Systematic Review in Immunology

Associations between Interleukin-4 and COVID-19 severity: A systematic review and meta-analysis

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Abstract

Introduction: This systematic review and meta-analysis aimed to determine the correlation between IL-4 concentrations and COVID-19 severity.

Methods: This study was designed as a systematic review and meta-analysis and was performed in accordance to the PRISMA statement. Titles, abstracts, and full texts of articles were independently reviewed by at least 2 authors. Continuous variables were compared by the mean difference (MD) with 95% confidence interval (CI).

Results: Thirty-three studies reported IL-4 levels among severe versus non-severe COVID-19 patients. Pooled analysis showed that levels of IL-4 among those groups varied and amounted to 2.72 ± 3.76 pg/mL vs 3.08 ± 4.14 pg/mL (MD = -0.26; 95%CI: -0.43 to -0.10; p = 0.002. In addition, eight studies reported levels of IL-4 among COVID-19 patients who survived vs deceased and was 2.61 ± 0.49 pg/mL vs (3.44 ± 16.4 pg/mL, respectively (MD = 0.22; 95%CI: 0.08 to 0.37; p = 0.002).

Discussion: This detailed systematic review and meta-analysis revealed that the plasma concentration of IL-4 is a potential risk factor for COVID-19 severity and mortality. Specifically, old age and male gender were associated with high IL-4 levels. Lung damage could result from the change in IL-4 concentration, thus making critical and severe COVID-19 cases at a very high risk of dying, thereby reducing their quality of life. Therefore, strategies such as using monoclonal antibodies to inhibit Th2 cytokines could be explored in developing an effective treatment regimen for COVID-19 patients.

Take-home message: An independent risk factor for the severity and fatality of COVID-19 is the plasma levels of IL-4. High IL-4 levels are specifically related to old age and male gender. Lung damage may be a result of the change in IL-4 concentration, placing COVID-19 critically and severely ill at a high risk of dying.

Keywords: Interleukin-4; IL-4; COVID-19; SARS-CoV-2; COVID-19 severity.

cytokine storm syndrome may be related to the severity of an individual’s COVID-19 status [10,11]. Serum interleukin levels significantly rose in severe and critical patients compared to mild COVID-19 cases, thus causing lung injury and acute respiratory distress. However, this does not connote that mild cases are not at risk for poor health outcomes [12–15]. Furthermore, there have been notable differences in cytokine profiles between COVID-19 patient survivors and non-survivors [16].

The cytokine storm and abnormal immune system have been noted, with leukocytes, neutrophils, infection biomarkers, and the concentrations of cytokines [interleukin (IL)-2R, IL-6, IL-8, IL-10, and tumor necrosis factor (TNF)-] being significantly higher in patients with SARS-CoV-2 infection [17,18]. These abnormal immune system changes include a decrease in the total number of T- and CD4+ cells [17]. The worse infection prognosis, the heightened inflammatory response, and the stimulation of the cytokine storm could all be explained by the consumption of CD4+ and CD8+ T cells [19].

Chang and colleagues conducted a systematic review investigating the associations between serum interleukins (IL-1β, IL-2, IL-4, IL-6, IL-8, and IL-10) and COVID-19 severity reported elevated levels of IL-6, IL-8, and IL-10 were associated with COVID-19 severity. In contrast, elevated levels of IL-1β, IL-6, and IL-8 were associated with poor COVID-19 prognosis [3]. No significant difference was found in IL-1β, IL-2, and IL-4 levels between severe and non-severe COVID-19 patients [3]. As the effector and inducer of this immunological mechanism, IL-4, the primary cytokine of the Th2 immune response, is crucial to the Th2 pathway. IL-4 and IL-13 are both primarily linked to fibrogenic inflammatory remodelling, whereas Th1 cells produce gamma interferon (IFN-) and IL-2 to inhibit fibrosis [20].

