


# Circulating B10 regulatory cells are decreased in severe and critical COVID-19

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## Abstract

The contribution of B cells in COVID-19 pathogenesis, beyond the production of specific antibodies against SARS-CoV-2, is still not well understood. Since one of their most relevant functional roles includes their immune-suppressive mechanisms, we decided to evaluate one of the most recognized human B regulatory subpopulations: the IL-10<sup>+</sup> B10 cells, during COVID-19 onset. After stimulation of PBMCs for IL-10 induction, we employed multiparametric flow cytometry to determine B10 frequencies in severe and critical COVID-19 patients and then correlated those with clinical and laboratory parameters. Compared with healthy individuals, we detected a significant reduction in the B10 subset in both patient groups, which correlates with some inflammatory parameters that define the disease severity. This evidence suggests an aberrant role of B10 cells in immune responses against SARS-CoV-2 that needs to be further explained.

## KEYWORDS

B cells, B10, Bregs, COVID-19

## 1 | INTRODUCTION

The recent emergence of Coronavirus disease 2019 (COVID-19) pandemic has dramatically accelerated the characterization of diverse antiviral immune-mediated mechanisms aimed at controlling this emergent illness. COVID-19 is caused by the Severe Acute Respira-

tory Syndrome CoronaVirus 2 (SARS-CoV-2) and may generate asymptomatic or mild symptomatic infection in most patients. In contrast, other infected people suffer from severe acute respiratory distress syndrome (ARDS) with a poor prognosis.<sup>1</sup>

The severity of COVID-19 depends on the balance of host immune responses against viral stimuli. Deep immune-profiling studies of SARS-CoV-2 infected patients have documented several immune dysfunctions that correlate with disease severity, including lymphopenia, altered type I IFNs response, myeloid cell aberrations, and high levels of inflammatory cytokines (cytokine storm).<sup>2-4</sup> The latter has a significant role in severe/critical COVID-19 manifestations, including ARDS.<sup>5</sup>

**Abbreviations:** ARDS, acute respiratory distress syndrome; ASC, Antibody-secreting cells; Bregs, B regulatory cells; COVID-19, Coronavirus disease 2019; FiO<sub>2</sub>, fraction of inspired oxygen; FMO, Fluorescence Minus One; PaO<sub>2</sub>, Arterial partial pressure of oxygen; PBMCs, Peripheral blood mononuclear cells; ROC, Receiver operating characteristic (curve); SARS-CoV-2, Severe acute respiratory syndrome coronavirus 2; SO<sub>2</sub>, Oxygen saturation; ssRNA, Single-stranded RNA; TLRs, Toll-like receptors; Tregs, T regulatory cells.

Diverse peripheral immune regulatory mechanisms have evolved to control and limit the aberrant hyperinflammatory responses. Regulatory T cells (Tregs) constitute a subset of CD4<sup>+</sup> T cells representing the most recognized players in preventing exacerbated immune responses. Tregs inhibit the activation of both innate and adaptive inflammatory responses through diverse mechanisms, including the secretion of immunosuppressive cytokines such as TGF- $\beta$ , IL-35, and IL-10.<sup>6</sup> Although limited information is available regarding human Tregs in acute viral infections, some evidence indicates that they contribute to host protection by downregulating immunopathogenic mechanisms of tissue damage.<sup>7,8</sup> Remarkably, these cells have been described to be highly reduced during COVID-19 onset.<sup>9,10</sup> Additionally, some reports indicate the success of the adoptive transfer of allogeneic Tregs as a therapeutic option for this disease.<sup>11</sup> Since the outcome of viral diseases depends on the interplay between pathogen and host pro- and anti-inflammatory responses, a dysbalanced regulatory cell number and/or function against SARS-CoV-2 could be associated with COVID-19 pathogenesis and severity.<sup>9</sup>

