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# Anakinra combined with methylprednisolone in patients with severe COVID-19 pneumonia and hyperinflammation: An observational cohort study



Giorgio Bozzi, MD,<sup>a,\*</sup> Davide Mangioni, MD,<sup>a,b,\*</sup> Francesca Minoia, MD,<sup>c</sup> Stefano Aliberti, MD,<sup>d,e</sup> Giacomo Grasselli, MD,<sup>e,f</sup> Laura Barbetta, MD,<sup>g</sup> Valeria Castelli, MD,<sup>a</sup> Emanuele Palomba, MD,<sup>a</sup> Laura Alagna, MD,<sup>a</sup> Andrea Lombardi, MD,<sup>a</sup> Riccardo Ungaro, MD,<sup>a</sup> Carlo Agostoni, MD,<sup>c,h</sup> Marina Baldini, MD,<sup>i</sup> Francesco Blasi, MD,<sup>d,e</sup> Matteo Cesari, MD, PhD,<sup>h,j</sup> Giorgio Costantino, MD,<sup>f,h</sup> Anna Ludovica Fracanzani, MD,<sup>e,k</sup> Nicola Montano, MD,<sup>e,l</sup> Valter Monzani, MD,<sup>g</sup> Antonio Pesenti, MD,<sup>e,f</sup> Flora Peyvandi, MD,<sup>e,m</sup> Marcello Sottocorno, PharmD,<sup>n</sup> Antonio Muscatello, MD,<sup>a</sup> Giovanni Filocamo, MD,<sup>c</sup> Andrea Gori, MD,<sup>a,e,o</sup> and Alessandra Bandera, MD, PhD<sup>a,e,o</sup>  
Milan, Italy

**Background:** Immunomodulants have been proposed to mitigate severe acute respiratory syndrome coronavirus 2-induced cytokine storm, which drives acute respiratory distress syndrome in coronavirus disease 2019 (COVID-19). **Objective:** We sought to determine efficacy and safety of the association of IL-1 receptor antagonist anakinra plus methylprednisolone in severe COVID-19 pneumonia with hyperinflammation.

**Methods:** A secondary analysis of prospective observational cohort studies was carried out at an Italian tertiary health care facility. COVID-19 patients consecutively hospitalized (February 25, 2020, to March 30, 2020) with hyperinflammation

(ferritin  $\geq 1000$  ng/mL and/or C-reactive protein  $\geq 10$  mg/dL) and respiratory failure (oxygen therapy from 0.4 FiO<sub>2</sub> Venturi mask to invasive mechanical ventilation) were evaluated to investigate the effect of high-dose anakinra plus methylprednisolone on survival. Patients were followed from study inclusion to day 28 or death. Crude and adjusted (sex, age, baseline PaO<sub>2</sub>:FiO<sub>2</sub> ratio, Charlson index, baseline mechanical ventilation, hospitalization to inclusion lapse) risks were calculated (Cox proportional regression model). **Results:** A total of 120 COVID-19 patients with hyperinflammation (median age, 62 years; 80.0% males; median PaO<sub>2</sub>:FiO<sub>2</sub> ratio, 151; 32.5% on mechanical ventilation) were evaluated. Of these, 65 were treated with anakinra and methylprednisolone and 55 were untreated historical controls. At 28 days, mortality was 13.9% in treated patients and 35.6% in controls (Kaplan-Meier plots,  $P = .005$ ). Unadjusted and adjusted risk of death was significantly lower for treated patients compared with controls (hazard ratio, 0.33, 95% CI, 0.15-0.74,  $P = .007$ , and HR, 0.18, 95% CI, 0.07-0.50,  $P = .001$ , respectively). No significant differences in bloodstream infections or laboratory alterations were registered.

**Conclusions:** Treatment with anakinra plus methylprednisolone may be a valid therapeutic option in COVID-19 patients with hyperinflammation and respiratory failure, also on mechanical ventilation. Randomized controlled trials including the use of either agent alone are needed to confirm these results. (J Allergy Clin Immunol 2021;147:561-6.)

**Key words:** SARS-CoV-2, COVID-19, hyperinflammation, anti-IL-1, anakinra, corticosteroids, methylprednisolone, immunomodulation, respiratory failure, mechanical ventilation

From <sup>a</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Infectious Disease Unit, <sup>b</sup>the Department of Medical Biotechnology and Translational Medicine, University of Milan, <sup>c</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, UOC Pediatria a Media Intensità di Cure, <sup>d</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Respiratory Unit and Cystic Fibrosis Adult Center, <sup>e</sup>the Department of Pathophysiology and Transplantation, University of Milan, <sup>f</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Department of Anesthesia, Critical Care and Emergency, <sup>g</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Acute Medical Unit, <sup>h</sup>the Department of Clinical Sciences and Community Health, University of Milan, <sup>i</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Internal Medicine Unit, <sup>j</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Geriatric Unit, <sup>k</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Internal Medicine and Metabolic Diseases Unit, <sup>l</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Internal Medicine Unit, Immunology and Allergology Unit, <sup>m</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Internal Medicine and Hemostasis and Thrombosis Unit, <sup>n</sup>Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Hospital Pharmacy, and <sup>o</sup>the Centre for Multi-disciplinary Research in Health Science, University of Milan, Milan.

