contraindicate methylprednisolone and which cannot be treated or resolved based on physician's clinical judgment. Therapy with steroid high dose pulses in the week preceding the enrolment in the study and enrolment in another clinical trial were also exclusion criteria.

Randomization

Eligible patients were randomly assigned in 1:1 ratio to receive either methylprednisolone pulse therapy or placebo, in both arms in addition to standard treatment with dexamethasone. Random assignment was performed centrally at the Trials Center of the Policlinico San Martino-IRCCS in Genova, Italy, stratified by participating centers and type of noninvasive respiratory support [standard oxygen versus High Flow Nasal Cannula (HFNC) or noninvasive ventilation (NIV)]. Computer generated random lists were prepared using permuted balanced blocks of size 4 in random sequence. An Internet-based randomization system ensured concealment of the treatment assignment until the patient had been registered in the system. Treatment allocation was communicated electronically only to the study center's pharmacy to allow local preparation of the methylprednisolone or placebo bag for the IV administration of the treatment.

Treatment

Patients in the experimental arm received 3 boluses of 1 g of methylprednisolone intravenously on days 1, 2 and 3 from randomization in 100 ml physiological in addition to standard treatment. Patients in the control arm received 100 ml of saline (placebo) administered on days 1, 2 and 3 from randomization in addition to standard treatment. Standard treatment included dexamethasone (6 mg / day oral or intravenous for 10 days), according to the RECOVERY study protocol (2). The standard therapy was allowed to change during the study in accordance with the indications of the Italian Medicines Agency (AIFA) cards regarding the choice of drugs, dosages, or duration of treatment.

Procedures

Patients were evaluated daily during their hospitalization, from randomization (day 1) through day 30. The type of noninvasive respiratory support was daily recorded. The medical evaluations on day 21, 28 and end of study (30 days) could be limited to a telephone contact if the patient was at home. The compilation of the end of study visit was mandatory and it could coincide with any clinical visit or telephone contact occurred during the 30 days' observation window. All the severe and non-severe adverse events observed during the hospitalization and in the 30 days from randomization were recorded in the clinical database. Adverse events were classified and graded according to the Common Terminology Criteria for Adverse Events (CTCAE) v 5.0 criteria. For each adverse event the maximum grade of toxicity was identified. Trial participants, investigators, study team care providers were unaware of the trial-group assignments until after all data queries were resolved and the database was locked. Only the hospital pharmacies were aware of the treatment allocation.

Outcomes

The primary endpoint was the duration of the patient hospitalization, calculated as the interval of time between the randomization and the hospital discharge without the need of supplementary oxygen during the first 30 days following randomization. The patients who were transferred to other hospital wards at different intensity of care were considered as still hospitalized. Patients who were transferred to other institutions outside the hospital (e.g., COVID-hotels) for the impossibility of transferring the patient at home were considered as discharged from the hospital.
Patients deceased within 30 days were considered as never discharged.
Patients discharged at home with supplementary oxygen were censored at the date of discharge because no further follow-up information was available. This choice implied the questionable assumption that their prognosis was similar to that of patients still in hospital: the effects of this assumption were evaluated in 2 post-hoc sensitivity analyses in which these patients were considered either discharged without oxygen, or never discharged. The results of these analyses closely resembled those of the primary analysis. The secondary endpoints of efficacy were the following:

1. Survival free from invasive ventilation with orotracheal intubation, defined as the interval between the randomization and the first use of invasive ventilation with orotracheal intubation or death.
2. Overall survival, defined as the interval between randomization and death for any cause.

According to the study protocol, the following explorative endpoints were also evaluated in selected populations:

1. Survival free from clinical worsening, defined as the interval between randomization and the first episode in which the ratio PaO_2/FiO_2 drops below the value 150 or death. Only patients with PaO_2/FiO_2, > 200 mm Hg at enrolment were included in this analysis.

2. Survival free from NIV or HFNC, defined as the interval between randomization and the first use of NIV or the first administration of HFNC, or death. Only patients who did not receive oxygen in this modality at randomization were included in this analysis.

Statistical analysis

Assuming a median time to hospital discharge of 14 days in the control arm, the experimental treatment was considered effective if it reduced time to discharge by 37.5% in relative terms (hazard ratio = 1.60). This corresponds to an absolute increase in the cumulative probability of discharge at 14 days from 50% to 67%, i.e., a decrease in the median time to discharge from 14 to less than 9 days. Assuming a statistical power of 90% and a 5% error rate α, for a two-tailed log rank test at least 198 hospital discharges needed to be observed. The sample size required to observe 198 hospital discharges, assuming a 5% overall dropout rate, was estimated to be at least 260 patients, 130 per arm.

Statistical analysis is detailed in the Statistical Analysis Plan. The cumulative probabilities of survival free from discharge in the two treatment groups were estimated according to the Kaplan Meier method and compared with a non-stratified log rank test. The hazard ratio and its 95% confidence interval (95%CI) were computed by fitting a univariate Cox's model. All secondary efficacy analyses were performed with the same approach described for the primary efficacy analysis. Primary and secondary efficacy analyses were performed on all patients included in the intention-to-treat population. Explorative endpoints analyses were carried out on the appropriate sub-populations, identified from the intention-to-treat population.

We reported the P value (2 tailed; significance defined as $P<0.05$) and bilateral 95%CI not adjusted for multiplicity.

We performed some subgroups analyses not planned into the Statistical Analysis Plan that we reported into the Appendix. The subgroups were defined according to the PaO_2/FiO_2 values at randomization (PaO_2/FiO_2, between 100 and 200 mm Hg versus PaO_2/FiO_2 between 201 and 300 mm Hg), the modality of oxygen administration (supplemental standard oxygen versus noninvasive ventilation or high-flow oxygen), CRP values (stratified in > 5 and $\leq$ 10 mg/dL, > 10 and $\leq$ 15mg/dL, and > 15 mg/dL) at randomization, age (≤ 60 years and > 60 years), and Body Mass Index (BMI) (stratified in quartiles: 1st quartile: ≤ 24,6; 2nd quartile: 24,7-27.6; 3rd quartile: 27.7- 30.9; 4th quartile: ≥ 31.0). Hazard ratios estimates obtained in the Cox model were reported within each subgroup. The presence of a significant variation of the Hazard Ratios across strata of each factor was assessed by means of the treatment-by-factor interaction tests.