Some works of literature have reported significant associations between IL-6, IL-10, and IL-13; however, there exists a paucity of evidence on the existence of a significant association between IL-4 plasma concentrations and COVID-19 severity [20–23]. Therefore, it becomes crucial to compare the IL-4 load in the sera of patients with COVID-19 with those of healthy and recovered individuals to further understand the cellular mechanism behind the pathogenicity of COVID-19. Thus, this systematic review and meta-analysis aimed to determine the correlation between IL-4 concentrations and COVID-19 severity.

METHODS

This study was designed as a systematic review and meta-analysis and was performed according to the recommendations of the Cochrane Collaboration Group [24] and the Preferred Reporting Items for Systematic Reviews and Meta-analyses (PRISMA) guidelines [25].

Search strategy and study selection

Two authors (M.M. and M.P.) independently performed the literature search. PubMed Central, Scopus, EMBASE and Cochrane Collaboration Library were used for a comprehensive search of relevant studies from January 2020 to September 10, 2022. We used the following search terms: “Interleukin-4” OR “IL-4” AND “SARS-CoV-2” OR “novel coronavirus” OR “COVID-19”. Additionally, the reference lists of included papers were also manually searched for additional studies. Titles, abstracts, and full texts of articles identified from database search were imported into EndNote X9 (Clarivate Analytics, Philadelphia, USA).

Inclusion and exclusion criteria
The adopted inclusion criteria were as follows: (1) original articles; (2) COVID-19 patients in different clinical conditions: mild, moderate, severe or critical; (3) COVID-19 patients who survived hospital discharge or died on admission; (4) all types of observational studies: cohort, cross-sectional, case-control, longitudinal; (5) full-text articles published in English. Exclusion criteria were as follows: (1) studies, which did not meet the above criteria, (2) letters, posters, editorials, review articles and meta-analyses.

**Data extraction**
The data extraction exercise was conducted by two authors (M.P. and M.M.), and disagreements concerning the selection criteria were discussed and resolved by consensus, including six authors (A.A.A., O.S.I., F.C., M.B., M.A.-J. and L.S). Data were extracted from the included studies using a predefined form.

**Quality assessment**
Three authors (M.M., A.N. and M.P.) independently completed the quality assessment. Any disagreements were also resolved by discussion with the third reviewer (L.S.). We used the Newcastle-Ottawa scale (NOS) to assess the methodological quality of observational studies with its design. According to the NOS criteria, the studies were rated low, moderate, and high quality in accordance with the scores, 0–3, 4–6 and 7–9, respectively. Additionally, we performed funnel plot tests for asymmetry to investigate potential publication bias if there were more than 10 trials in a single meta-analysis.

**Statistical analysis**
All the meta-analyses were performed using the STATA software (version 14, StataCorp LLC, College Station, TX, USA) and the RevMan software (version 5.4, The Cochrane Collaboration, Copenhagen, Denmark). Mean differences (MD) and 95% confidence intervals (CIs) were calculated to build forest plots of continuous data and evaluate differences in IL-4 concentrations between COVID-19 patients with severe vs non-severe groups or survivor vs non-survivor status during follow-up. P-values of <0.05 were considered to indicate statistical significance. In situations where IL-4 levels were reported as median with interquartile range, estimated means and standard deviations with the formula described by Hozo were used [26]. We evaluated heterogeneity between studies using the p-value of the Q-test and the F statistic. F of <50% was considered low or moderate heterogeneity, and a fixed-effects model was used. We additionally performed a sensitivity analysis to evaluate the influence of any given study on the pooled estimate.

