A Case of COVID-19 and *Pneumocystis jirovecii* Co-infection

Aravind A. Menon, M.D.[1]*, David D. Berg, M.D.[2]*, Elliot J. Brea, M.D., Ph.D.[3], Aaron J. Deutsch, M.D.[3], Khameer K. Kidia, M.D., M.Phil.[3], Emilia G. Thurber, M.D.[3], Sylvie B. Polsky, M.D.[3], Tiffany Yeh, M.D.[3], Jonathan A. Duskin, M.D.[3], Alison M. Holliday, M.D., M.P.H.[3], Elizabeth B. Gay, M.D.[1‡], Laura E. Fredenburgh, M.D.[1‡§]

[1] Division of Pulmonary and Critical Care Medicine, Brigham and Women's Hospital, Boston, MA; [2] Division of Cardiovascular Medicine, Brigham and Women's Hospital, Boston, MA; [3] Department of Medicine, Brigham and Women's Hospital, Boston, MA

* These authors contributed equally to this work.

‡ These authors contributed equally to this work.

§ **Corresponding Author:**

Laura E. Fredenburgh, M.D.

Division of Pulmonary and Critical Care Medicine Brigham and Women's Hospital

75 Francis Street

Boston, MA 02115

Phone: (617) 525-9563

lfredenburgh@bwh.harvard.edu

Author Contributions: A.A.M., D.D.B., E.B.G., and L.E.F. contributed to the literature review, data collection, and drafted the manuscript. All authors participated in the clinical care of the patient, and read, revised, and approved the manuscript.

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

To the Editor:

Lymphocytopenia has been identified as a common laboratory finding in patients with SARS-CoV-2 infection, particularly among those with more severe presentations (1); however, there are limited data on which specific lymphocyte populations may be affected or the clinical sequelae. In this report, we describe the case of a woman with hypoxemic respiratory failure found to have co-infection with SARS-CoV-2 and *Pneumocystis jirovecii*, a pathogen commonly seen in patients with defects in T cell immunity.

An 83-year-old female nonsmoker presented to our hospital on March 12, 2020 with fevers, malaise, headache, dry cough, and dyspnea. She had a history of mild intermittent asthma, managed with an albuterol inhaler as needed, mitral valve prolapse with moderate to severe mitral regurgitation, and mild-moderate ulcerative colitis, which was well-controlled on oral budesonide (3 mg daily and being tapered), as well as maintenance-dose sulfasalazine (1500 mg twice daily). Her symptoms had started approximately two weeks prior to presentation, shortly after travel from Florida to Massachusetts, and had failed to improve with courses of azithromycin and amoxicillin-clavulanate. In the emergency department, she had a fever of 39.3°C and oxygen saturation of 86% on room air, which improved to 95% on 5 liters per minute (L/min) of supplemental oxygen by nasal cannula. Initial laboratory evaluation revealed leukocytosis and relative lymphocytopenia (absolute lymphocyte count 1090 cells/μL) **(Table 1)**. Chest computed tomography (CT) was notable for diffuse bilateral ground glass opacities with patchy bands of atelectasis and small nodular foci of consolidation with a distribution suggestive of a viral pneumonia. Subtle cystic

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

changes were also seen in the affected regions **(Figure 1)**. She was admitted to the medical intensive care unit (ICU) and placed on strict isolation precautions given concern for community acquired SARS-CoV-2. She developed worsening tachypnea with a respiratory rate of 40 breaths per minute, and hypoxia with an oxygen saturation of 80% requiring supplemental oxygen through a non-rebreather mask at a rate of 15 L/min. An arterial blood gas showed a partial pressure of arterial oxygen (PaO_2) of 63 mm Hg on 15 L/min of supplemental oxygen. She was intubated for hypoxemic respiratory failure and supported on low tidal volume ventilation according to the Acute Respiratory Distress Syndrome (ARDS) Network lower tidal volume protocol. Her PaO_2/FiO_2 (ratio of the PaO_2 to the fraction of inspired oxygen [FiO_2]) was consistent with moderate ARDS.