Statistical analyses were performed using SAS, version 9.4 (SAS Institute) and SPSS, version 23 (IBM Corp). The trial is registered with EudraCT, 2020-004323-16, and ClinicalTrials.gov Identifier, NCT04673162.

RESULTS

Patients

Between December 21, 2020, and March 10, 2021, the 19 centres randomized 304 patients, 152 in the methylprednisolone pulses arm and 152 in the placebo and standard therapy arm. Three patients (1 in the experimental arm and 2 in the control arm) withdrew consent leaving 301 patients eligible for the intention-to-treat analysis (Figure 1).

One hundred and forty-nine patients (98.7%) assigned to receive methylprednisolone pulses and 148 (98.7%) assigned to receive placebo received

the therapy as assigned. Most patients, 144/151 (95.4%) in active treatment arm and 142/150 (94.7%) in the placebo arm, received all 3 pulses. Demographic and baseline clinical characteristics are summarized in Table 1.

Primary Outcome

One hundred and twelve of 151 (75.4%) patients in the pulse methylprednisolone arm and 111 of 150 (75.2%) patients in the placebo arm were discharged from hospital without oxygen within 30 days. No difference in time to discharge without oxygen was observed between the two groups: the median was 15 (95%CI, 13.0 to 17.0) days in methylprednisolone arm versus 16 (95%CI, 13.8 to 18.2) days in placebo arm (hazard ratio, 0.92; 95%CI, 0.71-1.20; P-value= 0.528) (Table 2 and Figure 2a).

Secondary Outcomes

Fifty-four patients were admitted to Intensive Care Unit and received invasive ventilation with orotracheal intubation or died, with no significant differences between pulse methylprednisolone and placebo arms (20.0% versus 16.1%; hazard ratio, 1.26; 95%CI, 0.74-2.16) (Table 2 and Figure 2b).

Thirty-three deaths occurred within 30 days since randomization, and mortality was comparable in the two arms (10.0% versus 12.2%; hazard ratio, 0.83; 95%CI, 0.42-1.64) (Table 2 and Figure 2c).

Explorative Endpoints and Subgroup Analyses

At randomization, 158 patients had PaO_2/FiO_2 ratio > 200 mm Hg. In these patients the incidence of clinical worsening defined as progression to a PaO_2/FiO_2 < 150 mm Hg or death was similar in methylprednisolone pulse and placebo arms (45.7% versus 40.3%; hazard ratio, 1.17; 95%CI, 0.73-1.89) (Table 2).

At randomization, 205 patients received standard supplementary oxygen. In these patients the incidence of progression to the use of HFNC or NIV, mechanical ventilation or death was not different in methylprednisolone pulse and placebo arms (52.9% versus 50.6%; hazard ratio, 1.16; 95%CI, 0.79-1.70).

No differences were observed between the two arms in subgroup analyses, where subgroups were defined according to PaO_2/FiO_2 values, different modalities of oxygen therapy, CRP values, age and BMI at randomization (see Appendix).

Safety Outcomes

Eighty-six adverse events were reported in 51/149 (34.2%) patients treated with placebo and 90 in 54/149 (36.2%) patients treated with pulses of methylprednisolone. Twenty-eight (32.6%) grade 3 or 4 adverse events occurred in placebo group versus 35 (38.9%) in pulse methylprednisolone group (Table 3). Forty-eight adverse events were judged to be treatment related by the principal investigators: 18 (20.9%) and 30 (33.3%) in the placebo and methylprednisolone arm, respectively. Adverse events by MedDRA system organ class (version 22.0) are listed in Appendix.

Sixteen serious adverse events (SAEs) occurred in the placebo arm and 9 in the methylprednisolone pulses. Eight grade 3 or 4 SAEs occurred in the placebo arm and 7 in methylprednisolone pulses. The number of SAEs considered by the investigators as treatment related were similar in the two arms, as well as the frequency of patients with SAEs (Table 3). A summary description of SAEs is reported in the Appendix. Bacterial infections were the most frequently reported severe adverse events in both arms and they were detailed in Table S2. In particular, serious infections thought to be related to the treatment were observed in 3.3% of the patients treated with methylprednisolone pulses and in 4% of those treated with

placebo. Cases of severe uncontrolled hyperglycemia were not reported in the two arms.

Discussion

In this double-blind, randomized, placebo-controlled trial no significant difference was observed in time to hospital discharge between the high-dose methylprednisolone pulse group and the standard of care group in patients with COVID-19 pneumonia. There was also no benefit in the secondary outcomes, survival and admission to intensive care unit. The lack of benefit on the primary outcome detected in the ITT population was also observed in subgroups of patients defined according to different PaO_2/FiO_2 values, different modalities of oxygen therapy, and different CRP values at randomization. Furthermore, we failed to observe beneficial effects of methylprednisolone pulses on progression of respiratory failure in the subgroups of 158 patients with PaO_2/FiO_2 ratio > 200 mm Hg and in the 206 patients who received standard supplementary oxygen at randomization. Therefore, a rapid and vigorous suppression of inflammation with the addition of methylprednisolone pulses did not provide clinical benefit compared to standard care with dexamethasone according to the RECOVERY trial schedule in COVID-19 hospitalized patients requiring oxygen therapy (7).