\*These authors contributed equally to this work and should both be considered first coauthors.

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Corresponding author: Davide Mangioni, MD, Department of Internal Medicine, Infectious Diseases Unit, IRCCS Ca' Granda Ospedale Maggiore Policlinico Foundation, Via Francesco Sforza 35, 20122 Milan, Italy. E-mail: [davide.mangioni@unimi.it](mailto:davide.mangioni@unimi.it).

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## INTRODUCTION

As of November 2020, the ongoing pandemic of coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) affected 46 million people worldwide, resulting in more than 1.2 million deaths.<sup>1</sup>

High levels of proinflammatory cytokines, C-reactive protein (CRP), and ferritin correlate with worse outcomes in patients with severe COVID-19.<sup>2-4</sup> Growing evidence suggests that these

**Abbreviations used**

CCI:	Charlson comorbidity index
COVID-19:	Coronavirus disease 2019
CRP:	C-reactive protein
MPD:	Methylprednisolone
MV:	Mechanical ventilation
SARS-CoV-2:	Severe acute respiratory syndrome coronavirus 2
SOC:	Standard of care

patients develop a hyperinflammatory syndrome resembling cytokine storm syndromes, potentially benefiting from immunomodulatory treatment.<sup>5</sup>

IL-1-receptor antagonist anakinra is one of the cytokine-blocking agents used for COVID-19 treatment.<sup>5</sup> Although randomized clinical trials are ongoing,<sup>6</sup> single-center experiences have reported encouraging findings.<sup>7-10</sup> The short half-life of anakinra enables to rapidly discontinue its action in case of adverse reactions or secondary infections, making its use suitable for critically ill patients also.<sup>7,10</sup> IL-1 inhibition is also associated with reduction in endothelial dysfunction and microvascular alteration,<sup>11</sup> which seem crucial in COVID-19-related thromboembolic events.<sup>12</sup>

Corticosteroid treatment is a cornerstone in the management of noninfectious hyperinflammatory conditions, namely cytokine storm syndromes.<sup>13</sup> Favorable data have recently emerged in support of the use of corticosteroids in patients with severe COVID-19, especially in those receiving invasive MV.<sup>14-17</sup> In a recent meta-analysis of prospective, randomized clinical trials on critically ill patients with COVID-19, use of corticosteroids compared with placebo or standard of care (SOC) resulted in a significantly lower 28-day mortality.<sup>18</sup>

With this study, we aimed at investigating the efficacy and safety of combined treatment with anakinra and methylprednisolone (anti-IL-1 + MPD) in COVID-19 patients with hyperinflammation and respiratory failure.

## RESULTS AND DISCUSSION

Of 476 COVID-19 patients admitted at our hospital between February 25 and March 30, 2020, a total of 120 (25.2%) patients with hyperinflammation and respiratory failure were included according to inclusion/exclusion criteria (see this article's [Methods](#) section in the Online Repository at [www.jacionline.org](http://www.jacionline.org)). Of these, 65 were treated with anti-IL-1 + MPD and 55 were untreated historical controls.

Median age of the study population was 62 years (interquartile range, 54.5-70 years), 80.0% (96 of 120) were males, and median Charlson comorbidity index (CCI) was 0 (interquartile range, 0-1). At inclusion, median PaO<sub>2</sub>:FiO<sub>2</sub> ratio was 151 (105-204.5), 32.5% (39 of 120) were on mechanical ventilation (MV), median ferritin was 1555 µg/L (1239-2679 µg/L), and median CRP was 15.2 mg/dL (10.8-23.1 mg/dL). Compared with historical controls, patients treated with anti-IL-1 + MPD had less frequently CCI less than or equal to 1 (25% vs 45.4%;  $P = .017$ ), longer duration of hospitalization before inclusion (3 vs 1 median days;  $P < .0001$ ), lower baseline PaO<sub>2</sub>:FiO<sub>2</sub> ratio (median of 142 vs 173;  $P = .049$ ), reduced proportion of lopinavir/ritonavir treatment (30.8% vs 70.9%;  $P < .0001$ ), and higher proportion of anticoagulant therapy (63.1% vs 38.9%;  $P = .009$ ). The 2

groups did not differ by age, sex, number of patients on MV at inclusion, baseline ferritin, CRP, lymphocyte and platelet counts, hemoglobin and liver enzyme levels, and use of remdesivir and hydroxychloroquine during hospitalization ([Table 1](#)).