A broad infectious work-up for viral, bacterial, and fungal organisms **(Table 1)** confirmed the diagnosis of SARS-CoV-2 infection, based on positive detection for the presence of SARS-CoV-2 RNA from a nasopharyngeal swab (N1 target [Ct 31.33]; N2 target [Ct 33.38]; RNase P control [Ct 25.66]), a qualitative test result (positive when Ct<40.00 for N1 and N2 targets) reported by the MA State Public Health Laboratory using its FDA Emergency Use Authorization approved CDC 2019-nCoV Real-Time RT-PCR Diagnostic Panel. In addition, a serum (1,3)-β-D-glucan level was markedly elevated at 305 pg/mL (reference value <80 pg/mL), prompting additional testing for *P. jirovecii* with a qualitative real-time PCR assay from a tracheal aspirate, which was positive (fluorescent value 0.160 at melting temperature [Tm] 62.4°C; minimum fluorescent signal intensity for positive test ≥0.020). Notably, she had no apparent clinical characteristics associated with false positive (1,3)-β-D-glucan measurements,

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

such as exposure to hemodialysis membranes, intravenous immunoglobulin, albumin, gauze packing, or intravenous β-lactam antibiotics. Human immunodeficiency virus (HIV)-1/2 antibody/antigen testing was non-reactive. However, a CD4+ T lymphocyte count was low at 291 cells/μL (reference value 441-2156 cells/μL) as was the CD4+/CD8+ ratio (1.18; reference value 1.20-5.30). She was treated with trimethoprim-sulfamethoxazole (TMP-SMX) and successfully extubated on hospital day 7. A follow-up serum (1,3)-β-D-glucan level obtained one week after initiating treatment was significantly reduced (90 pg/mL). Moreover, a follow-up CD4+ T lymphocyte count obtained ten days after initial presentation demonstrated improvement (730 cells/μL).

CD4+ T lymphocytes play a critical role in the immune response against *P. jirovecii.* Classically, when patients with untreated HIV develop severe CD4+ lymphocytopenia (<200 cells/μL), the risk of *Pneumocystis* pneumonia increases significantly (2). In the present case, we hypothesize that SARS-CoV-2 infection led to a state of functional immune suppression related to CD4+ lymphocytopenia, which then predisposed the patient to *P. jirovecii* infection. Although the patient's CD4+ T cell count was >200 cells/μL, the sample was collected nearly a week into her course after her total lymphocyte count had started to recover. It is also possible that an underlying immune defect predisposed the patient independently to SARS-CoV-2 and *P. jirovecii* infection; however, the patient did not have a known underlying immunodeficiency, nor did she have any classical risk factors for *Pneumocystis* pneumonia, such as malignancy, organ transplantation, or prolonged exposure to systemic corticosteroids. Although patients with inflammatory bowel disease on systemic corticosteroids, biologics, and other immunosuppressants may be at

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

increased risk of *Pneumocystis* pneumonia (3), the overall incidence in ulcerative colitis is low (approximately 8/100,000 person-years) (4) and has not been associated with oral budesonide use (5). Given the high sensitivity of *P. jirovecii* PCR (6), *Pneumocystis* colonization cannot be completely excluded. However, taken together, the highly positive PCR test, significant elevation in (1,3)-β-D-glucan, cystic lesions on chest imaging, progressive hypoxemia in the setting of CD4+ lymphocytopenia, and response to TMP-SMX therapy are highly supportive of a diagnosis of *Pneumocystis* pneumonia.

Respiratory viral infections, particularly influenza, predispose patients to the development of secondary bacterial infections (7) and invasive fungal infections, including aspergillosis, most notably in immunocompromised patients (8). While no cases of *Pneumocystis* pneumonia have been reported in patients infected with SARS-CoV-1 or Middle East respiratory syndrome (MERS) coronavirus, co-infection with *P. jirovecii* has been reported in HIV and hematopoietic stem cell transplant patients with influenza A infection (9, 10). Furthermore, two cases of *Pneumocystis* pneumonia and H1N1 influenza A co-infection have been reported in immunocompetent patients, possibly secondary to influenza-induced CD4+ lymphocytopenia (11).