Pulse glucocorticoid therapy is used in many immuno-inflammatory conditions to obtain a quick and strong suppression of inflammation in emergency situations (9-12, 14). However, despite its diffuse use in clinical practice, very few studies, often uncontrolled and underpowered, have evaluated the efficacy and safety of this therapy (10-12, 14-17). Indeed, a systematic literature review on safety and efficacy of pulse glucocorticoid therapy for SARS-CoV, Middle East Respiratory Syndrome (MERS)-CoV or SARS-CoV-2 showed that the quality of the evidence is poor and randomised controlled trials are highly needed (14). A recent Iranian randomized

trial showed a significant pulmonary improvement and a reduced mortality in hospitalized patients with severe COVID-19 treated with pulse methylprednisolone compared to those treated with standard therapy (18). However, this study has several limitations. It was not double-blind placebo controlled and enrolled only 62 patients. Even more important, differently from our study, standard of care did not include glucocorticoid treatment. A Spanish randomized, open-label, controlled study in hospitalized patients with COVID-19 pneumonia needing oxygen therapy compared low dose dexamethasone (6 mg once daily for 10 days) to high dose dexamethasone (20 mg once daily for 5 days, followed by 10 mg once daily for additional 5 days) (19). Similarly to our study, this trial showed no difference in efficacy between high dose and low dose of dexamethasone. However, the two studies were not completely comparable because we used in pulse therapy a 10 times higher steroid dosage (125 mg of prednisone equivalent per day versus 1250 mg of prednisone equivalent per day), maximizing the corticosteroid genomic and nongenomic actions (9). Although all our patients had severe inflammation at enrollment (CRP values had to be more than 10 times the upper reference), they represented a spectrum of COVID-19 disease, ranging from patients more inflamed and critically ill at the time of randomization to those with less severe inflammatory disease receiving supplemental oxygen by nasal prongs. We only excluded patients in early symptomatic phases (i.e., interval between initial symptoms of infection and randomization $\leq$ 5 days) as we did not want to prevent functional immune response in the early stages of the infection, and patients in invasive mechanical ventilation or with presence of shock or concomitant organ failure requiring an Intensive Care Unit admittance.

Despite the overall negative results of our study, we cannot exclude potential benefit in specific subgroups of patients with more severe disease and more severe inflammatory response. Also, our study cannot rule out a role for pulse

glucocorticoid therapy as a rescue therapy in patients who failed to improve after dexamethasone and that a gradual tapering of pulse methylprednisolone would have better controlled the inflammation, avoiding possible rebound. Of interest, our study did not show an increased risk of glucocorticoid side effects, particularly infections or hyperglycemia, in the patients treated with methylprednisolone pulses. Previous studies observed only an increased frequency of mild acute adverse events (e.g., sleep disturbance, mood change) in patients treated with pulse glucocorticoids compared to those treated with placebo (10,12). Therefore, our results confirm that pulses of methylprednisolone are safe also in the inflammatory phase of patients with COVID-19.

Our trial has some strengths, but also some limitations. One of the strengths is that the trial was multicenter double-blind placebo controlled. Our trial population was intentionally chosen to have a severe inflammatory status as reflected by the elevated CRP values at baseline which were similar in the pulse methylprednisolone and placebo patients. These patients were presumably those with better chance of responding to methylprednisolone pulses. The two groups were balanced in terms of their baseline characteristics, including demographics, days from symptom onset to randomization, PaO_2/FiO_2 value and modality of noninvasive support at randomization, coexisting conditions, and concurrent treatment. The selected primary outcome, duration of patient hospitalization, may have important limitations, particularly regarding differences in local clinical practice. However, in Italy, during the study period, the treatment of hospitalized patients with COVID-19 was standardized by AIFA and the treatments were similar across all trial sites.

The main limitation of this trial derived from its limited sample size, which implies that the observed results, do not allow to rule out the hypothesis that pulses of methylprednisolone are associated with a modest beneficial effect, e.g., a 15-16% reduction in time to discharge. More important, we cannot exclude possible benefits in specific subpopulations of patients that were underpowered to detect clinically

relevant differences. Platform trials enrolling large numbers of patients could better evaluate the efficacy of pulse glucocorticoid therapy in these specific subgroups of patients.

Author contributions: CS, MC, DFM, GLM, LB, PB, CT and SC had access to the raw data; CS, MM, MC, DFM, GLM, PV, SN, GD, LB, LS, PB, CT and NF contributed to the study design, data collection, data review, interpretation, writing and approval of the manuscript, and the decision to submit; MC, DFM, GLM, LB, LS, and SC accessed and verified the data and did the statistical analysis; GG, MC, AMM, CB, AV, FF, GM, RC, FT, AB, EB, CB, GJB, AP, WI, RS, CB, FL, MC, KET, GC, MS, GF, RD, GB, EAN, GC, GF, IB, LZ, TA contributed to collection, verification, and interpretation of the data, and critical review and revision of the manuscript; CS, MM and MC had final responsibility for the decision to submit for publication.

Declaration of interests: We declare no competing interests.

This study is registered with EudraCT, 2020-004323-16, and ClinicalTrials.gov Identifier, NCT04673162.

Data sharing: Deidentified participant data including data dictionaries will be made available within 3 months of publication. The proposed use of the data and analyses must be approved by the Scientific Committee before to have access to the data. The Scientific Committee will have the right to review and comment any draft manuscripts prior to publication. Those wishing to request access should write to the corresponding author: carlo.salvarani@ausl.re.it.

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Figure 1. The CONSORT Flow Diagram of the study

Figure 2. Kaplan-Meier estimates of hospital discharge without oxygen (A) orotracheal intubation or death (B), and death (C).

Table 1. Demographic and Clinical Characteristics of Patients at Randomization.