**RESULTS**

**Study characteristics**
Based on the above-mentioned inclusion criteria, we identified 5,722 reports and screened their summaries for eligibility after removing duplicates. Overall, 1,394 articles were screened according to the titles and abstracts. Full-text screening was performed on 55 studies, and data for 39 studies [27–66] were extracted for this meta-analysis. A flow chart of the literature search and study selection is presented in Figure 1. Thirty-three studies reported the IL-4 values among severe vs. non-severe COVID-19 patients. Eight studies reported the correlation coefficient between IL-4 concentration and COVID-19 survivability.
The systematic review included articles published between 2020 and 2022, comprising 8,722 COVID-19 participants. The baseline characteristics of selected studies are presented in Table 1. The study quality assessed using the NOS scores was ≥7 for all included trials (Table 1).

Figure 1. Flowchart detailing selection and screening of the studies included in this review.
<table>
<thead>
<tr>
<th>Study and year</th>
<th>Country</th>
<th>Study group</th>
<th>No. of patients</th>
<th>Age (ys)</th>
<th>Sex, male IL-4, pg/mL</th>
<th>NOS score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Akinosoglou et al, 2022 [27]</td>
<td>Greece</td>
<td>Severe</td>
<td>9</td>
<td>61.5 ± 28</td>
<td>6 (66.7%)</td>
<td>4.9 ± 24.9</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>18</td>
<td>63.0 ± 19</td>
<td>9 (50.0%)</td>
<td>0.0 ± 17.8</td>
</tr>
<tr>
<td>Belaid et al, 2022 [28]</td>
<td>Algeria</td>
<td>Severe</td>
<td>26</td>
<td>66.8 ± 12.3</td>
<td>21 (80.8%)</td>
<td>0.67 ± 0.38</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>31</td>
<td>53.7 ±14.4</td>
<td>19 (61.3%)</td>
<td>1.05 ± 0.38</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Survival</td>
<td>42</td>
<td>NS</td>
<td>NS</td>
<td>1.01 ± 0.37</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Death</td>
<td>15</td>
<td>NS</td>
<td>NS</td>
<td>0.64 ± 0.35</td>
</tr>
<tr>
<td>Cabaro et al, 2021 [29]</td>
<td>Italy</td>
<td>Severe</td>
<td>19</td>
<td>67.5 ± 4.0</td>
<td>15 (78.9%)</td>
<td>3.44 ± 0.58</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>46</td>
<td>57.8 ± 5.8</td>
<td>27 (58.7%)</td>
<td>3.59 ± 0.79</td>
</tr>
<tr>
<td>Chi et al, 2020 [30]</td>
<td>China</td>
<td>Severe</td>
<td>8</td>
<td>54.0 ±12.3</td>
<td>5 (62.5%)</td>
<td>0.09 ± 0.95</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>58</td>
<td>41.8 ± 14.5</td>
<td>32 (55.2%)</td>
<td>9.35 ± 1.16</td>
</tr>
<tr>
<td>Gadotti et al, 2020 [31]</td>
<td>Brazil</td>
<td>Survival</td>
<td>38</td>
<td>56.8 ± 7.3</td>
<td>23 (60.5%)</td>
<td>1.65 ± 0.15</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Death</td>
<td>18</td>
<td>66.3 ± 5.3</td>
<td>16 (88.9%)</td>
<td>1.53 ± 0.18</td>
</tr>
<tr>
<td>Gao et al, 2021 [32]</td>
<td>China</td>
<td>Severe</td>
<td>32</td>
<td>54.3 ± 11.4</td>