There is emerging evidence that patients with SARS-CoV-2 are at high risk for co-infection (12), and this case highlights the importance of being vigilant about excluding treatable respiratory pathogens, including *P. jirovecii.* Since COVID-19 and *Pneumocystis* pneumonia may share common clinical features (e.g., bilateral multifocal infiltrates, profound hypoxemia), co-infection with *P. jirovecii* may not be

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

appreciated in patients with severe SARS-CoV-2 infection. It may therefore be reasonable to consider additional diagnostic testing for *P. jirovecii* in patients with SARS-CoV-2 infection, particularly when there are other clinical characteristics that may support co-infection (e.g., elevated LDH, cystic findings on chest CT), even in the absence of classical *P. jirovecii* risk factors. Finally, this case extends the potential utility of (1,3)-β-D-glucan testing for diagnosing *Pneumocystis* pneumonia (13) in patients with suspected SARS-CoV-2 infection, which is particularly relevant given concerns about healthcare transmission associated with performing bronchoscopy in these patients.

Acknowledgments

We would like to thank Sandra C. Smole, Ph.D., Director of the MA State Public Health Laboratory, Bureau of Infectious Disease and Laboratory Sciences for assistance with interpretation of the results of the SARS-CoV-2 Real-Time RT-PCR assay.

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

References

1. Guan WJ, Ni ZY, Hu Y, Liang WH, Ou CQ, He JX, Liu L, Shan H, Lei CL, Hui DSC, Du B, Li LJ, Zeng G, Yuen KY, Chen RC, Tang CL, Wang T, Chen PY, Xiang J, Li SY, Wang JL, Liang ZJ, Peng YX, Wei L, Liu Y, Hu YH, Peng P, Wang JM, Liu JY, Chen Z, Li G, Zheng ZJ, Qiu SQ, Luo J, Ye CJ, Zhu SY, Zhong NS, China Medical Treatment Expert Group for Covid-19. Clinical characteristics of coronavirus disease 2019 in China. *N Engl J Med* 2020. doi: 10.1056/NEJMoa2002032 (In press).

2. Thomas CF, Jr., Limper AH. Pneumocystis pneumonia. *N Engl J Med* 2004;350:2487-2498.

3. Cotter TG, Gathaiya N, Catania J, Loftus EV, Jr., Tremaine WJ, Baddour LM, Harmsen WS, Zinsmeister AR, Sandborn WJ, Limper AH, Pardi DS. Low risk of pneumonia from Pneumocystis jirovecii infection in patients with inflammatory bowel disease receiving immune suppression. *Clin Gastroenterol Hepatol* 2017;15:850-856.

4. Long MD, Farraye FA, Okafor PN, Martin C, Sandler RS, Kappelman MD. Increased risk of Pneumocystis jiroveci pneumonia among patients with inflammatory bowel disease. *Inflamm Bowel Dis* 2013;19:1018-1024.

5. Lichtenstein GR, Bengtsson B, Hapten-White L, Rutgeerts P. Oral budesonide for maintenance of remission of Crohn's disease: a pooled safety analysis. *Aliment Pharmacol Ther* 2009;29:643-653.

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

6. Arcenas RC, Uhl JR, Buckwalter SP, Limper AH, Crino D, Roberts GD, Wengenack NL. A real-time polymerase chain reaction assay for detection of Pneumocystis from bronchoalveolar lavage fluid. *Diagn Microbiol Infect Dis* 2006;54:169-175.

7. Martin-Loeches I, van Someren Greve F, Schultz MJ. Bacterial pneumonia as an influenza complication. *Curr Opin Infect Dis* 2017;30:201-207.