Characteristics	**Placebo + Standard (N=150)**	**MTP pulses + Standard (N=151)**
Age, median (IQR) - y	64.0 (55.0-72.2)	64.0 (54.0-74.0)
Male sex – no. (%)	106 (70.7)	111 (73.5)
Days from symptom onset to randomisation, median (IQR)	8.0 (6.0-10.0)	9.0 (6.0-11.0)
Days from Pneumonia diagnosis to randomisation, median (IQR)	1.0 (1.0-2-0)	1.0 (1.0-2.0)
PaO_2/FiO_2, median (IQR) – mmHg	204.0 (158.0-243.0)	208.0 (158.0-248.0)
Respiratory rate, median (IQR) – (breaths/min)	20.0 (18.0-24.0)	20.5 (18.0-24.0)
CRP, median (IQR) – mg/dL	10.9 (7.6-14.6)	10.6 (7.3-14.9)
Modality of oxygen administration (%)		
Supplemental standard oxygen	101 (67.3)	104 (68.9)
NIV or high-flow oxygen	49 (32.7)	47 (31.1)
Coexisting conditions – no. (%)		
Diabetes mellitus	19 (12.7)	26 (17.2)
Hypertension	74 (49.3)	83 (55.0)
COPD	8 (5.3)	5 (3.3)
Heart failure	7 (4.7)	9 (6.0)
Obesity (BMI ≥ 30 kg/m^2)	35/112 (31.3)	34/121 (28.1)
Other Treatments – no. (%) *		
Glucocorticoids	135 (90.0)	131 (86.8)
Remdesivir	27 (18.0)	19 (12.6)
Antibiotics	41 (27.3)	40 (26.5)
LMWH	117 (78.0)	120 (79.5)

MTP = methylprednisolone; IQR = interquartile range; CRP = C-reactive protein; NIV = non-invasive ventilation; COPD = Chronic obstructive pulmonary disease; LMWH = Low-molecular-weight heparin

*any treatments administered during the period from the onset of symptoms until randomization.

Table 2. Clinical Outcomes in the Intention-to-Treat Population

Endpoints	Placebo + Standard no. (% at 30 days)	MTP pulses + Standard no. (% at 30 days)	Hazard Ratio (95% CI)	P-value
Primary endpoint				
Discharge without oxygen [a]	111/150 (75.2)	112/151 (75.4)	0.92 (0.71-1.20)	0.528
Time to discharge within 30 days – median (95%CI)	16 (13.8-18.2)	15 (13.0-17.0)		
Secondary endpoints				
Admission to ICU or death [b]	24/150 (16.1)	30/151 (20.0)	1.26 (0.74-2.16)	0.176
Deaths	18/150 (12.2)	15/151 (10.0)	0.83 (0.42-1.64)	0.584
Explorative endpoints				
Clinical worsening or death [c]	31/77 (40.3)	37/81 (45.7)	1.17 (0.73-1.89)	0.430
Use of HF, NIV, or death [d]	51/101 (50.6)	55/104 (52.9)	1.16 (0.79-1.70)	0.430

All proportions were cumulative probabilities estimated with the Kaplan Meier Product Limit Estimator and compared with the log-rank test

a) 1 patient in the Placebo arm and 3 patients in the MTP arm had been admitted to ICU but had a full recovery and were discharged without oxygen within 30 days.

b) 8 patients in the Placebo arm and 7 patients in the MTP arm died after admission to ICU, while 10 patients and 8 patients, respectively, died before being admitted to ICU.

c) in the 158 patients with PaO2/FIO2 ratio > 200 mm Hg at randomization.

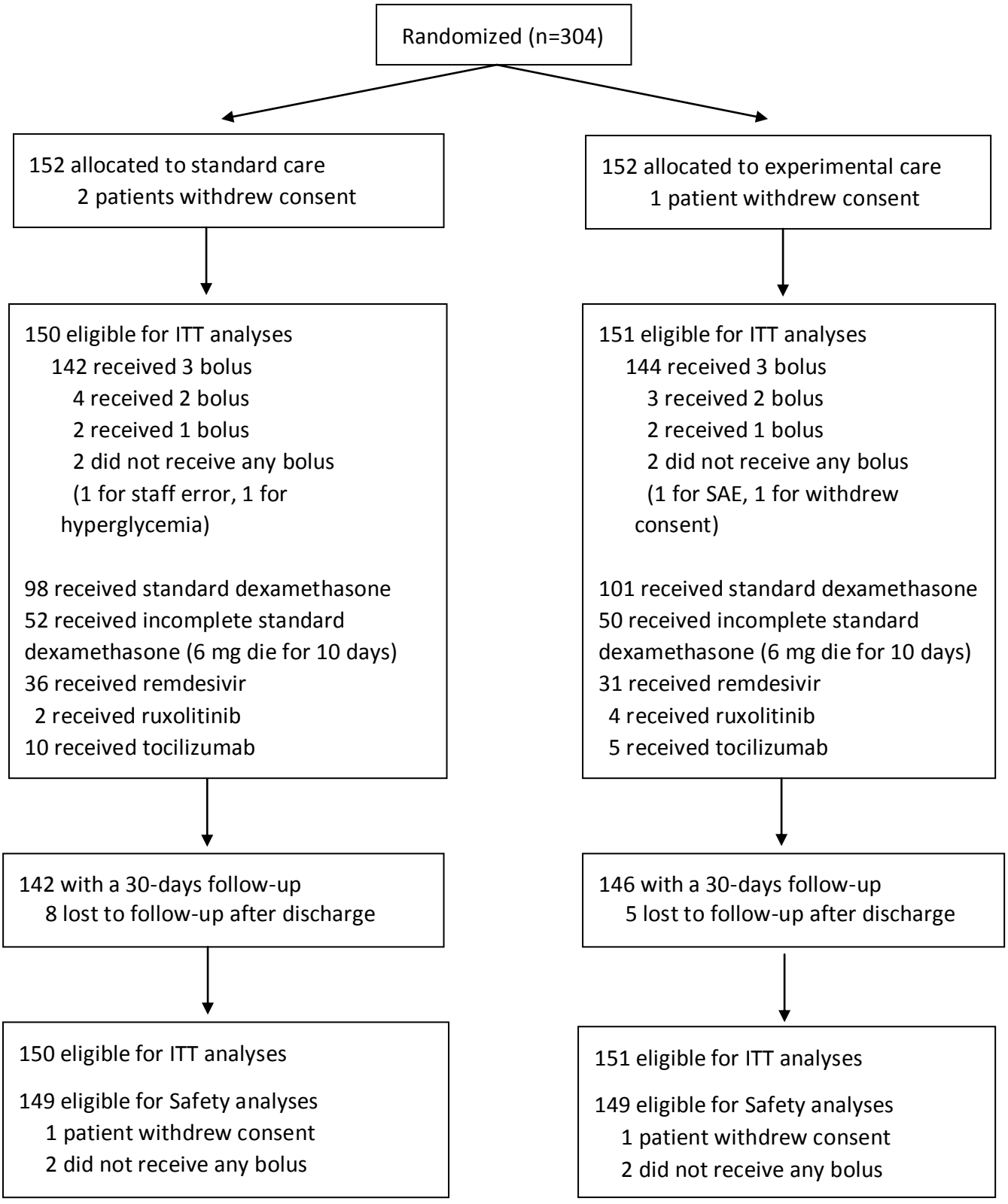
d) in the 205 patients who received standard supplementary oxygen at randomization.