<td>20 (62.5%)</td>
<td>1.3 ± 1.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>135</td>
<td>48.1 ±13.7</td>
<td>51 (37.8%)</td>
<td>1.6 ± 0.9</td>
</tr>
<tr>
<td>Gil-Etayo et al, 2021 [33]</td>
<td>Spain</td>
<td>Survival</td>
<td>46</td>
<td>53 ± 4.5</td>
<td>30 (65.2%)</td>
<td>2.44 ± 1.25</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Death</td>
<td>9</td>
<td>87 ± 2.8</td>
<td>7 (77.8%)</td>
<td>96.33 ± 109.65</td>
</tr>
<tr>
<td>He et al, 2020 [34]</td>
<td>China</td>
<td>Severe</td>
<td>33</td>
<td>43.6 ± 10.4</td>
<td>18 (54.5%)</td>
<td>1.52 ± 0.91</td>
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<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>60</td>
<td>36.3 ± 8.0</td>
<td>31 (51.7%)</td>
<td>1.67 ± 1.06</td>
</tr>
<tr>
<td>He et al, 2020 [35]</td>
<td>China</td>
<td>Severe</td>
<td>69</td>
<td>62.0 ± 5.5</td>
<td>37 (53.6%)</td>
<td>4.30 ± 0.27</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>135</td>
<td>42.5 ± 3.7</td>
<td>42 (31.1%)</td>
<td>3.76 ± 0.03</td>
</tr>
<tr>
<td>Hu et al, 2020 [36]</td>
<td>China</td>
<td>Severe</td>
<td>13</td>
<td>61.5 ± 2.5</td>
<td>8 (61.5%)</td>
<td>1.03 ± 0.29</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>63</td>
<td>48.2 ± 1.1</td>
<td>26 (41.3%)</td>
<td>1.30 ± 0.23</td>
</tr>
<tr>
<td>Jin et al, 2021 [37]</td>
<td>China</td>
<td>Severe</td>
<td>40</td>
<td>55.5 ± 15</td>
<td>19 (47.5%)</td>
<td>2.95 ± 2.44</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>106</td>
<td>43.8 ± 12.8</td>
<td>58 (54.7%)</td>
<td>2.24 ± 0.98</td>
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<tr>
<td>Lai et al, 2022 [38]</td>
<td>China</td>
<td>Survival</td>
<td>2,343</td>
<td>58.0 ± 3.3</td>
<td>1.072 (45.8%)</td>
<td>2.50 ± 0.30</td>
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<tr>
<td></td>
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<td>Death</td>
<td>212</td>
<td>69.5 ± 2.7</td>
<td>150 (70.8%)</td>
<td>2.12 ± 0.18</td>
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<tr>
<td>Li et al, 2020 [39]</td>
<td>China</td>
<td>Survival</td>
<td>1,327</td>
<td>54.0 ± 4.0</td>
<td>643 (48.5%)</td>
<td>3.0 ± 0.33</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Death</td>
<td>122</td>
<td>69.8 ± 2.5</td>
<td>90 (73.8%)</td>
<td>2.5 ± 0.33</td>
</tr>
<tr>
<td>Liao, 2020 [40]</td>
<td>China</td>
<td>Severe</td>
<td>231</td>
<td>67.7 ± 3.0</td>
<td>137 (59.3%)</td>
<td>2.16 ± 0.19</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>149</td>
<td>55.3 ± 4.3</td>
<td>69 (46.3%)</td>
<td>2.05 ± 0.15</td>
</tr>
<tr>
<td>Ling et al, 2021 [41]</td>
<td>China</td>
<td>Severe</td>
<td>17</td>
<td>64.0 ± 3.5</td>
<td>11 (64.7%)</td>
<td>2.15 ± 1.30</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>15</td>
<td>47.5 ± 8.5</td>
<td>4 (26.7%)</td>
<td>4.72 ± 2.55</td>
</tr>
<tr>
<td>Author</td>
<td>Year</td>
<td>Country</td>
<td>Severe Group</td>
<td>Non-severe Group</td>
<td>Severe Rate (%)</td>