Within the 28-day follow-up, 28 of 120 (23%) patients died, 9 of 65 (13.9%) in the anti-IL-1 + MPD group compared with 19 of 55 (35.6%) controls (Kaplan-Meier curves,  $P = .004$ ; [Fig 1, A](#)). Among patients without MV, mortality rate was 6 of 47 (12.8%) in the anti-IL-1 + MPD group compared with 10 of 34 (29.4%) in controls ( $P = .04$ ; [Fig 1, B](#)). Among those with MV, it was 3 of 18 (16.7%) in the anti-IL-1 + MPD group and 9 of 21 (42.8%) in controls ( $P = .076$ ; [Fig 1, C](#)). Overall cumulative risk of death at 28 days was significantly lower for the anti-IL-1 + MDP group compared with controls (hazard ratio, 0.33; 95% CI, 0.15-0.74;  $P = .007$ ). Other factors significantly associated with survival were age less than 65 years, baseline PaO<sub>2</sub>:FiO<sub>2</sub> ratio more than 100, and CCI 0 compared with 1 or more. No association to survival was found for antiviral treatment or for anticoagulant therapy (see [Tables E1](#) and [E2](#) in this article's Online Repository at [www.jacionline.org](http://www.jacionline.org)). At multivariable analysis, treatment with anti-IL-1 + MPD was found to be independently associated with survival when adjusted by sex, age, baseline PaO<sub>2</sub>:FiO<sub>2</sub> ratio, CCI, MV at inclusion, and days between hospitalization and inclusion (hazard ratio, 0.18; 95% CI, 0.07-0.50;  $P = .001$ ) (see [Table E3](#) in this article's Online Repository at [www.jacionline.org](http://www.jacionline.org)).

Treated patients experienced consistent improvements in respiratory function and a rapid lowering of serum CRP levels during treatment ([Fig 2](#)).

Overall, anti-IL-1 + MPD treatment was well tolerated. Grade 3 or greater gamma-glutamyl transferase increase (27.7%), anemia (24.6%), alanine transaminase increase (6.2%), and granulocytopenia (1.5%) were observed in treated patients. However, a comparable proportion of these adverse events was observed within controls. No differences in adverse events were reported between intravenous and subcutaneous routes of administration. Nine bloodstream infections (13.8%) were observed in the anti-IL-1 + MDP group and 4 (7.3%) in controls ( $P = .23$ ).

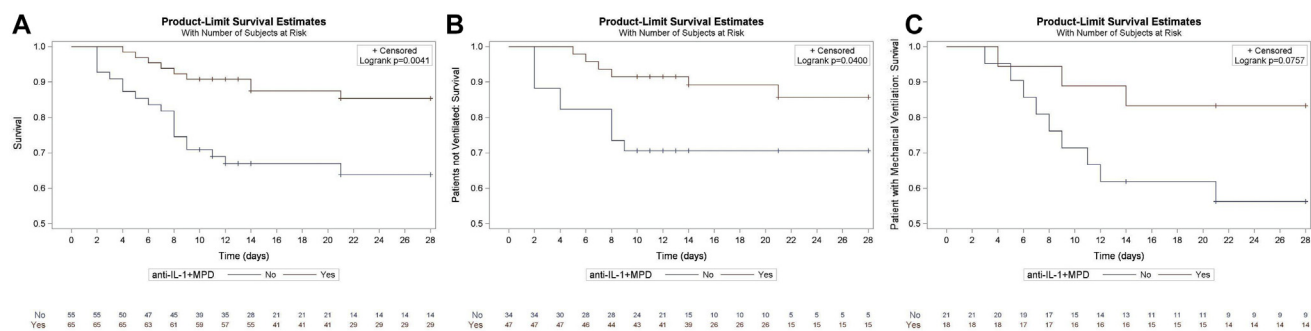
To our knowledge, this is the largest observational study evaluating the efficacy of anakinra associated with MPD in COVID-19 patients with hyperinflammation and respiratory failure.

Several clinical trials are currently in progress to evaluate the benefits of anakinra treatment in COVID-19.<sup>6</sup> In a retrospective study of COVID-19 patients with respiratory failure outside the intensive care unit, Cavalli et al<sup>9</sup> found a survival benefit in high-dose anakinra (5 mg/kg twice a day intravenously) use compared with SOC (90% vs 56% at day 21). A significant reduction in a composite outcome of mortality and/or intensive care unit admission was also observed in a French cohort treated with subcutaneous anakinra (100 mg twice a day for 72 hours, then 100 mg daily for 7 days) compared with historical controls (25% vs 73% at day 20).<sup>10</sup> In contrast to these studies, our analysis encompassed almost one-third of patients (32.5%) who were on MV at inclusion. Moreover, combined treatment with high-dose anakinra and MPD was chosen on the basis of widely approved treatment regimens used in severe cytokine storm syndromes.<sup>13</sup> Of note, corticosteroids such as dexamethasone<sup>14,15</sup> and MPD<sup>16,17</sup> have recently been shown to be beneficial in COVID-19 patients with respiratory failure. In the Randomised Evaluation of COVID-19 Therapy (RECOVERY) trial, the addition of short-

