Letter to *Blood*

TO THE EDITOR:

The BTK inhibitor ibrutinib may protect against pulmonary injury in COVID-19–infected patients

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The Bruton tyrosine kinase (BTK) inhibitor ibrutinib is used to treat indolent B-cell malignancies and chronic graft-versus-host disease (cGVHD). The potential for ibrutinib to abrogate pulmonary inflammatory cytokines, lung injury, and death was demonstrated in a highly relevant lethal flu animal model.[1] Therefore, we sought to clarify the impact of ibrutinib in COVID-19 patients. We care for 600 to 800 Waldenstrom macroglobulinemia (WM) patients each year, ~300 of whom are on a BTK inhibitor. We identified 6 patients receiving ibrutinib for WM who were diagnosed with COVID-19; these patients consented to the use of their data. Their clinical characteristics appear in Table 1. Their median age was 66 years, and 5 were on the recommended treatment dose of 420 mg/d; the sixth patient was on a reduced dose of 140 mg/d because of arthralgias. For all patients, the median time on ibrutinib was 52 months. Their median time with COVID-19–related symptoms prior to diagnostic testing was 5 days, and the median time since diagnosis of COVID-19 was 22 days. All 6 patients experienced cough and fever as prodromal symptoms. The 5 patients on ibrutinib, 420 mg/d, did not experience dyspnea and did not require hospitalization. Their course was marked by steady improvement, and resolution or near resolution of COVID-19–related symptoms during the follow-up period.

The patient on reduced-dose ibrutinib (Patient 6; Table 1) experienced progressive dyspnea and hypoxia prompting hospitalization. Chest computed tomography showed bilateral ground glass opacities and a pleural effusion on admission prompting a hold on ibrutinib, during which his hypoxia acutely worsened, necessitating supplemental oxygen use. Hydroxychloroquine (HCQ) and azithromycin were administered. Azithromycin was stopped after 3 days because of wide QRS complex tachyarrhythmia; HCQ was given for a total of 5 days. Hypoxia worsened and fever persisted during HCQ course. Ibrutinib was restarted at 140 mg/d, and tocilizumab, 400 mg, was coadministered on hospital day 5 with improved oxygenation, as well as decreased C-reactive protein (CRP) levels (83 mg/L to 9 mg/L). IV immunoglobulin was also given on hospital days 6 through 10. On day 10 of hospitalization, the patient experienced worsening hypoxia that was accompanied by increased CRP (28 mg/L) and required mechanical ventilation. Given the lack of hypoxia in the other COVID-19–infected WM patients on full-dose ibrutinib, ibrutinib was increased to 420 mg/d on days 11 and 12. A rapid improvement in oxygenation followed, and the patient was successfully extubated late on day 12 and maintained oxygen

saturations of 94% to 96% on 3 L/min supplemental oxygen by nasal cannula. The next day, supplemental oxygen was decreased to 2 L/min, with oxygen saturations of 96% to 98% and a CRP level of 10 mg/L. On day 14, oxygen saturation was 95% on room air, repeat CRP level was 6 mg/L, and he was discharged home off supplemental oxygen and on 420 mg/d of ibrutinib. Seven days later, he continues to do well, without fever, cough, or dyspnea at rest. He remains on ibrutinib, 420 mg/d, and is tolerating therapy well.

Pulmonary failure is the main cause of mortality related to COVID-19 infection.[2,3] Up to 80% of patients hospitalized for COVID-19 infection require supplemental oxygenation, of whom 30% to 40% may require mechanical ventilation.[2,4,5] SARS-CoV-2 binds via the ACE2 receptor that is highly expressed on alveolar type II (ATII) cells in the lung.[6] ATII cells constitute 5% to 15% of the lung epithelium. Although ATI cells are highly adapted for gas exchange, ATII cells have a specialized role in innate immune response.[7-9] ATII cells express Toll-like receptors (TLRs) and can trigger inflammatory cytokines and chemoattractants in response to pathogens that recruit and activate other immune cells, including macrophages and neutrophils.[7-9] Highly relevant to coronavirus infection, expression of proinflammatory and chemoattractant cytokines interleukin-1β (IL-1β), IL-6, IP-10/CXCL10, monocyte chemoattractant protein-1 (MCP-1), and tumor necrosis factor-α (TNF-α) was identified in ACE2$^+$ cells from autopsy tissue of SARS-CoV-1–infected patients, which appeared to be causally related to the acute lung injury and pathogenesis observed with SARS-CoV-1.[10] A similar profile of elevated cytokine levels was reported in the plasma of SARS-CoV-1 patients during the progressive and end stage of infection,[11] which was consistent with an M1-polarized macrophage response.[12]