Interestingly, other immune-suppressive elements than Tregs have not received enough attention yet; these not-enough characterized players include the diverse subsets of regulatory B cells (Bregs). B cells develop “classical” immune functions, including antibody production and antigen presentation.<sup>12</sup> More recently, Bregs have been found to exert immunoregulatory activity,<sup>13</sup> although they account for a small percentage (around 0.5%) of the total B lymphocytes in healthy humans.<sup>14</sup> These B cells are characterized by their suppressive properties of immune-mediated inflammatory responses. Also, they could facilitate the inflammation-recovery processes, thus contributing to maintaining immune homeostasis through different effector mechanisms, including the secretion of anti-inflammatory cytokines such as IL-10.<sup>15,16</sup> Among the different and well-characterized Bregs in humans,<sup>13</sup> we can highlight one of the first described and most abundant subpopulation: the mature CD24<sup>hi</sup>CD27<sup>+</sup> B lymphocytes named as B10 cells due to their capability of IL-10 secretion in response to innate signals such as TLR7/9 ligation, that has been shown to suppress pro-inflammatory responses.<sup>17,18</sup>

Breg implications during SARS-CoV-2 infection have not been widely explored, but indirect evidence indicates that total B cells (CD19<sup>+</sup>) producing IL-10 are significantly reduced in convalescent COVID-19 patients with a poor outcome.<sup>19</sup> Besides that, it has been well documented that acute COVID-19 correlates with impaired type I interferon (IFN-I) responses, predominantly in critically unwell patients.<sup>20</sup> Interestingly, it has been shown that some IL-10<sup>+</sup> Breg subsets depend on antiviral IFN-I as a critical signal for their differentiation,<sup>21</sup> thus making it possible that B10 cells could be affected in COVID-19 patients. Additionally, our previous findings have shown a decline in total memory CD27<sup>+</sup> B cells in severe and critical COVID-19 cases.<sup>22</sup> As the B10 subset is included in this CD27<sup>+</sup> compartment, these regulatory cells could also be found numerically reduced in these individuals. Therefore, we hypothesized that these regulatory B10 cells could also be involved or affected in the pathogenesis of COVID-19. Thus, we look for them in a small cohort of hospitalized patients.

## 2 | RESULTS AND DISCUSSION

Beyond Ab production or Ag-presentation abilities, B cells can also secrete IL-10 that restricts inflammation and counteracts potential pro-inflammatory cytokine excessive production. IL-10 secreted by these cells, further denominated collectively as Bregs, acts as an immunoregulator that inhibits pro-inflammatory reactions, thus preventing tissue damage derived from exacerbated innate or adaptive immune responses against pathogenic stimuli.<sup>15,18</sup>