**TABLE I.** Summarization of the study population characteristics according to treatment with anakinra and MPD

Characteristic	N	Treated	N	Not treated	P value
<b>Demographic</b>					
Age (y)	65	60 (54-69)	55	63 (55-76)	.339
Sex: male	65	52 (80)	55	44 (80)	1.000
CCI	65	0 (0-0)	55	0 (0-1)	<b>.037</b>
CCI ≥1	64	16 (25)	55	25 (45.4)	<b>.017</b>
Days between hospitalization and inclusion	65	3 (1-6)	55	1 (0-2)	<b>&lt;.0001</b>
<b>Respiratory function at inclusion</b>					
PaO <sub>2</sub> :FiO <sub>2</sub> ratio					
<100	62	19 (30.7)	50	7 (14.0)	<b>.049</b>
100-200	62	32 (51.6)	50	24 (48.0)	
200-300	62	9 (14.5)	50	14 (28.0)	
300-400	62	2 (3.2)	50	5 (10.0)	
MV	65	18 (27.7)	55	21 (37.5)	.222
<b>Laboratory markers at inclusion</b>					
Ferritin (ng/mL)					
<2000	63	35 (56.45)	38	27 (71.0)	.144
>2000	63	27 (43.55)	38	11 (29.0)	
Lymphocyte count (10 <sup>3</sup> /L)	63	0.7 (0.5-0.9)	55	0.8 (0.5-1.1)	.458
CRP (mg/dL)	65	14.8 (9.0-24.5)	51	15.6 (11.5-21.9)	.969
Hemoglobin (g/dL)	65	12.9 (10.6-14.1)	55	12.5 (10.9-14.1)	.912
Platelet count (10 <sup>3</sup> /L)	65	244 (177-326)	55	230 (189-304)	.436
Alanine transaminase (U/L)	62	41 (28-56)	51	38.0 (25.0-73.0)	.899
Gamma-glutamyl transferase (U/L)	41	59.0 (34.4-110.8)	41	53.0 (26.6-95.0)	.792
d-dimer (μg/L)	56	1220 (855-2906)	47	1271 (1059-1854)	.944
<b>Concomitant medications</b>					
Remdesivir	65	8 (12.3)	55	11 (20.0)	.250
Hydroxychloroquine	65	65 (100)	55	52 (94.6)	.057
Lopinavir/ritonavir	65	20 (30.8)	55	39 (70.9)	<b>&lt;.0001</b>
Anticoagulant therapy	65	41 (63.1)	54	21 (38.9)	<b>.009</b>

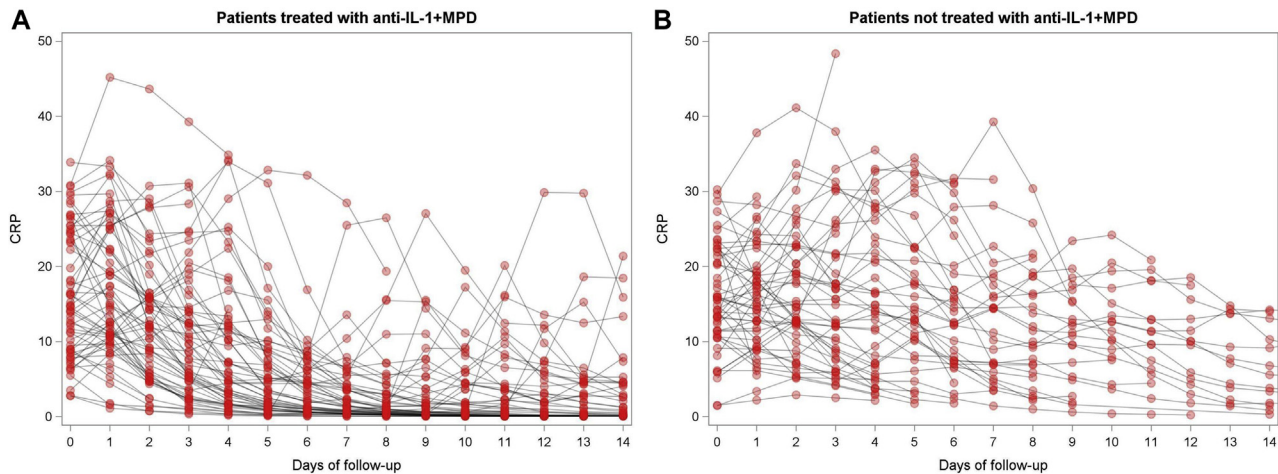
Continuous variables are presented as median (interquartile range), and categorical variables are reported as absolute number (percentage). P values < .05 are indicated in boldface.



**FIG 1.** Survival according to treatment with anakinra and MPD (anti-IL-1 + MPD). Both treated patients and controls were characterized by hyperinflammation and respiratory failure and fulfilled inclusion/exclusion criteria (see this article's [Methods](#) section in the Online Repository). **A**, Survival of all individuals exposed to combined treatment is shown in the red color, dotted line; survival of the control group is shown in the blue color, continuous line. **B** and **C**, Survival of individuals exposed to combined treatment compared with controls in patients without and with MV at inclusion, respectively.

course dexamethasone (6 mg every 24 hours for 10 days or less) to SOC resulted in lower 28-day mortality compared with SOC alone among hospitalized COVID-19 patients (22.9% vs 25.7%, respectively).<sup>14</sup> Interestingly, the highest beneficial effect was obtained in patients on invasive MV (29.3% mortality in the dexamethasone group compared with 41.4% mortality in the SOC group at day 28), whereas no difference was seen among those receiving no respiratory support. No treatment with anakinra was reported in any of the study arms. Conversely, in the COVID-19 Dexamethasone (CoDEX) trial, the addition of intravenous dexamethasone (20 mg every 24 hours for 5 days,

followed by 10 mg every 24 hours for an additional 5 days) to SOC compared with SOC alone in mechanically ventilated COVID-19 patients with moderate to severe acute respiratory distress syndrome (ARDS) resulted in a significant benefit in the number of ventilator-free days (6.6 vs 4.0 days) but not in all-cause 28-day mortality (56.3% vs 61.5%, respectively).<sup>15</sup> In their multicenter quasi-experimental study, Fadel et al<sup>16</sup> compared mortality and/or intensive care unit admission of patients with moderate to severe COVID-19 either on early, short-course MPD (0.5-1 mg/kg/d for 3 days) or SOC.<sup>16</sup> The composite end point occurred at a lower rate in the MPD group (34.9% vs



**FIG 2.** Daily changes in serum CRP from inclusion to day 14 (overall duration of the treatment with anakinra and MPD) for treated (A) and untreated (B) patients.