<td>Non-severe Rate (%)</td>
</tr>
<tr>
<td>-------------------------</td>
<td>-------</td>
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</tr>
<tr>
<td>Liu et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>62.8 ± 2.8</td>
<td>100 (49.5%)</td>
</tr>
<tr>
<td>Liu et al, 2021</td>
<td>2021</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>64.7 ± 3.8</td>
<td>33 (58.8%)</td>
</tr>
<tr>
<td>Lv et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>61.1 ± 10.4</td>
<td>117 (49.0%)</td>
</tr>
<tr>
<td>Meng et al, 2021</td>
<td>2021</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>51.8 ± 5.8</td>
<td>17 (63.0%)</td>
</tr>
<tr>
<td>Nie et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>57.5 ± 5.0</td>
<td>13 (52.0%)</td>
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<td>Ozger et al, 2021</td>
<td>2021</td>
<td>Turkey</td>
<td>Survival</td>
<td>Death</td>
<td>58.1 ± 7.4</td>
<td>20 (69.0%)</td>
</tr>
<tr>
<td>Queiroz et al, 2022</td>
<td>2022</td>
<td>Brazil</td>
<td>Severe</td>
<td>Non-severe</td>
<td>67.9 ± 16.9</td>
<td>25 (62.5%)</td>
</tr>
<tr>
<td>Ren et al, 2021</td>
<td>2021</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>55.8 ± 4.1</td>
<td>30 (71.4%)</td>
</tr>
<tr>
<td>Rutkowska et al, 2021</td>
<td>2021</td>
<td>Poland</td>
<td>Severe</td>
<td>Critical</td>
<td>59.1 ± 12.0</td>
<td>14 (93.3%)</td>
</tr>
<tr>
<td>Shi et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>61.2 ± 5.5</td>
<td>16 (51.6%)</td>
</tr>
<tr>
<td>Song et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>63.0 ± 4.9</td>
<td>63 (63.0%)</td>
</tr>
<tr>
<td>Wang et al, 2021</td>
<td>2021</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>69.9 ± 12.6</td>
<td>71 (58.7%)</td>
</tr>
<tr>
<td>Wei et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>62.5 ± 4.5</td>
<td>27 (69.2%)</td>
</tr>
<tr>
<td>Wu et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>57.9 ± 11.8</td>
<td>18 (75.0%)</td>
</tr>
<tr>
<td>Yang et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>61.7 ± 9.2</td>
<td>5 (35.7%)</td>
</tr>
<tr>
<td>Yin et al, 2021</td>
<td>2021</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>68.3 ± 5.4</td>
<td>26 (46.4%)</td>
</tr>
<tr>
<td>Yuan et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>63.3 ± 4.3</td>
<td>30 (49.2%)</td>
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<tr>
<td>Zhang et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Severe</td>
<td>Non-severe</td>
<td>61.7 ± 9.2</td>
<td>5 (35.7%)</td>
</tr>
<tr>
<td>Zhang et al, 2020</td>
<td>2020</td>
<td>China</td>
<td>Survival</td>
<td>Death</td>
<td>37.5 ± 2.8</td>
<td>32 (34.4%)</td>
</tr>
</tbody>
</table>
Thirty-three studies reported IL-4 levels among patients with severe vs. non-severe COVID-19 patients. Pooled analysis showed that levels of IL-4 among those groups varied and amounted to 2.72 ± 3.76 pg/mL vs. 3.08 ± 4.14 pg/mL (MD = -0.26; 95%CI: -0.43 to -0.10; p=0.002; Figure 2).

