Pulmonary Vascular Pathology in Covid-19

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infection (as determined by two consecutive negative PCR tests), were available for analysis (Fig. S1 in the Supplementary Appendix).

The group of persons with asymptomatic SARS-CoV-2 infection consisted of 58 passengers and 32 crew members, with median age of 59.5 years (interquartile range, 36 to 68; range, 9 to 77). A total of 24 of these persons (27%) had coexisting medical conditions, including hypertension (in 20%) and diabetes (9%). The first PCR test at the hospital was performed a mean of 6 days after the initial positive PCR test on the ship. The median number of days between the first positive PCR test (either on the ship or at the hospital) and the first of the two serial negative PCR tests was 9 days (interquartile range, 6 to 11; range, 3 to 21), and the cumulative percentages of persons with resolution of infection 8 and 15 days after the first positive PCR test were 48% and 90%, respectively. The risk of delayed resolution of infection increased with increasing age (mean delay in resolution for an increase in age from 36 to 68 years, 4.41 days; 95% CI, 2.28 to 6.53) (Fig. 1).

In this cohort, the majority of asymptomatically infected persons remained asymptomatic throughout the course of the infection. The time to the resolution of infection increased with increasing age.

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Disclosure forms provided by the authors are available with the full text of this letter at NEJM.org.

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Pulmonary Vascular Pathology in Covid-19

TO THE EDITOR: In their article, Ackermann et al. (online May 21; July 9 issue)[1] showed that vascular lesions in the lung — namely, sprouting and intussusceptive angiogenesis, along with disruption of intercellular junctions — were found in patients who died from coronavirus disease 2019 (Covid-19). The authors hypothesized that the mechanism responsible for the vasculopathy was a direct viral effect on endothelial cells or perivascular inflammation, but they did not comment on pericytes. Pericytes are perivascular cells that have a key role in the maintenance of microvessel integrity; their loss triggers both sprouting and intussusceptive angiogenesis.[2] Because they are among the cells that have the highest expression of angiotensin-converting enzyme 2 (ACE2), the receptor of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2),[3,4] pericytes could be a target of the virus. We recently showed a profound loss of pericytes coexisting with preserved endothelial cells in alveolar capillaries of the lungs of patients with Covid-19.[5] Could the authors comment on the possibility that alteration of pericytes by a direct effect of SARS-CoV-2 may be the initial trigger of the microvasculopathy?

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TO THE EDITOR: Ackermann et al. observed widespread vascular abnormalities in autopsy specimens from patients with Covid-19 pneumonia, including thrombosis, microangiopathy, and a higher degree of angiogenesis than was seen in patients with influenza pneumonia. These histologic findings suggest possible mechanisms for the pulmonary vascular abnormalities on computed tomographic (CT) pulmonary angiography that we recently described in patients with Covid-19 pneumonia.[1]

Although focal vessel enlargement within ground-glass opacities was described in early imaging investigations of Covid-19,[2] we have noted additional extensive vascular abnormalities, including regionally dilated segmental and subsegmental pulmonary vessels, increased branching and tortuosity of pulmonary vasculature, and perfusion abnormalities on dual-energy CT (Fig. 1). As previously suggested,[1] vascular injury leading to abnormal vasoregulation and vascular shunting may be one explanation for the presence of hypoxemia that is disproportionate to the reduction in pulmonary compliance in some patients with Covid-19 pneumonia.[3]

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Dr. Little reports being an academic author and textbook associate editor for Elsevier, and receives royalties for his work. No other potential conflict of interest relevant to this letter was reported.

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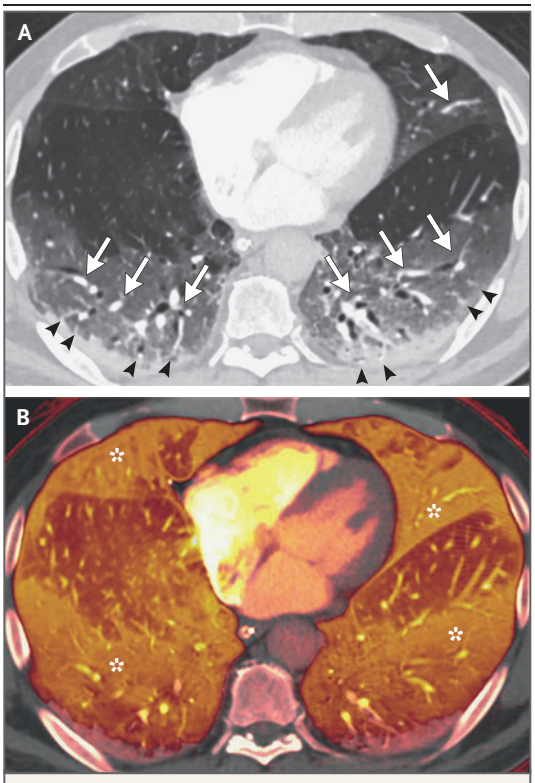

Figure 1. A 65-Year-Old Man with Covid-19 Pneumonia and Acute Respiratory Distress Syndrome.