SARS-CoV-1 shares 86% homology with SARS-CoV-2. SARS-Cov-2 patients requiring intensive care also showed elevated plasma levels of inflammatory cytokines and chemoattractants, such as IL-2, IL-6, IL-7, IL-10, granulocyte colony-stimulating factor, IP-10/CXCL-10, MCP-1/CCL2, MIP-1a/CCL3, and TNF-α.[13] The importance of inflammatory cytokines to lung injury in SARS-CoV-2–infected patients has been suggested by reports of benefit with IL-6 and IL-6 receptor–blocking antibodies, and clinical trials to examine their use have been initiated (NCT04317092, NCT04306705, NCT04315298).

We and other investigators previously showed that BTK and its upstream activator HCK were involved in TLR-mediated

Table 1. Clinical characteristics of 6 patients with WM on ibrutinib with COVID-19 infection

Demographics	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5	Patient 6
Age, y	65	61	72	67	71	58
Sex	M	M	F	F	M	M
Time since B-cell diagnosis, mo	39	54	95	202	52	107
Received treatment prior to ibrutinib for WM	No	No	Yes	Yes	No	Yes
Time on ibrutinib, mo	39	54	83	50	47	85
Dose of ibrutinib, mg/d	420	420	420	420	420	140-HELD-420
COVID-19 symptoms						
Time with symptoms prior to COVID-19 diagnostic testing, d	5	2	6	7	10	5
Time since COVID-19 diagnostic testing, d	24	20	17	28	13	29
Cough	Yes	Yes	Yes	Yes	Yes	Yes
Fever	Yes	Yes	Yes	Yes	Yes	Yes
Dyspnea	No	No	No	No	No	Yes
Sore throat	Yes	No	No	No	No	Yes
Taste loss	No	No	Yes	No	Yes	No
Smell loss	No	No	Yes	No	Yes	No
Hospitalization	No	No	No	No	No	Yes
Required ICU admission	Yes	No	No	No	No	Yes
Required supplemental O2	No	No	No	No	No	Yes
Required mechanical ventilation	No	No	No	No	No	Yes
Other COVID-19 symptoms	No	Anorexia	Diarrhea	Headache	No	No
Other medication for COVID-19	HCQ, AZ	NA	No	NA	No	HCQ, AZ, TOCI
Disposition						
COVID-19 symptoms resolved	No	Yes	Yes	Yes	Yes	No
COVID-19 symptoms persist	Yes	No	Yes	Yes	No	Yes
COVID-19 symptoms improved	Yes	Yes	Yes	Yes	Yes	Yes

140-HELD-420 denotes that this patient was on 140 mg/d of ibrutinib prior to hospitalization that was held upon admission; he experienced worsening hypoxia after ibrutinib was held and required mechanical ventilation, following which he was restarted on 420 mg/d of ibrutinib and showed rapid improvement in oxygenation.

AZ, azithromycin; F, female; HCQ, hydroxychloroquine; ICU, Intensive Care Unit; M, male; TOCI, tocilizumab.

signaling.[14-16] BTK and HCK are triggered by MYD88, a TLR adaptor protein that signals for all TLRs, with the exception of TLR3, in response to viral and bacterial pathogens, including coronaviruses.[17] ATII cells express TLRs, as do alveolar macrophages that coordinate inflammatory responses with ATII cells.[7-9] As components of TLR/MYD88 signaling, BTK and HCK can drive inflammatory cytokine production through ERK1/2.[18]

In a transgenic mouse model, activated HCK overexpression promoted extensive pulmonary inflammation and an enhanced innate immune response, particularly in older mice.[19] Elevated levels of TNF-α were identified in the bronchoalveolar lavage fluids of these mice following lipopolysaccharide challenge. The pulmonary pathology findings from these mice show great overlap with those from patients with COVID-19 infection, which included serous and fibrin exudation with alveolar infiltration consisting mostly of macrophages and monocytes.[20]