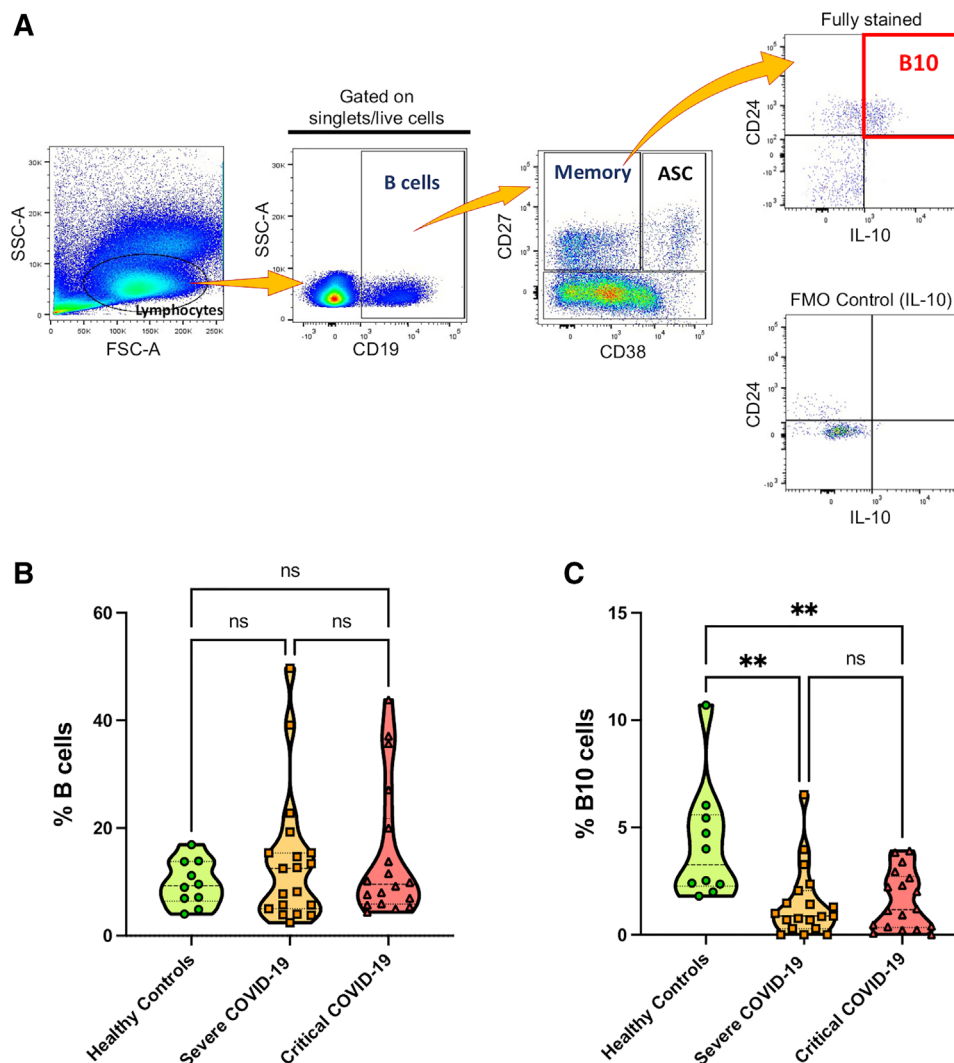
The SARS-CoV-2 infection seems to be mainly associated with a systemic hyper-inflammatory status where several immune-suppressive mechanisms appeared to be altered or dysfunctional in these patients, thus contributing to disease pathogenesis.<sup>23,24</sup> Accordingly, and up today, only one report indicates that total CD19<sup>+</sup> IL-10<sup>+</sup> B cells are reduced in the circulation of acute COVID-19 patients.<sup>19</sup> This observation only suggests that one or more Breg subsets could be affected since most of the identified and well defined human CD19<sup>+</sup> Breg subpopulations produce IL-10, including the CD24<sup>hi</sup>CD38<sup>hi</sup> transitional B cells, the CD24<sup>+</sup>CD38<sup>+</sup>TIM-1<sup>+</sup> B cells, the CD27<sup>int/hi</sup>CD38<sup>hi</sup> plasmablasts, and the mature CD24<sup>hi</sup>CD27<sup>+</sup> B10 cells.<sup>15,25</sup>

Therefore, we analyzed those B cell subsets by flow cytometry during the acute COVID-19 onset, finding that most do not display alterations in their frequencies (Supplementary Fig. 1A–C), except for the B10 subpopulation. Accordingly, using a simple multiparametric flow cytometry strategy (Fig. 1A), we first segregated total CD19<sup>+</sup> B cells from PBMCs samples; then CD27<sup>+</sup> CD24<sup>hi</sup> cells were gated to identify IL-10-producers as B10 cells.

As previously reported,<sup>22</sup> we could not detect any significant differences among frequencies (Fig. 1B) or absolute numbers (Supplementary Fig. 2) of total CD19<sup>+</sup> B cells of any group. Interestingly, regulatory B10 cells displayed a significant reduction in their frequencies in severe and critical COVID-19 patients compared to healthy controls (Fig. 1C).

The reduced circulating B10 cells are possibly a result of the impaired IFN-I responses, previously reported in severe or critical COVID-19 patients.<sup>20</sup> Accordingly, both COVID-19 and influenza viral infections have been associated with an impaired IFN-I and -III host response relative to other pathogens; more interestingly, the COVID-19 severity seems to correlate with the degree of IFN-I/III impairment.<sup>26,27</sup> As mentioned before, IFN- $\alpha$  (IFN-I) is a crucial signal for some IL-10<sup>+</sup> Breg subsets differentiation, both in mouse or human models.<sup>21,28</sup> Hence it would be possible that B10 cells could be depleted under this specific infectious context.