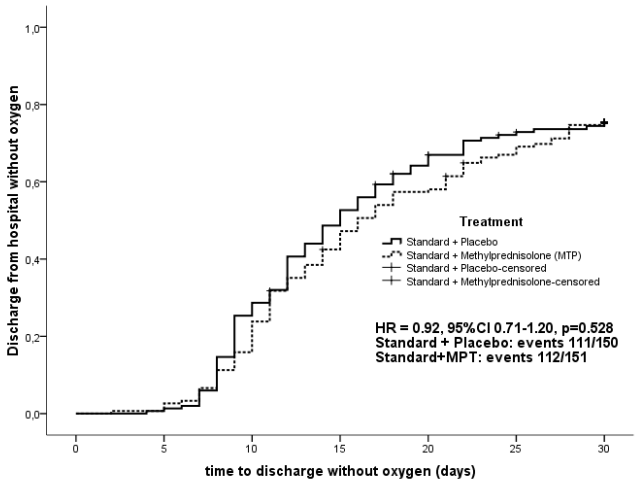
ICU = Intensive Care Unit; HF=High Flow oxygen; NIV = non-invasive ventilation.

Table 3. Adverse Events by Treatment Arm, Grade and Relatedness in the Safety Population of 297 patients.

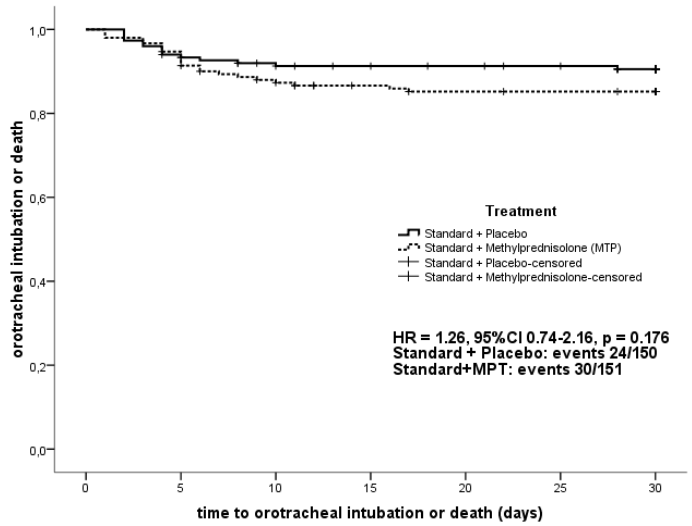
	Treatment Arm	
	Placebo + Standard	**MTP pulses + Standard**
Adverse Events – no. (%)		
Reported AEs	86	90
Grade AEs *		
1-2	51 (59.3)	52 (57.8)
3-4	28 (32.6)	35 (38.9)
5	7 (8.1)	3 (3.3)
Relatedness AEs (Yes)	18 (20.9)	30 (33.3)
Patients with AEs	51/149 (34.2)	54/149 (36.2)
Serious Adverse Events – no. (%)		
Reported SAEs	16	9
Grade SAEs *		
1-2	1 (6.2)	-
3-4	8 (50.0)	7 (77.8)
5	7 (43.8)	2 (22.2)
Relatedness SAEs (Yes)	3 (18.7)	2 (22.2)
Patients with SAEs	12/149 (8.0)	8/149 (5.4)