54.3% at day 14). Again, no patient was treated with anakinra. Ramiro et al<sup>17</sup> prospectively investigated the effect of high-dose intravenous MPD (250 mg on day 1 followed by 80 mg on days 2-5) on the outcome of patients with severe COVID-19-associated cytokine storm syndromes and respiratory failure. In 43% of cases, anti-IL-6 tocilizumab was added as escalation of immunosuppressive treatment, whereas no patients received anakinra. Compared with matched historical controls, hospital mortality was 65% lower and the need of MV was 71% lower in the treatment group. Table II summarizes the major clinical studies that have used either anakinra alone or steroids alone for the treatment of severe COVID-19 so far.

In our study, patients treated with the combination of anakinra plus MPD experienced lower mortality than controls (13.9% vs 35.6% at day 28;  $P = .004$ ). Notably, mortality in treated patients who were on MV at baseline was as low as 16.7%, yet only a trend toward significance emerged compared with the SOC group, possibly due to limited sample size. The outcomes of this population can be compared with the results of MV patients in the RECOVERY trial (no comparison can be made for non-MV patients due to different disease severity between studies).<sup>14</sup> Although the 28-day mortality is similar between MV patients in control groups (42.8% vs 41.4%), our cohort of patients treated with anti-IL-1 + MPD seemed to have experienced a better outcome than patients in the dexamethasone arm of the RECOVERY trial (16.7% vs 29.3% mortality at day 28, respectively). The use of anakinra as add-on therapy to corticosteroids may provide meaningful clinical benefits in this setting and warrants further consideration. The impact of combined treatment was confirmed after adjusting by age, comorbidities, respiratory dysfunction, and length of hospitalization before inclusion, with a 18% reduction in mortality. Combined treatment was overall well tolerated, with no significant differences in adverse event compared with controls. Frequencies of bloodstream infections and laboratory alterations of patients treated with anakinra plus MPD were similar to those reported in studies investigating anakinra as a single agent.<sup>8-10</sup>

Our work has limitations. First, the monocentric nature of the study might affect the generalizability of our results. Second, although controls have been recruited in the same setting, their

number is lower than the cases, mainly because the association of anti-IL-1 + MPD has been implemented relatively early during the pandemic. Third, because no groups treated either with anakinra alone or MPD alone have been included in the analysis, no definitive conclusions could be drawn on the single or synergistic effect of the 2 drugs. Moreover, SOC consisted of evolving combinations of antivirals and anticoagulant therapy, which, although not significantly associated with survival, represent a potential bias. Lastly, no primary hard end point other than 28-day mortality was considered: intermediate end points may help better evaluating treatment efficacy in patients with different severity and length of disease.

In conclusion, combined treatment with anakinra and MPD may be a valid therapeutic option in COVID-19 patients with hyperinflammation and respiratory failure, and also in mechanically ventilated patients. Randomized controlled trials that include arms for steroids and anti-IL-1 therapy alone are needed to confirm these results.

For detailed methods, please see the Methods section in this article's Online Repository at [www.jacionline.org](http://www.jacionline.org).

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"COVID-19 NETWORK" Working Group: Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico. Scientific Direction: Silvano Bosari, Luigia Scudeller, Giuliana Fusetti, Laura Rusconi, and Silvia Dell'Orto; Department of Transfusion Medicine and Biobank: Daniele Prati, Luca Valenti, Giuseppe Lamorte, Maria Manunta, Guido Baselli, and Luigi Santoro; Infectious Diseases Unit: Andrea Gori, Alessandra Bandera, Antonio Muscatello, Davide Mangioni, Laura Alagna, Giorgio Bozzi, Andrea Lombardi, Riccardo Ungaro, Teresa Itri, Valentina Ferroni, Valeria Pastore, Roberta Massafra, and Ilaria Rondolini; Internal Medicine, Hemophilia and Thrombosis Center and Fondazione Luigi Villa: Flora Peyvandi, Roberta Gualtierotti, Barbara Ferrari, Raffaella Rosso, Elisabetta Corona, Nicolò Rampi, and Costanza Massimo; Internal Medicine, Immunology and

**TABLE II.** Summarization of major clinical studies that have used either anakinra alone or steroids alone for the treatment of severe COVID-19