Moreover, this reduction in B10 cell numbers after SARS-CoV-2 infection, in response to excessive inflammation present in acute severe/critical COVID-19, could also result from innate receptors-mediated stimulation of these cells. It has been recently reported that mouse B10 cells (CD19<sup>+</sup>CD1d<sup>hi</sup>CD5<sup>+</sup>) significantly reduced their frequency and IL-10-producing capacity after TLR7 overexpression or overstimulation with synthetic agonists.<sup>29</sup> Additionally, the decline in the number and functional capacity of this Breg subset is enhanced by high levels of IFN- $\gamma$ .<sup>29</sup> Although human B10 are phenotypically different from their mouse equivalent, it is possible that during COVID-19 acute onset, single-stranded RNA (ssRNA) fragments from the

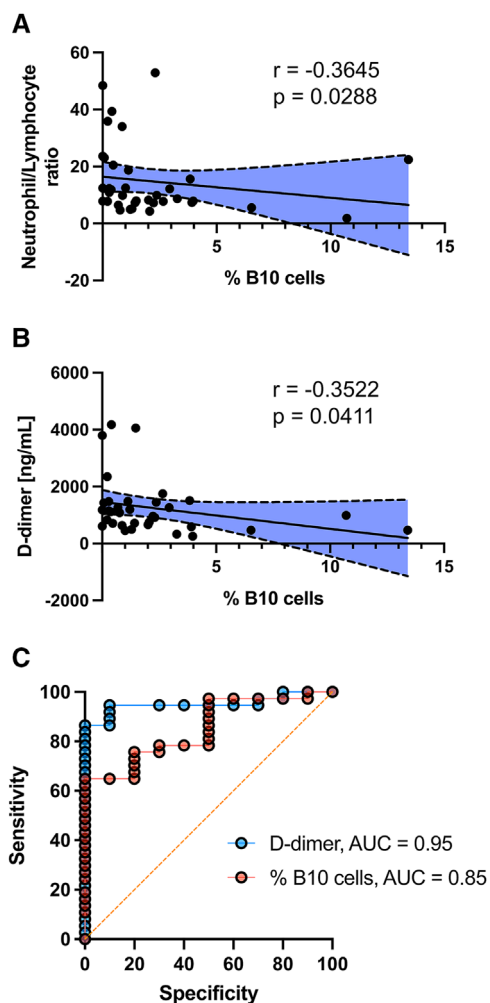


**FIGURE 1** Circulating B10 cell frequencies are significantly reduced in severe and critical COVID-19 patients. (A) Gating strategy for the identification of the B10 subset from CD19<sup>+</sup> B cells, CD27<sup>+</sup> (memory) and CD24<sup>hi</sup> cells (excluding the CD38<sup>hi</sup> Ab-secreting cells, ASC), previously selected from singlets gate (SSC-A vs. SSC-H), live Zombie UV<sup>-</sup> cells, and lymphocytes region gated from an SSC-A vs. FSC-A density plot. Data are representative of a healthy control. (B) Frequency of total CD19<sup>+</sup> B cells in PBMCs from patients infected with SARS-CoV-2 (total n = 37; 19 severe and 18 critical) and healthy controls (n = 10; all with negative PCR for SARS-CoV-2). (C) Frequency of B10 (CD27<sup>+</sup> CD24<sup>hi</sup> IL-10<sup>+</sup>) cells in PBMCs from patients infected with SARS-CoV-2 (n = 37; 19 severe and 18 critical) and healthy controls (n = 10; all negative PCR for SARS-CoV-2). All frequency values are displayed as mean (dashed line) plus lower and upper quartiles (dotted lines). The data were analyzed by a Kruskal-Wallis test followed by a Dunn's post hoc test. \*\* $p \leq 0.01$ .