8. Schauwvlieghe A, Rijnders BJA, Philips N, Verwijs R, Vanderbeke L, Van Tienen C, Lagrou K, Verweij PE, Van de Veerdonk FL, Gommers D, Spronk P, Bergmans D, Hoedemaekers A, Andrinopoulou ER, van den Berg C, Juffermans NP, Hodiamont CJ, Vonk AG, Depuydt P, Boelens J, Wauters J, Dutch-Belgian Mycosis study group. Invasive aspergillosis in patients admitted to the intensive care unit with severe influenza: a retrospective cohort study. *Lancet Respir Med* 2018;6:782-792.

9. Franconi I, Monari C, Tutone M, Ciusa G, Corradi L, Franceschini E, Meschiari M, Puzzolante C, Gennari W, Pecorari M, Guaraldi G, Mussini C. Pneumocystosis as a complication of H1N1 influenza A infection in an HIV-positive patient on effective cART. *Open Forum Infect Dis* 2019;6:ofz105.

10. Burke J, Soubani AO. Influenza and Pneumocystis jirovecii pneumonia in an allogeneic hematopoietic stem cell transplantation recipient: Coinfection or superinfection? *Transpl Infect Dis* 2018;20: e12802.

11. Marwah V, Singh S, Govind R, Shergill S, Vasan A. Pneumocystis jiroveci

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

pneumonia and H1N1 co-infection in an immunocompetent patient. *Indian J Chest Dis Allied Sci* 2019;61:219-220.

12. Kim D, Quinn J, Pinsky B, Shah NH, Brown I. Rates of co-infection between SARS-CoV-2 and other respiratory pathogens. *JAMA* 2020. doi: 10.1001/jama.2020.6266 (In press).

13. Karageorgopoulos DE, Qu JM, Korbila IP, Zhu YG, Vasileiou VA, Falagas ME. Accuracy of beta-D-glucan for the diagnosis of Pneumocystis jirovecii pneumonia: a meta-analysis. *Clin Microbiol Infect* 2013;19:39-49.

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

Table 1. Clinical Laboratory Results.

Measure	Result	Reference
Hematology and Chemistry		
Hemoglobin (g/dL)	8.9	11.5-16.4
Hematocrit (%)	27.3	36-48
Leukocytes ($\times 10^3$ per μL)	15.2	4-10
Differential (%)		
Neutrophils	89.5	40-70
Lymphocytes	7.2	22-44
Monocytes	2.4	4-11
Eosinophils	0.0	0-8
Basophils	0.2	0-3
Platelets ($\times 10^3$ per μL)	562	150-450
Ferritin (µg/L)	54	13-150
Procalcitonin (ng/mL)	0.1	0.00-0.08
Lactate dehydrogenase (U/L)	348	135-225
Microbiology		
Respiratory culture (tracheal aspirate)	3+ neutrophils, negative Gram stain and no growth on culture	No growth
Blood culture	No growth	No growth
SARS-CoV-2 (COVID-19 PCR)	Positive	Negative
Influenza A and B PCR	Negative	Negative
Parainfluenza PCR	Negative	Negative
Adenovirus PCR	Negative	Negative
Respiratory syncytial virus PCR	Negative	Negative
Human metapneumovirus PCR	Negative	Negative
Rhinovirus PCR	Negative	Negative
S. pneumoniae urine antigen	Negative	Negative
Legionella urine antigen	Negative	Negative
Histoplasma urine antigen	Negative	Negative
Coccidioides urine antigen	Negative	Negative
Blastomyces urine antigen	Negative	Negative
Cryptococcal antigen	Negative	Negative
Galactomannan antigen	0.08	0-0.49