Reference	Investigated drug	Study design	Study population	Treatment/intervention	Outcomes
Cavalli et al, <sup>9</sup> Lancet Rheumatol 2020	Anakinra	Monocentric retrospective case-control study (Italy)	<ul style="list-style-type: none"> <li>● Hyperinflammation (CRP ≥100 mg/L and/or ferritin ≥900 ng/mL)</li> <li>● Bilateral pneumonia</li> <li>● PaO<sub>2</sub>:FiO<sub>2</sub> ≤200 mm Hg on noninvasive ventilation</li> <li>● No mechanically ventilated patients</li> </ul>	IV anakinra 5 mg/kg twice a day (no. 29) vs SOT (no. 16, historical controls)	<ul style="list-style-type: none"> <li>● 21-d survival: 90% in the anakinra group vs 56% the in SOT group (<i>P</i> = .009)</li> <li>● MV-free survival: 72% in the anakinra group vs 50% in the SOT group (<i>P</i> = .15)</li> </ul>
Huet et al, <sup>10</sup> Lancet Rheumatol 2020	Anakinra	Monocentric case-control study (prospective cohort with historical controls) (France)	<ul style="list-style-type: none"> <li>● Bilateral pneumonia</li> <li>● Oxygen saturation of ≤93% under oxygen 6 L/min or more, or saturation ≤93% under oxygen 3 L/min with a loss of 3% in 24 h</li> <li>● No mechanically ventilated patients</li> </ul>	SC anakinra 100 mg twice daily for 72 h followed by 100 mg daily for 7 d (no. 52) vs SOT (no. 44, historical controls)	<ul style="list-style-type: none"> <li>● Need for invasive MV or death: 25% in the anakinra group vs 73% in the SOT group (95% CI, 0.10-0.49; <i>P</i> = .00021)</li> </ul>
Cauchois et al, PNAS 2020	Anakinra	Multicenter retrospective case-control study (France)	<ul style="list-style-type: none"> <li>● Hyperinflammation (CRP ≥110 mg/L)</li> <li>● Bilateral pneumonia</li> <li>● Increase of more than 4 L/min in the previous 12 h in oxygen requirement</li> <li>● Mechanically ventilated patients included (2 in the anakinra group vs 4 in the SOT group)</li> </ul>	IV anakinra 300 mg daily for 5 d tapered to 200 mg daily for 2 d and 100 mg for 1 d (no. 12) vs SOT (no. 10)	<ul style="list-style-type: none"> <li>● Mortality: 0% in the anakinra group vs 10% in the SOT group (<i>P</i> = .45)</li> <li>● Ventilator-free days during the first 20 d (number of days alive and free from MV): 20 in the anakinra group vs 17 in the SOT group (<i>P</i> = .06)</li> <li>● Number of days with oxygen requirement &lt;3 L/min: 15.5 in the anakinra group vs 8 in the SOT group (<i>P</i> &lt; .05)</li> </ul>
Horby et al, <sup>14</sup> N Engl J Med 2020	Dexamethasone	Multicenter randomized open-label trial (United Kingdom)	<ul style="list-style-type: none"> <li>● Hospitalized patients with SARS-CoV-2 infection</li> <li>● Mechanically ventilated patients included</li> </ul>	SOT + oral or IV dexamethasone 6 mg once daily for up to 10 d (no. 2104) vs SOT (no. 4321)	<ul style="list-style-type: none"> <li>● Overall 28-d mortality: 22.9% in the dexamethasone group vs 25.7% in the SOT group (95% CI, 0.75-0.93); 29.3% vs 41.4% in mechanically ventilated patients (95% CI, 0.51-0.81); 23.3% vs 26.2% in patients with oxygen requirement (95% CI, 0.72-0.94); 17.8% vs 14.0% in patients with no respiratory support (95% CI, 0.91-1.55)</li> </ul>
Tomazini et al, <sup>15</sup> JAMA 2020	Dexamethasone	Multicenter randomized open-label trial (Brazil)	<ul style="list-style-type: none"> <li>● Mechanically ventilated patients only</li> <li>● MV for &lt;48 h</li> <li>● Moderate to severe ARDS (PaO<sub>2</sub>:FiO<sub>2</sub> ≤200 mm Hg)</li> <li>● No corticosteroid use in the previous 15 d</li> </ul>	SOT + IV dexamethasone 20 mg daily for 5 d followed by 10 mg daily for an additional 5 d or until ICU discharge (no. 151) vs SOT (no. 148)	<ul style="list-style-type: none"> <li>● Ventilator-free days during the first 28 d: 6.6 in the dexamethasone group vs 4.0 in the SOT group (<i>P</i> = .04)</li> <li>● 28-d mortality: 56.3% in the dexamethasone group vs 61.5% in the SOT group (<i>P</i> = .85)</li> </ul>
Fadel et al, <sup>16</sup> Clin Infect Dis 2020	MPD	Multicenter quasi-experimental study (United States)	<ul style="list-style-type: none"> <li>● Bilateral pneumonia</li> <li>● Oxygen requirement of 4 L/min or more, or escalating oxygen requirement from baseline</li> <li>● Mechanically ventilated patients included</li> </ul>	IV MPD 0.5-1 mg/kg/d for 3 d (up to 7 d in ICU patients) (no. 132) vs SOT (no. 81, historical controls)	<ul style="list-style-type: none"> <li>● Mortality: 13.6% in the MPD group vs 26.3% in the SOT group (<i>P</i> = .024)</li> <li>● Need for MV: 21.7% in the MPD group vs 36.6% in the SOT group (<i>P</i> = .025)</li> <li>● ICU admission during hospitalization: 27.3% in the MPD group vs 44.3% in the SOT group (<i>P</i> = .017)</li> <li>● Composite outcome (all 3 above): 34.9% in the MPD group vs 54.3% in the SOT group (<i>P</i> = .005)</li> </ul>