SARS-CoV-2 genome act as direct activators of their endosomal TLR7 pathway, as demonstrated for monocyte-derived DCs.<sup>30</sup> This stimulation, enhanced by high levels of IFN- $\gamma$  described in acute-COVID-19 patients,<sup>31–33</sup> maybe also induce the depletion of human B10 cells.

The contraction of the B10 subpopulation in hospitalized COVID-19 patients raises the possibility that the loss of these cells could be linked or contribute to the inflammatory pathogenesis of their severe disease. Correspondingly, it has been reported that total CD19<sup>+</sup> B cells from convalescent patients of COVID-19 with a favorable clinical outcome displayed a higher IL-10 production upon TLR9 activation, compared to those with a poor outcome with abnormal chest-radiographic findings,<sup>19</sup> suggesting that a positive outcome may be associated with the expansion of regulatory B cells.

To gain insight into the involvement of B10 lymphocytes in regulating excessive inflammation in acute COVID-19 and their potential utility as follow up clinical markers, we calculated Spearman's correlation coefficients between the B10 cell frequencies and the clinical and laboratory available features (depicted in Supplementary Table 1) of both severe and critical COVID-19 patients. As expected, the B10 cell subset diminishing seems to be associated with a hyper-inflammatory status in severe and critical COVID-19 patients, at least demonstrated by the significant correlations displayed between their numbers and well established inflammation-associated clinical parameters such as neutrophil/lymphocyte ratio (Fig. 2A) and the D-dimer presence (Fig. 2B), that has been previously described as highly increased in these patients.<sup>22,34,35</sup>



**FIGURE 2** Frequencies of circulating B10 cells correlate with clinical parameters assessed in hospitalized COVID-19 patients. Correlation analysis calculating Spearman's coefficient ( $r$ ) between the B10 subset frequencies and indicated laboratory variables: neutrophil/lymphocyte ratio (A) or D-dimer (B). Values from  $n = 37$  hospitalized COVID-19 patients (19 severe and 18 critical). Significant  $p$ -values ( $p < 0.05$ ) are shown. C) Receiver operator characteristic (ROC) curves of circulating B10 cell frequencies and D-dimer serum levels for the discrimination of COVID-19 severe/critical disease (constructed from data of  $n = 37$  hospitalized COVID-19 patients and  $n = 10$  healthy controls). Area under the curve (AUC) values are depicted

Finally, to assess the potential usefulness of B10 frequency measurement in medical diagnosis or management of COVID-19 patients, we generated a receiver operating characteristic (ROC) curve to determine the discriminative capacity of B10 percentages in hospitalized (severe/critical) patients vs. healthy donors. To appreciate its significance, we decided to compare it to a well-defined COVID-19-related biomarker.

Diverse meta-analyses have shown that D-dimer levels possess prognostic value, correlate with disease severity and in-hospital mortality, and can be used as an early marker to guide the management of SARS-CoV-2 infected individuals.<sup>36</sup> For comparative purposes, we

also constructed a ROC curve for D-dimer. After determining the areas under the curve (AUC) of both parameters, we observed that B10 cell percentage displayed an outstanding discriminative value (AUC = 0.85) distinguishing hospitalized COVID-19 patients, not so far from that given when measuring D-dimer levels (AUC = 0.95) (Fig. 2C).

Limitations of our study include the lack of additional viral-respiratory disease controls such as influenza or respiratory syncytial virus-infected patients, which would allow us to determine if B10 cell changes could also be detected in different infectious contexts. Moreover, although our work is also limited by the missing group of mild COVID-19 patients, thus not allowing evaluation of the potential of B10 percentage as a severity-associated marker, our results indicate that measurement of these cells could be helpful in clinical contexts and need to be more explored, even in longitudinal cohort studies, to a better understanding of its prognostic value.