Ibrutinib is a highly potent covalent inhibitor of BTK (biochemical 50% inhibitory concentration [IC_{50}], 0.5 nM). Ibrutinib is also a potent reversible inhibitor of HCK (IC_{50}, 49 nM). The IC_{50} levels for BTK and HCK are within the pharmacologically attainable dosimetry of orally administered ibrutinib.[16] Serially collected blood samples

from patients with chronic lymphocytic leukemia (CLL), WM, and cGVHD on ibrutinib monotherapy showed marked reductions in proinflammatory and chemoattractant cytokines that greatly overlapped with those reported to be elevated in the plasma of SARS-CoV-1 and SARS-CoV-2 patients, as well as in ACE2+ cells from lung tissue of SARS-CoV-1 patients (Table 2).[10,11,13,21-23] In the ILLUMINATE randomized study, CLL subjects treated with ibrutinib immediately prior to infusion with obinutuzumab also showed significantly decreased levels of inflammatory cytokines associated with infusion-related reactions (a cytokine release syndrome).[24] These findings are consistent with a shift from an M1- to an M2-polarized macrophage response following ibrutinib and are supported by preclinical and clinical studies showing dependence of macrophage lineage commitment on BTK function.[25]

The potential for ibrutinib to abrogate lung injury and death was also demonstrated in an experimental model wherein mice challenged with a lethal intranasal inoculum of a mouse-adapted strain of H1N1 influenza virus were protected against lung injury. Control mice developed respiratory failure, along with histological and computed tomography findings consistent with lung injury, in sharp contrast to the mice that received ibrutinib.[1] Control mice also lost weight and died, whereas those treated with ibrutinib

Table 2. Summary of proinflammatory and chemoattractant cytokine patterns in patients infected with SARS-CoV-1 and SARS-CoV-2 and following ibrutinib treatment in patients with CLL, WM, or and cGVHD

	He et al[10]	Jiang et al[11]	Huang et al[13]	Niemann et al[21]	Greil et al[24]	Vos et al[22]	Miklos et al[23]
Patient population	CoV-1*	CoV-1*	CoV-2*	CLL on ibrutinib†	CLL on ibrutinib†	WM on ibrutinib†	cGVHD on ibrutinib†
Tissue	ACE2+ cells	Plasma	Plasma	Plasma	Plasma	Plasma	Plasma
GM-CSF			↑				↓
IL-1β	↑						
IL-2			↑				↓ (IL2RA)
IL-6	↑	↑		↓	↓	↓	
IL-7			↑				
IL-8		↑		↓	↓	↓	↓
IL-10			↑	↓	↓	Variable	
IP-10/CXCL10		↑	↑	↓		↓	↓
MCP-1/CCL2	↑	↑	↑	↓	↓		↓
MIP-1A/CCL3			↑	↓			↓
MIP-1B/CCL4			↑	↓		↓	↓
TNF-α	↑			↓	↓	↓	↓

↑, denotes elevated in patients with SARS-CoV-1 or SARS-CoV-2; ↓, denotes levels decreased or inhibited in patients with the indicated condition with ibrutinib treatment; GM-CSF, granulocyte-macrophage colony-stimulating factor.

*Patients infected with SARS-CoV-1 or SARS-CoV-2.

†Patients with CLL, WM, or cGVHD.

recovered their weight after a brief loss, and all survived.[1] Notably, mice treated with ibrutinib also showed decreased inflammatory cell infiltration, as well as proinflammatory cytokines in lung tissues, that included proinflammatory and chemoattractant cytokines, such as IL-1β, IL-6, KC/CXCL1, TNF-α, and MCP-1, in SARS-Cov-1 and SARS-CoV-2 patients.[1] The findings provide rationale that an exaggerated cytokine release syndrome triggered in ATII cells and resident macrophages by SARS-CoV-2 may underlie pulmonary injury associated with COVID-19.

Therefore, ibrutinib, and possibly other BTK inhibitors, may provide protection against lung injury and even improve pulmonary function in hypoxic patients with COVID-19, as we observed in this series of WM patients on ibrutinib. These findings should be considered hypothesis generating and preliminary in nature. Patients on ibrutinib, and possibly other BTK inhibitors, may well benefit with continuation of their therapy, despite the diagnosis of COVID-19. It will be important to validate these findings in other patient populations who are taking BTK inhibitors, including CLL patients. Clinical trials examining the benefit of BTK inhibitors are being initiated by us and other investigators in COVID-19 patients in pulmonary distress, and the outcome of these prospective randomized studies will be needed to confirm these preliminary observations.

Authorship

Contribution: S.P.T. conceptualized and designed the study and wrote the first draft; J.J.C., A.P.S., I.M.G., and K.M. provided patient care and data; S.P.T., J.D.S., M.L.G., and G.Y. provided input for supportive basic science studies; and all authors provided editorial review.

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