(Continued)

TABLE II. (Continued)

Reference	Investigated drug	Study design	Study population	Treatment/intervention	Outcomes
Ramiro et al, <sup>17</sup> Ann Rheum Dis 2020	MPD	Monocentric case-control study (prospective cohort with historical controls) (the Netherlands)	<ul style="list-style-type: none"> <li>● Hyperinflammation (at least 2: CRP <math>\geq</math>100 mg/L, ferritin <math>\geq</math>900 ng/mL, D-dimer <math>&gt;</math>1500 <math>\mu</math>g/L)</li> <li>● Bilateral pneumonia</li> <li>● Oxygen saturation of <math>\leq</math>94% in ambient air or tachypnea <math>&gt;</math>30/min</li> <li>● Mechanically ventilated patients included (1 in the MPD group vs 13 in the SOT group)</li> </ul>	IV MPD 250 mg on day 1 followed by 80 mg daily for 2-7 d with possible escalation with TCZ (single dose 8 mg/kg) at day 2-5 if worsening in clinical or respiratory status (no. 86) vs SOT (no. 86, historical controls)	<ul style="list-style-type: none"> <li>● Clinical improvement (2 points in the WHO 7-point ordinal scale): 74.4% in the MPD group vs 51.2% in the SOT group (<math>P = .0025</math>)</li> <li>● Mortality: 16.3% in the MPD group vs 47.7% in the SOT group (<math>P = .0004</math>)</li> <li>● Need for MV: 11.6% in the MPD group vs 27.9% in the SOT group (<math>P = .0003</math>)</li> </ul>

ICU, Intensive care unit; IV, intravenous; SC, subcutaneous; SOT, standard of therapy; TCZ, tocilizumab (humanized mAb against the IL-6 receptor); WHO, World Health Organization.

Allergology: Nicola Montano, Barbara Vigone, Chiara Bellocchi, Elisa Fiorelli, Valerie Melli, and Eleonora Tobaldini; Respiratory Unit and Cystic Fibrosis Adult Center: Francesco Blasi, Stefano Aliberti, Maura Spotti, Edoardo Simonetta, Leonardo Terranova, Francesco Amati, Carmen Miele, Sofia Misuraca, Alice D'Adda, Silvia Della Fiore, Marta Di Pasquale, Marco Mantero Martina Contarini, Margherita Ori, Letizia Morlacchi, Valeria Rossetti, Andrea Gramagna, Maria Pappalettera, Mirta Cavallini, and Annalisa Vigni; Cardiology Unit: Marco Vicenzi and Irena Rota. Emergency Medicine: Giorgio Costantino, Monica Solbiati, Ludovico Furlan, Marta Mancarella, Giulia Colombo, Giorgio Colombo, and Alice Fanin; Acute Internal Medicine: Valter Monzani, Angelo Rovellini, Laura Barbetta, Filippo Billi, and Christian Folli; Internal Medicine: Marina Baldini, Irena Motta, and Natalia Scaramellini; Internal Medicine and Metabolic Diseases: Anna Ludovica Fracanzani, Rosa Lombardi, and Federica Iuculano; Geriatric Unit: Matteo Cesari, Marco Proietti, and Laura Calcaterra. Istituto di Ricerche Farmacologiche Mario Negri IRCCS: Alessandro Nobili, Mauro Tettamanti, and Igor Monti.

**Clinical implications: In the search for an optimal support treatment, combination of high-dose anakinra plus MPD may be beneficial in COVID-19 severe pneumonia with hyperinflammation. This combined treatment is candidate for further investigation.**

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## METHODS

A secondary analysis of prospective, observational cohort studies (COVID-19\_Network; nCOV-2019\_ICU Study) was performed at Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy (Institutional Review Board #241\_2020; #236\_2020). All patients with COVID-19 who fulfilled the following inclusion criteria were analyzed: age more than 18 years; evidence of pneumonia; ferritin greater than or equal to 1000 ng/mL and/or CRP greater than 10 mg/dL (see Qin et al,<sup>E1</sup> Zhou et al,<sup>E2</sup> Chen et al,<sup>E3</sup> and King et al<sup>E4</sup>); respiratory failure with need of supplemental oxygen (oxygen therapy from 0.4 FiO<sub>2</sub> Venturi mask to invasive MV). Exclusion criteria were data available for less than 48 hours or death within 48 hours from inclusion; symptoms for less than 7 days; uncontrolled bacterial infections (ie, sepsis/septic shock); treatment with anti-IL-1 or MPD alone.