In conclusion, our results indicate that circulating B10 cells are decreased during SARS-CoV-2 infection and can be considered as potential biomarkers for severe COVID-19. It remains unclear if this phenomenon is specific to SARS-CoV-2 and whether this loss resulted from these cells differentiating into IL-10 producing plasma cells or undergoing apoptosis, thus warranting future studies focusing on delineating the mechanisms by which this or other viral infections could negatively regulate B10 cell development.

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#### DISCLOSURES

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest. The data that support the findings of this study are available from the corresponding author, upon reasonable request.

#### AUTHORSHIP

R.C.-D., V.A.S.-H., and S.R.-R. contributed equally to this work, designed and performed experiments, analysis, and interpretation of data. D.E.M.-S. assisted in the processing and preservation of patient samples. J.T.-R. and A.P.-F. collected patient samples, data, generated, and organized our clinical database. R.C.-D., V.A.S.-H., and S.R.-R. assisted in writing and editing the manuscript. D.G.-M. and J.L.M.-M. conducted experiments, supervised general work, wrote, and edited the

manuscript. All authors contributed to the article and approved the submitted version.

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## REFERENCES

- Schultze JL, Aschenbrenner AC. COVID-19 and the human innate immune system. *Cell*. 2021;184:1671-92.
- Lee JS, Shin EC. The type I interferon response in COVID-19: implications for treatment. *Nat Rev Immunol*. 2020;20:585-6.
- Mathew D, Giles JR, Baxter AE, et al. Deep immune profiling of COVID-19 patients reveals distinct immunotypes with therapeutic implications. *Science*. 2020;369:1-17.
- Wang J, Jiang M, Chen X, Montaner LJ. Cytokine storm and leukocyte changes in mild versus severe SARS-CoV-2 infection: Review of 3939 COVID-19 patients in China and emerging pathogenesis and therapy concepts. *J Leukoc Biol*. 2020;108:17-41.
- Luo XH, Zhu Y, Mao J, Du RC. T cell immunobiology and cytokine storm of COVID-19. *Scand J Immunol*. 2021;93:1-15. (e12989).
- Plitas G, Rudensky AY. Regulatory T Cells: Differentiation and function. *Cancer Immunol Res*. 2016;4:721-5.
- Karkhah A, Javanian M, Ebrahimpour S. The role of regulatory T cells in immunopathogenesis and immunotherapy of viral infections. *Infect Genet Evol*. 2018;59:32-7.
- Anghelina D, Zhao J, Trandem K, Perlman S. Role of regulatory T cells in coronavirus-induced acute encephalitis. *Virology*. 2009;385:358-67.
- Rahimzadeh M, Naderi N. Toward an understanding of regulatory T cells in COVID-19: A systematic review. *J Med Virol*. 2021;93:4167-81.
- Galvan-Pena S, Leon J, Chowdhary K, et al. Profound Treg perturbations correlate with COVID-19 severity. *Proc Natl Acad Sci U S A*. 2021;118:1-9.
- Gladstone DE, Kim BS, Mooney K, Karaba AH, D'Alessio FR. Regulatory T Cells for treating patients with COVID-19 and acute respiratory distress syndrome: two case reports. *Ann Intern Med*. 2020;173:852-3.
- Romero-Ramirez S, Navarro-Hernandez IC, Cervantes-Diaz R, et al. Innate-like B cell subsets during immune responses: beyond antibody production. *J Leukoc Biol*. 2019;105:843-56.
- Mauri C, Menon M. Human regulatory B cells in health and disease: Therapeutic potential. *J Clin Invest*. 2017;127:772-9.
- Chekol Abebe E, Asmamaw Dejenie T, Mengie Ayele T, Dagnaw Baye N, Agegnehu Teshome A, Tilahun Muche Z. The role of regulatory B cells in health and diseases: A systemic review. *J Inflamm Res*. 2021;14:75-84.