Eight studies reported levels of IL-4 among COVID-19 patients who survived vs. deceased. IL-4 levels among patients who survived were 2.61 ± 0.49 pg/mL and were statistically significantly lower than in the deceased group (3.44 ± 16.4 pg/mL; MD = 0.22; 95%CI: 0.08 to 0.37; p=0.002; Figure 3).

In addition, based on the available articles, we averaged the data on IL-4 levels and obtained IL-4 levels at 2.55 ± 0.4 pg/mL in the asymptomatic COVID-19 patients’ group, 2.89 ± 5.16 pg/mL among mild COVID-19 patients’ group, 2.76 ± 1.98 pg/mL in the moderate COVID-19 patients’ group, 2.68 ± 3.33 pg/mL in the severe COVID-19 patients’ group and 2.63 ± 5.85 pg/mL in the critical COVID-19 patients’ group (Figure 4).

Sensitivity analysis based on the leave-one-out analysis showed that a single trial did not influence the pooled results. The above dependence applied to all comparisons included in the meta-analysis.

<table>
<thead>
<tr>
<th>Study</th>
<th>Country</th>
<th>Severe</th>
<th>Non-severe</th>
<th>IL-4 Level (Mean ± SD)</th>
<th>p-value</th>
<th>Nature of Death (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zhang et al, 2020 [63]</td>
<td>Italy</td>
<td>Severe</td>
<td>67</td>
<td>NS</td>
<td>3.29 ± 0.32</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>81</td>
<td>NS</td>
<td>3.44 ± 0.42</td>
<td></td>
</tr>
<tr>
<td>Zheng et al, 2020 [64]</td>
<td>China</td>
<td>Severe</td>
<td>74</td>
<td>56.9 ± 3.1</td>
<td>0.85 ± 0.03</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>22</td>
<td>47 ± 5.6</td>
<td>0.90 ± 0.20</td>
<td></td>
</tr>
<tr>
<td>Zhu et al, 2020 [65]</td>
<td>China</td>
<td>Severe</td>
<td>16</td>
<td>57.5 ± 11.7</td>
<td>1.99 ± 0.47</td>
<td>8</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>111</td>
<td>49.9 ± 15.5</td>
<td>1.93 ± 0.19</td>
<td></td>
</tr>
<tr>
<td>Zhu et al, 2021 [66]</td>
<td>China</td>
<td>Severe</td>
<td>17</td>
<td>56.8 ± 11.6</td>
<td>1.92 ± 0.50</td>
<td>9</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-severe</td>
<td>125</td>
<td>48.0 ± 16.6</td>
<td>1.84 ± 0.22</td>
<td></td>
</tr>
</tbody>
</table>

Note: NS = Not specified
Figure 2. Forest plot of interleukin 4 levels among severe vs. non-severe COVID-19 patients. The center of each square represents the mean differences for individual trials, and the corresponding horizontal line stands for a 95% confidence interval. The diamonds represent pooled results.

Figure 3. Forest plot of interleukin 4 levels among COVID-19 patients who survive vs. decrease. The center of each square represents the mean differences for individual trials, and the corresponding horizontal line stands for a 95% confidence interval. The diamonds represent pooled results.

DISCUSSION

This systematic review and meta-analysis found that COVID-19 alters IL-4 plasma concentration levels in COVID-19-positive individuals, regardless of symptomatology. We observed no remarkable difference in the plasma concentration of IL-4 in severe and non-severe COVID-19-positive cases. However, IL-4 levels were significantly lower among COVID-19 survivors compared to the deceased. It is also interesting to note that IL-4 plasma concentration was lowest in both severe and non-severe COVID-19 groups compared to the concentration of other cytokines [33]. These
findings are similar to the results of a clinical trial conducted among healthy, moderate, and severe COVID-19 patients enrolled from three public hospitals in the Erbil city, Kurdistan, Iraq where the recovery group had lower levels of IL-4 compared to the severe COVID-19 group [67]. Notably, treatment with antiviral medications did not effectively lower IL-4 concentration among COVID-19 cases presenting with mild symptoms [67], thus increasingly placing the individuals at risk for poor prognosis and likely mortality.

Figure 4. Mean interleukin 4 levels among different COVID-19 severity groups.