From March 5, 2020, patients were treated off-label with anti-IL-1 + MPD according to local standard operating procedures. Treatment was implemented at a different time in distinct settings (ie, COVID-19 intensive care unit [ICU], sub-ICU, internal medicine), starting from the ICU. Written informed consent for off-label use was obtained from all patients (except those on MV). The control group included patients with COVID-19 admitted and followed from February 25, 2020, to the time of anti-IL-1 + MPD introduction. Patients who retrospectively fulfilled all the inclusion and exclusion criteria for treatment were consecutively included in the control group.

Anakinra (Swedish Orphan Biovitrum AB, Stockholm, Sweden) was administered subcutaneously at 200 mg every 8 hours for 3 days, then 100 mg every 8 hours up to day 14.<sup>E5</sup> Patients on MV were treated with off-label intravenous administration (3-hour infusion time).<sup>E5-E8</sup> The intravenous route was chosen in view of the pharmacokinetic alterations of critically ill patients in the ICU (ie, high volume of distribution, massive generalized cutaneous edema consequent to water retention, and low albumin). Also, because patients on MV were on anticoagulant therapy, subcutaneous administration could lead to hematomas or infectious complications.

MPD was administered at 1 mg/kg loading dose, then 1 mg/kg/d (fractioned, 2 doses) for 5 days, then 0.5 mg/kg/d (fractioned, 2 doses) for 5 days, followed by 0.25 mg/kg/d (every 24 hours or fractioned) up to day 14.

All subjects received the treatment that was considered SOC at time of the study, which includes hydroxychloroquine in most cases and lopinavir/ritonavir in some. Some patients were also subjected to the use of experimental antiviral remdesivir through compassionate use (Table I). According to the hospital internal guidelines, all patients received antithrombotic prophylaxis/treatment with enoxaparin sodium during hospitalization. Specifically, until mid-March 2020, hospital guidelines recommended prophylaxis with 100 U/kg every 24 hours for patients weighing 80 kg and 5000 U every 12 hours for patients weighing 80 kg (if normal renal function), irrespective of the severity of the disease. From mid-March, based on increased observations of thromboembolic events in patients with severe COVID-19, dosage was

increased and stratified according to the severity of the disease (and the corresponding risk of thromboembolic events): 100 U/kg every 24 hours in COVID-19 internal medicine (70 U/kg every 12 hours for obese patients), 70 U/kg every 12 hours in COVID-19 sub-ICUs, and 100 U/kg every 12 hours in COVID-19 ICU. This latter scheme was considered in evaluating the percentages of anticoagulant therapy reported in Table I.

The primary outcome was 28-days survival rate. Adverse events were graded according to CTCAE\_v4.0. Differences between groups were assessed using 2-sample *t* test or Wilcoxon rank-sum test for parametric and nonparametric continuous variables and Fisher exact test for categorical variables. Study inclusion (t0) started at anti-IL-1 + MPD initiation (cases) or when ferritin/CRP levels above thresholds were registered (controls). Kaplan-Meier plots were used for survival data. Patients were followed from t0 to day 28 or death. If discharged earlier than day 28, patient status was assessed by postdischarge follow-up phone calls. Unadjusted and adjusted Cox proportional regression models were used after controlling for proportional hazards assumption. Factors associated with mortality at univariate analysis and hospitalization setting (days elapsed from hospitalization to t0, MV at inclusion) were considered as covariates. Statistical significance was set at  $\alpha$  less than 0.05. Analyses were performed using SAS software v.9.4 (SAS Institute Inc, Cary, NC).

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**TABLE E1.** Sensitivity analysis of the impact of anticoagulant therapy on the clinical outcome of treated and control patients

Characteristic	Ranges	No. of patients	Deaths	P value
Anti-IL-1 + MPD				
Anticoagulant therapy	No	24	2 (8.3)	.466
	Yes	41	7 (17.1)	
No anti-IL-1 + MPD				
Anticoagulant therapy	No	33	10 (30.3)	.554
	Yes	21	8 (38.1)	

Variables are reported as absolute number (percentage).

**TABLE E2.** Sensitivity analysis of the impact of antiviral therapy (lopinavir/ritonavir + hydroxychloroquine) on the clinical outcome of treated and control patients

Characteristic	Ranges	No. of patients	Deaths	P value
Anti-IL-1 + MPD				
Antiviral therapy	No	45	8 (17.8)	.169
	Yes	20	1 (5.0)	
No anti-IL-1 + MPD				
Antiviral therapy	No	13	3 (23.1)	.488
	Yes	39	13 (33.3)	

Variables are reported as absolute number (percentage). Three patients in the control group who were treated with lopinavir/ritonavir alone were excluded.

**TABLE E3.** Crude and adjusted Cox proportional regression models of the treatment with anakinra and MPD

	<b>Hazard ratio</b>	<b>95% CI</b>	<b>P value</b>
Crude	0.33	0.15-0.77	.007
Adjusted*	0.18	0.07-0.50	.001

The proportional hazards assumption was checked by using a transform of the Schoenfeld residuals and performing a supremum test of the null hypothesis that the observed pattern of martingale residuals was not different from the expected pattern (<https://stats.idre.ucla.edu/sas/seminars/sas-survival/>).

\*Adjusted Cox proportional regression model by sex, age, PaO<sub>2</sub>/FiO<sub>2</sub> at baseline, CCI, MV at inclusion, and days elapsed from hospitalization to inclusion.