* Grade AEs: 1 = Mild; 2 = Moderate; 3 = Severe; 4 = Life-Threatening; 5 = Death

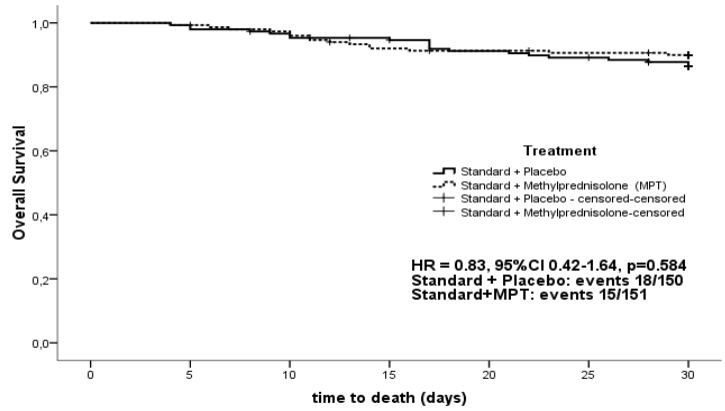

Randomized (n=304)
152 allocated to standard care
2 patients withdrew consent
152 allocated to experimental care
1 patient withdrew consent
150 eligible for ITT analyses
142 received 3 bolus
4 received 2 bolus
2 received 1 bolus
2 did not receive any bolus
(1 for staff error, 1 for hyperglycemia)
98 received standard dexamethasone
52 received incomplete standard dexamethasone (6 mg die for 10 days)
36 received remdesivir
2 received ruxolitinib
10 received tocilizumab
151 eligible for ITT analyses
144 received 3 bolus
3 received 2 bolus
2 received 1 bolus
2 did not receive any bolus
(1 for SAE, 1 for withdrew consent)
101 received standard dexamethasone
50 received incomplete standard dexamethasone (6 mg die for 10 days)
31 received remdesivir
4 received ruxolitinib
5 received tocilizumab
142 with a 30-days follow-up
8 lost to follow-up after discharge
146 with a 30-days follow-up
5 lost to follow-up after discharge
150 eligible for ITT analyses
149 eligible for Safety analyses
1 patient withdrew consent
2 did not receive any bolus
151 eligible for ITT analyses
149 eligible for Safety analyses
1 patient withdrew consent
2 did not receive any bolus



n. at risk							
S + placebo	150	149	112	77	51	37	32
S + MTP	151	150	127	85	63	47	41



n. at risk							
S+placebo	150	140	132	128	126	124	118
S+MTP	151	142	129	121	118	117	116



n. at risk							
S+placebo	150	149	144	140	133	130	124
S+MTP	151	150	146	137	135	133	131

Supplementary appendix

This appendix has been provided by the authors to give readers additional information about their work.

Supplement to: Carlo Salvarani, Marco Massari, Massimo Costantini, et al. Intravenous methylprednisolone pulses in hospitalized patients with severe COVID-19 pneumonia

Table of contents:

1. THE RCT-MP-COVID-19 STUDY GROUP

(The list was ordered by number of patients enrolled)

The following institutions were involved in this study. For each institution the study group members involved with the design and implementation of the study are reported.

Azienda USL -IRCCS di Reggio Emilia:

- Malattie Infettive: Marco Massari, Fabio Sampaolesi, Romina Corsini
- Pneumologia: Nicola Facciolongo, Chiara Barbieri; Francesco Menzella; Matteo Fontana, Silvia Capobelli.
- Reumatologia: Carlo Salvarani, Maria Grazia Catanoso, Gianluigi Bajocchi
- Direzione Scientifica: Massimo Costantini, Franco Merlo, Gabriella Mariani, Luisa Savoldi, Francesca Franzoni, Silvio Cavuto.
- Unità per il coinvolgimento dei pazienti nei processi di ricerca: Chiara Barbieri
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- Alta Intensità di Cure: Emanuele Alberto Negri
- Direzione Sanitaria: Giorgio Mazzi

Regione Emilia Romagna: Anna Maria Marata

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IRCCS Ospedale Policlinico San Martino Genova

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- Farmacia: Sabrina Beltramini, Federica Mina
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- Pneumologia: Cecilia Burattini, Franco Cosimo, Ielpo Antonella
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- Farmacia: Silvia Di Francesco, Roberta Mazzocchi

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Azienda Ospedaliera S. Antonio e Biagio e C. Arrigo di Alessandria

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- Infrastruttura ricerca, formazione e innovazione: Serena Penpa, Antonio Maconi
- Farmacia: Francesca Cammalleri

Ospedale di Bolzano

- Malattie Infettive: Greta Spoladore, Raffaella Binazzi
- Servizio Farmaceutico: Marta Mazzer, Paola Cristina Cappelletto.

2. ADDITIONAL DETAILS ON METHODS

The trial was designed between September and October 2021, by a multidisciplinary group at the Azienda USL-IRCCS of Reggio Emilia. The group included clinical specialists in Infectious Diseases (Marco Massari), Rheumatology (Carlo Salvarani), Respiratory Disease (Nicola Facciolongo), Pharmacology (Caterina Turrà), a group of statisticians (Silvio Cavuto, Luisa Savoldi) and clinical epidemiologists (Domenico Franco Merlo, Paolo Bruzzi and Massimo Costantini).

No funding was obtained for this study. The coordinator centre and all participating centres are using local resources to conduct the trial.

CS, MC, FM, PB, GM drafted the first version of the manuscript. A second revision of the manuscript was performed by PV, SN, GD. PB, LB, MC, FM contributed to the statistical section of the manuscript. CT revised the pharmacological section. The manuscript was revised and approved by all the authors, who agreed the submission for publication.

3. POPULATIONS IN STUDY

3.1. Randomised patients: 304

3.2. Population for Intention To Treat (ITT) analysis: 301
Three patients withdrew consent to the study during the first day after randomisation:

ID-code	Arm	comments
017-123	Standard	withdrew consent 1 day after randomization
016-150	Standard	withdrew consent 1 day after randomization
001-300	Experimental	withdrew consent 1 day after randomization

3.3. Population for safety analyses: 298

ID-code	Arm	comments
003-036	Standard	No experimental treatment
008-101	Experimental	No experimental treatment
016-150	Standard	Withdrew consent
012-206	Standard	No experimental treatment
017-251	Experimental	No experimental treatment
001-300	Experimental	Withdrew consent

Note: 1 patient (code 017-123) withdrew consent after one bolus and was included in the safety analysis until this date.