- Maravillas-Montero JL, Acevedo-Ochoa E. Human B regulatory cells: the new players in autoimmune disease. *Rev Invest Clin*. 2017;69:243-6.
- Rosser EC, Mauri C. Regulatory B cells: origin, phenotype, and function. *Immunity*. 2015;42:607-12.
- Iwata Y, Matsushita T, Horikawa M, et al. Characterization of a rare IL-10-competent B-cell subset in humans that parallels mouse regulatory B10 cells. *Blood*. 2011;117:530-41.
- Wang L, Fu Y, Chu Y. Regulatory B Cells. *Adv Exp Med Biol*. 2020;1254:87-103.
- Shuwa HA, Shaw TN, Knight SB, et al. Alterations in T and B cell function persist in convalescent COVID-19 patients. *Med (N Y)*. 2021;2:720-35 e4.
- Acharya D, Liu G, Gack MU. Dysregulation of type I interferon responses in COVID-19. *Nat Rev Immunol*. 2020;20:397-8.
- Menon M, Blair PA, Isenberg DA, Mauri C. A regulatory feedback between plasmacytoid dendritic cells and regulatory b cells is aberrant in systemic lupus erythematosus. *Immunity*. 2016;44:683-97.
- Sosa-Hernandez VA, Torres-Ruiz J, Cervantes-Diaz R, et al. B Cell Subsets as severity-associated signatures in COVID-19 Patients. *Front Immunol*. 2020;11:1-12.
- De Candia P, Prattichizzo F, Garavelli S, Matarese G. T Cells: warriors of SARS-CoV-2 Infection. *Trends Immunol*. 2021;42:18-30.
- Tascioglu D, Akkaya E, Genc S. The understanding of the immunopathology in COVID-19 infection. *Scand J Clin Lab Invest*. 2021;81:255-63.
- Menon M, Hussell T, Ali Shuwa H. Regulatory B cells in respiratory health and diseases. *Immunol Rev*. 2021;299:61-73.
- Zhou Z, Ren L, Zhang L, et al. Heightened innate immune responses in the respiratory tract of COVID-19 Patients. *Cell Host Microbe*. 2020;27:883-90 e2.
- Blanco-Melo D, Nilsson-Payant BE, Liu WC, et al. Imbalanced host response to SARS-CoV-2 drives development of COVID-19. *Cell*. 2020;181:1036-45 e9.
- Obieglo K, Costain A, Webb LM, et al. Type I interferons provide additive signals for murine regulatory B cell induction by *Schistosoma mansoni* eggs. *Eur J Immunol*. 2019;49:1226-34.
- Chodisetti SB, Fike AJ, Domeier PP, Choi NM, Soni C, Rahman ZSM. TLR7 negatively regulates b10 cells predominantly in an ifngamma signaling dependent manner. *Front Immunol*. 2020;11:1-12.
- Salvi V, Nguyen HO, Sozio F, et al. SARS-CoV-2-associated ssRNAs activate inflammation and immunity via TLR7/8. *JCI Insight*. 2021;6:1-15.
- Galani IE, Rovina N, Lampropoulou V, et al. Untuned antiviral immunity in COVID-19 revealed by temporal type I/III interferon patterns and flu comparison. *Nat Immunol*. 2021;22:32-40.
- Gadotti AC, de Castro Deus M, Telles JP, et al. IFN-gamma is an independent risk factor associated with mortality in patients with moderate and severe COVID-19 infection. *Virus Res*. 2020;289:1-7.
- Yang L, Liu S, Liu J, et al. COVID-19: immunopathogenesis and Immunotherapeutics. *Signal Transduct Target Ther*. 2020;5:128.
- Iwamura APD, Tavares da Silva MR, Hummelgen AL, et al. Immunity and inflammatory biomarkers in COVID-19: a systematic review. *Rev Med Virol*. 2021;31:1-11. (e2199).
- Soria-Castro R, Meneses-Preza YG, Rodriguez-Lopez GM, et al. Severe COVID-19 is marked by dysregulated serum levels of carboxypeptidase A3 and serotonin. *J Leukoc Biol*. 2021;110:425-31.
- Samprathi M, Jayashree M. Biomarkers in COVID-19: an up-to-date. *Review Front Pediatr*. 2020;8:1-12.

## SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

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