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

(1,3)-β-D-glucan (pg/mL)	305	<80
Pneumocystis jirovecii PCR	Positive	Negative

Measure	Result	Reference
Immunology		
CD4+ T lymphocytes (absolute)	291	441-2156

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE

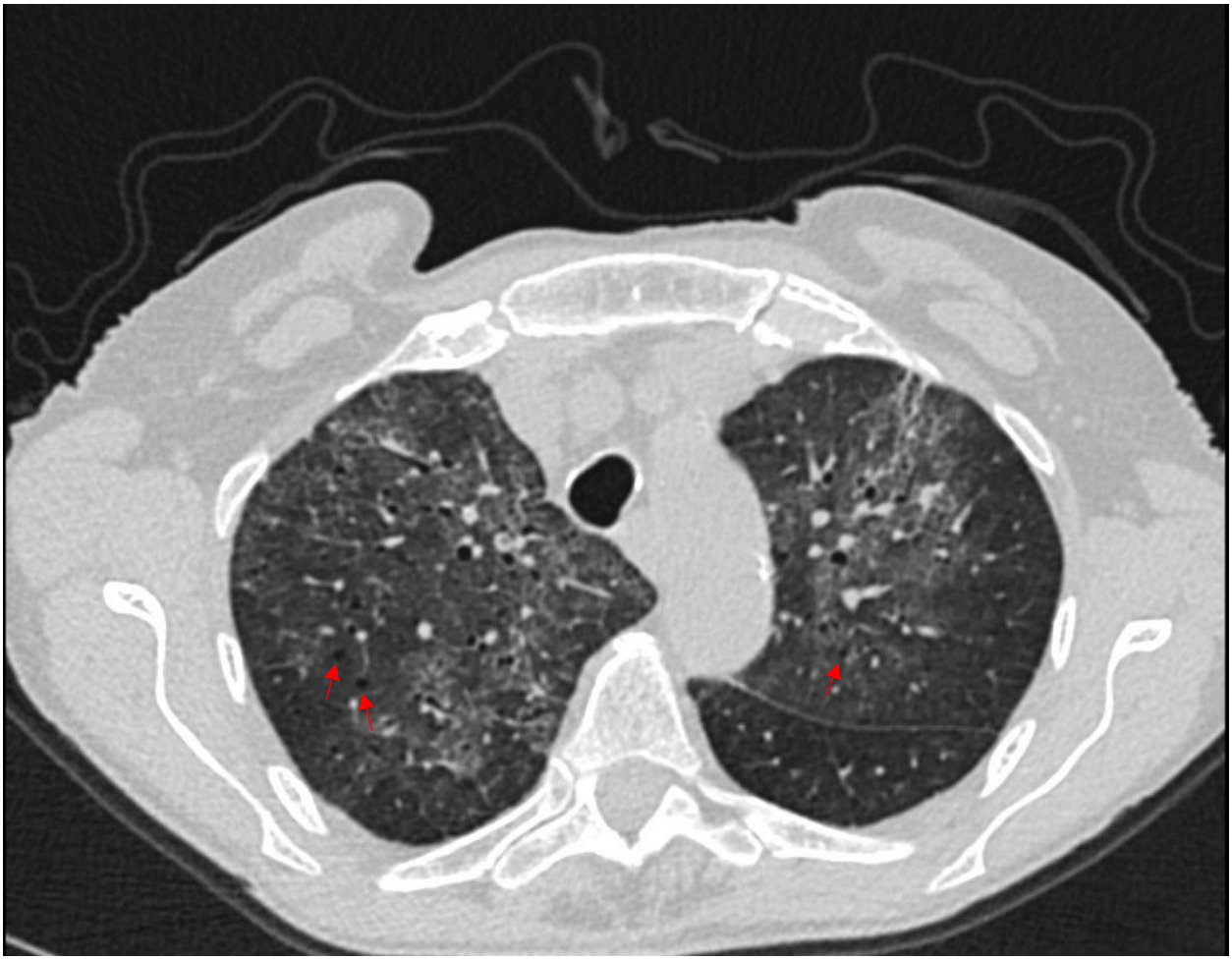

Figure 1. Chest Computed Tomographic (CT) Image. Representative axial image from the patient's chest CT scan. Red arrows indicate cystic changes.

AJRCCM Articles in Press. Published May 15, 2020 as 10.1164/rccm.202003-0766LE