4. ADDITIONAL DETAILS ON RESULTS

Other data about safety

Table S1. Adverse Events by MedDRA system organ class in the Safety Population (no.=298)

	Adverse Events (no.)							
	Placebo + Standard				**MTP pulses + Standard**			
	Grades *				***Grades ****			
MedDRA system organ class	***1 -2***	***3 -4***	***5***	***Total***	***1 -2***	***3 -4***	***5***	***Total***
Accidents and injuries (SMQ)	1	0	0	1	0	0	0	0
Blood and lymphatic system disorders	0	0	0	0	1	1	0	2
Breast disorders	0	0	0	0	1	0	0	1
Cardiac disorders	2	1	0	3	2	3	0	5
Endocrine disorders	0	0	0	0	1	0	0	1
Gastrointestinal disorders	2	1	0	3	3	1	1	5
Gastrointestinal haemorrhage	0	1	0	1	0	0	0	0
General disorders and administration site conditions	7	1	0	8	2	6	0	8
General system disorders NEC	1	0	0	1	0	0	0	0
Infections and infestations	5	3	4	12	6	5	0	11
Investigations	2	3	0	5	1	0	0	1
Metabolism and nutrition disorders	17	6	0	23	22	8	1	31
Musculoskeletal and connective tissue disorders	3	0	0	3	1	0	0	1
Psychiatric disorders	2	2	0	4	2	2	0	4
Renal and urinary disorders	4	2	0	6	1	3	0	4
Respiratory, thoracic, and mediastinal disorders	2	5	1	8	4	5	0	9
Skin and subcutaneous tissue disorders	1	0	0	1	1	0	0	1
Surgical and medical procedures	0	1	1	2	0	0	1	1
Vascular disorders	2	2	1	5	4	1	0	5
Total	51	28	7	86	52	35	3	90

* Grade AEs: 1 = Mild; 2 = Moderate, 3 = Severe; 4 = Life-Threatening; 5 = Death

Table S2. Summary of Serious Adverse Events (SAEs)

ID - Centre	MedDRA SOC term	MedDRA SOC code	MedDRA PT term	MedDRA PT code	Seriousness	Grade *	Related	Interval onset /outcome (days)	Outcome
Placebo + standard									
271-011	**Gastrointestinal haemorrhage**	10017955	Gastrointestinal disorders	10017947	Important event	4	Yes		Unknown
063-004	**General disorders and administration site conditions**	10018065	Pyrexia	10037660	Important event	2	No	1	Resolved
021-003	**Infections and infestations**	10021881	Septic shock	10040070	Life threatening	5	No	6	Fatal
035-004			Lung abscess	10025028	Hospitalisation	4	Yes		Unknown
049-001			Enterococcal infection	10061124	Important event	3	No	20	Resolved
286-017			Staphylococcal infection	10058080	Death	5	Yes	12	Fatal
286-017			Pneumonia klebsiella	10035717	Death	5	No	12	Fatal
286-017			Candida infection	10074170	Death	5	No	6	Fatal
042-004	**Renal and urinary disorders**	10038359	Renal failure	10038435	Important event	3	No		Unknown
035-004	**Respiratory, thoracic and mediastinal disorders**	10038738	Pneumothorax	10035759	Important event	3	No	3	Resolved
042-004			Pneumothorax	10035759	Hospitalisation	3	No	5	Resolved
071-006			Pneumonia aspiration	10035669	Life threatening	5	No	21	Fatal
130-008	**Surgical and medical procedures**	10042613	Endotracheal intubation	10067450	Life threatening	4	No		Unknown
293-008			Endotracheal intubation	10067450	Death	5	No	9	Fatal
105-013	**Vascular disorders**	10047065	Cerebral haemorrhage	10008111	Life threatening	5	No	1	Fatal
166-001			Cerebral haemorrhage	10008111	Hospitalisation	3	No	16	Resolved
MTP pulses + Standard									
209-013	**Cardiac disorders**	10007541	Cardiac failure	10007554	Important event	3	No		Improved
085-001	**Gastrointestinal disorders**	10017947	Haemorrhagic necrotic pancreatitis	10076058	Life threatening	5	No	7	Fatal
024-001	**Infections and infestations**	10021881	Pseudomonas infection	10061471	Important event	3	No	10	Resolved
245-001			Escherichia sepsis	10015296	Important event	3	No	13	Resolved
250-020			Septic shock	10040070	Important event	3	Yes		Improved
012-003	**Respiratory, thoracic and mediastinal disorders**	10038738	Pneumonia bacterial	10060946	Life threatening	4	No		Improved
250-020			Pneumonia bacterial	10060946	Important event	3	Yes		Improved
278-008	**Surgical and medical procedures**	10042613	Endotracheal intubation	10067450	Life threatening	5	No	9	Fatal
077-005	**Vascular disorders**	10047065	Cerebral ischaemia	10008120	Disability	4	No		Ong./Worsen.

* Grade: 1 = Lightweight; 2 = Moderate, 3 = Severe; 4 = Very Severe; 5 = Death

5. Subgroup analyses

In these subgroup analyses Hazard ratios (HRs) estimates obtained in the Cox model were reported within each subgroup, while the presence of a significant variation of the HRs across strata of each factor was assessed by means of the appropriate treatment-by-factor interaction tests. The P values of the interaction tests are reported, but should be considered with caution, due to the lack of correction for multiplicity.

Table S3: Clinical outcomes at 30 days according to PaO_2/FiO_2 at randomisation

Strata:

- PaO_2/FiO_2, between 100 and 200 (no.=143)
- PaO_2/FiO_2, between 201 and 300 (no.=158)

	Placebo + Standard no. (% at 30 dd)	**MTP pulses + Standard no. (% at 30 dd)**	**Rate Ratio (95% CI)**
PaO_2/FiO_2, between 100 and 200			
Discharge without oxygen	51/73 (70.7)	45/70 (66.7)	0.71 (0.48-1.08)
Admission to ICU or death	17/73 (23.4)	18/70 (25.7)	1.14 (0.59-2.21)
Deaths	12/73 (16.5)	9/70 (12.9)	0.77 (0.32-1.83)
PaO_2/FiO_2, between 201 and 300			
Discharge without oxygen	60/77 (79.5)	67/81 (82.9)	1.16 (0.82-1.65)
Admission to ICU or death	7/77 (9.1)	11/81 (13.7)	1.50 (0.58-3.88)
Deaths	6/77 (8.1)	5/81 (6.3)	0.79 (0.24-2.59)

Tests for interaction for discharge without oxygen (P=0.083), admission to ICU or death (P=0.522), deaths (P=0.758).