From this study, we identified that a large fraction of males had poor COVID-19 outcomes overall. In COVID-19 infection, male sex was found to be a risk factor for severe and critical illness and greater mortality [68–70]. Understanding the gender differences in COVID-19 outcomes is crucial for clinical care and remission strategies [68, 71]. It is important to note that IL-4 has been demonstrated to activate various signalling pathways crucial for controlling cell growth. IL-1 activation inhibits several crucial cytokines that may be released by proinflammatory monocytes, preventing macrophages from becoming cytotoxic and even creating nitric oxide [72, 73]. TNF-, IL-1, and PGE-2 are other inflammatory cytokines that are inhibited by the release and activation of IL-4, ultimately stimulating the IL-4 receptor [7]. LDL oxidation, which reduces inflammation, is also increased. On the other hand, IL-4 can effectively activate JAK-STAT, causing infertility issues in men as one of its adverse effects. It has also been demonstrated that Th2 cells can activate this interleukin, stimulating the STAT signalling pathway to cause apoptosis [74]. However, if Th2 levels were increased, patients should undergo intensive treatment as SARS-CoV-2 has been observed to dramatically enhance Th2, Th1/Th17 cells, and antibody production in the body of patients with COVID-19 [71].

This study found that COVID-19-positive cases in severe and critical conditions belonged to older age groups compared to those in less severe conditions. Also, people who died due to COVID-19-related causes were older than the survived cohort. Literature suggests a higher prevalence of systemic pro-inflammatory cytokines and a reduced prevalence of systemic anti-inflammatory cytokines as people age [74, 75]. As a result, “inflamme-aging”—a chronic inflammatory condition—
may develop in older subjects [74,75]. Numerous investigations have shown that older adults had higher levels of the inflammatory proteins IL-6, IL-1, tumor necrosis factor (TNF), and C-reactive protein (CRP) [76,77]. The precise cause of the cytokine storm in elderly persons with severe COVID-19 infection is not yet known [78]. The likelihood of a cytokine storm and subsequent acute respiratory distress syndrome in some elderly patients with severe COVID-19 infection, however, is probably significantly influenced by disruption of the cytokine homeostasis in the "inflamm-aging" phenomena [79]. The inflammatory phenotype of senescent cell activity, particularly in adipose tissue, immune-senescence, and lack of vitamin D content, as well as age-related pathophysiologic processes, are associated with the "cytokine storm" phenomenon in elderly patients with severe COVID-19 infection [80–84]. These processes include altered angiotensin-converting enzyme 2 (ACE2) receptor expression, excess ROS production, and altered autophagy [85,86].

**Strengths and limitations**

A solid point of our study is the comprehensive inclusion of all previous research on the subject of IL-4 in the disease, which is COVID-19, as well as a detailed analysis of all related factors, thus providing current evidence required for improving the management of COVID-19-positive patients. However, there are some limitations to the study. The considerable heterogeneity of the studies included in the meta-analysis and the observational character of the studies are the first and most significant limitations (retrospective analysis). Another drawback might be that some drugs affect the amounts of circulating biomarkers and influence the prognosis for COVID-19. As a result, it is important to reconsider the same biomarkers in light of the current treatments. Another problem is the small patient populations in the studies that made up the meta-analysis.

**CONCLUSION**

This detailed systematic review and meta-analysis revealed that the plasma concentration of IL-4 is a potential risk factor for COVID-19 severity and mortality. Specifically, old age and male gender were associated with high IL-4 levels. Lung damage could result from the change in IL-4 concentration, thus placing critical and severe COVID-19 cases at a very high risk of dying, thereby reducing their quality of life. Therefore, strategies such as using monoclonal antibodies to inhibit Th2 cytokines could be explored in developing an effective treatment regimen for COVID-19 patients.

**Author Contributions:** Conceptualization: MM, LS.; methodology: MM, FC, LS.; software: MP, LS.; validation: LS, MM, FC; formal analysis, LS, MM; investigation: MM, MP, MB, AAA, OSI, AN, LS, MY, GN, FC; resources: MM, LS; data curation: MP, MM, FC, AAA, MB, OSI, MA]; writing—original draft preparation: MM, MP, FC, AAA, OSI; writing—review and editing, all authors; visualization: MM, LS; supervision: LS; project administration: MM. All authors have read and agreed to the published version of the manuscript.

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**Conflicts of Interest:** None

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reveals IL-6 and IL-10 are disease severity predictors. Emerg Microbes Infect. 2020;9(1):1123–1130.


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