Table S4: Clinical outcomes at 30 days according to the modality of oxygen administration at randomisation

Strata:

- Supplemental standard oxygen (no. 205)
- NIV or high-flow oxygen (no.=96)

	Placebo + Standard no. (% at 30 dd)	**MTP pulses + Standard no. (% at 30 dd)**	**Rate Ratio (95% CI)**
Standard oxygen			
Discharge without oxygen	79/101 (79.6)	84/104 (81.1)	0.93 (0.68-1.26)
Admission to ICU or death	12/101 (12.0)	14/104 (13.5)	1.15 (0.53-2.48)
Deaths	9/101 (9.1)	7/104 (6.9)	0.75 (0.28-2.01)
NIV or high-flow oxygen			
Discharge without oxygen	32/49 (66.3)	28/47 (62.0)	0.86 (0.52-1.43)
Admission to ICU or death	12/49 (24.6)	15/47 (31.9)	1.33 (0.62-2.85)
Deaths	9/49 (18.6)	7/47 (14.9)	0.82 (0.31-2.20)

Tests for interaction for discharge without oxygen (P=0.822), admission to ICU or death (P=0.885), deaths (P=0.932).

Table S5: Clinical outcomes at 30 days according to C-reactive protein (CPR) at randomisation

Strata:

- CPR greater than 5 and less or equal 10 (no. 137)
- CPR greater than 10 and less or equal 15 (no. 93)
- CPR greater than 15 (no.=71)

	Placebo + Standard no. (% at 30 dd)	**MTP pulses + Standard no. (% at 30 dd)**	**Rate Ratio (95% CI)**
CPR > 5 and ≤ 10			
Discharge without oxygen	55/68 (82.2)	52/69 (75.4)	0.73 (0.50-1.07)
Admission to ICU or death	6/68 (8.9)	13/69 (19.0)	2.29 (0.87-6.03)
Deaths	5/68 (7.5)	8/69 (11.8)	1.61 (0.53-4.93)
CPR > 10 and ≤ 15			
Discharge without oxygen	31/46 (67.8)	34/47 (75.0)	1.14 (0.70-1.86)
Admission to ICU or death	11/46 (23.9)	8/47 (17.1)	0.68 (0.27-1.70)
Deaths	8/46 (17.4)	1/47 (2.3)	0.12 (0.01-0.93)
CPR > 15			
Discharge without oxygen	25/36 (71.2)	26/35 (75.9)	1.07 (0.62-1.85)
Admission to ICU or death	7/36 (19.8)	8/35 (22.9)	1.22 (0.44-3.36)
Deaths	5/36 (14.5)	5/35 (14.3)	1.04 (0.30-3.60)

Tests for interaction for discharge without oxygen (P=0.276), admission to ICU or death (P=0.145), deaths (P=0.021).

Table S6: Clinical outcomes at 30 days according to the age at randomisation

Strata:

- Age (years) ≤ 60 (no. 120)
- Age (years) > 60 (no. 181)

	Placebo + Standard no. (% at 30 dd)	**MTP pulses + Standard no. (% at 30 dd)**	**Rate Ratio (95% CI)**
Age (years) ≤ 60			
Discharge without oxygen	57/59 (96.6)	57/61 (93.4)	0.78 (0.54-1.13)
Admission to ICU or death	2/59 (3.4)	3/61 (4.9)	1.46 (0.24-8.77)
Deaths	2/59 (3.4)	0/61 (-)	0.19 (0.01-4.28)
Age (years) > 60			
Discharge without oxygen	54/91 (59.3)	55/90 (61.1)	0.97 (0.66-1.41)
Admission to ICU or death	22/91 (24.2)	27/90 (30.0)	1.27 (0.73-2.24)
Deaths	16/91 (17.6)	15/90 (16.7)	0.97 (0.47-1.97)

Tests for interaction for discharge without oxygen (P=0.435), admission to ICU or death (P=0.884), deaths (P=0.054).

Table S7: Clinical outcomes at 30 days according to Body Mass Index at randomisation

Strata:

- BMI 1st quartile, ≤ 24,6 (no.=58)
- BMI 2nd quartile, 24,7-27.6 (no.=59)
- BMI 3rd quartile, 27.7- 30.9 (no.=58)
- BMI 4th quartile, ≥ 31.0 (no.=58)

	Placebo + Standard no. (% at 30 dd)	**MTP pulses + Standard no. (% at 30 dd)**	**Rate Ratio (95% CI)**
BMI 1st quartile			
Discharge without oxygen	17/31 (54.8)	19/27 (70.4)	1.36 (0.71-2.62)
Admission to ICU or death	8/31 (25.8)	7/27 (25.9)	0.98 (0.36-2.71)
Deaths	7/31 (22.6)	2/27 (7.4)	0.37 (0.08-1.72)
BMI 2nd quartile			
Discharge without oxygen	15/23 (65.2)	26/36 (72.2)	1.01 (0.53-1.90)
Admission to ICU or death	6/23 (26.1)	7/36 (19.4)	0.74 (0.25-2.22)
Deaths	5/23 (21.7)	4/36 (11.1)	0.50 (0.13-1.91)
BMI 3rd quartile			
Discharge without oxygen	22/29 (75.9)	22/29 (75.9)	0.88 (0.49-1.59)
Admission to ICU or death	5/29 (17.2)	4/29 (13.8)	0.78 (0.21-2.91)
Deaths	3/29 (10.3)	2/29 (6.9)	0.68 (0.12-3.94)
BMI 4th quartile			
Discharge without oxygen	25/29 (86.2)	22/29 (75.9)	0.76 (0.43-1.35)
Admission to ICU or death	2/29 (6.9)	5/29 (17.2)	2.68 (0.52-13.8)
Deaths	0/29 (-)	2/29 (6.9)	5.19 (0.20-136)

Tests for interaction for discharge without oxygen (P=0.6071), admission to ICU or death (P=0.581), deaths (P=0.046).