Panel A is an axial CT image obtained at lung-window settings, showing large regions of ground-glass opacity in the lower lungs bilaterally; a pronounced mosaic perfusion pattern is present, in the absence of any visible pulmonary emboli. Subsegmental pulmonary vessels are dilated within areas of ground-glass opacity (arrows), whereas vessels in areas of clear lung are smaller in diameter. The distal subsegmental pulmonary vessels are engorged and tortuous (arrowheads). Panel B is a corresponding axial dual-energy CT image showing heterogeneous distribution of iodine in the lungs, suggesting higher perfusion of the areas of ground-glass opacity containing enlarged vessels (asterisks) and lower perfusion of the clear regions. Decreased perfusion in the posterior lungs corresponds to a small amount of consolidation seen in Panel A.

TO THE EDITOR: Ackermann et al. published an image obtained by transmission electron microscopy that depicted ultrastructural features of endothelial cell destruction. We measured the diameters of the two structures pinpointed in their Figure 3D as 250±7 nm in one and 264±13 nm in the other (mean [±SD] of four measurements). We also conducted a meta-analysis of published studies of SARS-CoV-2 (Fig. 1), in which we found a mean virion size of 60±9 nm in images from thin-section transmission electron microscopy. The two dark spots in the image provided by

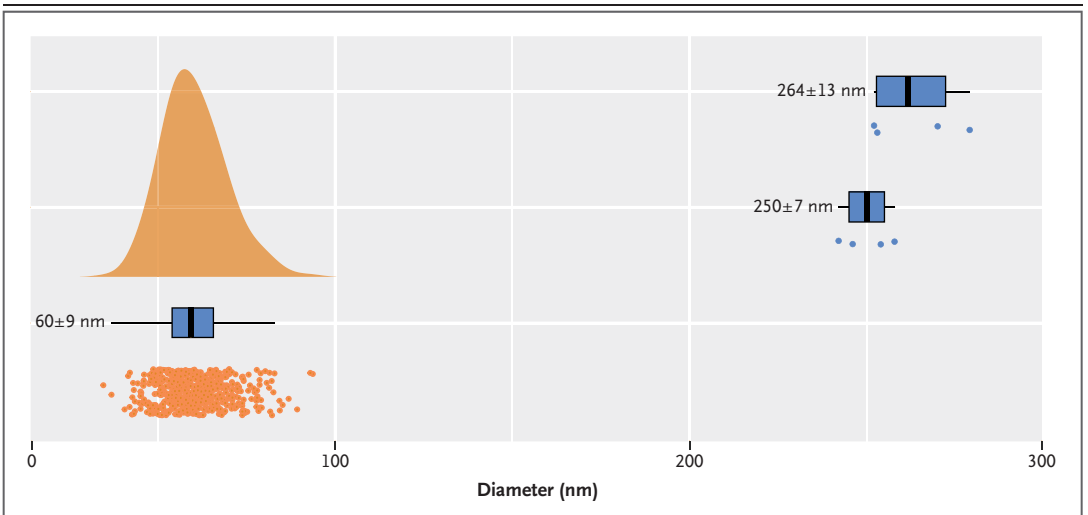

Figure 1. Comparison of SARS-CoV-2 Virion Sizes.

Shown is a comparison of virion size based on our meta-analysis of images from thin-section transmission electron microscopy from 12 sources (published studies as well as images from the Chinese National Institute for Viral Disease Control and Prevention and the University of Hong Kong) (mean [±SD] diameter, 60±9 nm) and the sizes of the two structures referred to as SARS-CoV-2 in Figure 3D of the article by Ackermann et al. (mean diameters from four measurements, 250±7 nm and 264±13 nm). In the box-and-whisker plots, the bold vertical line indicates the median, the left and right sides of the box the interquartile range, and the whiskers the range.

Ackermann et al. are thus approximately 4 times as large as expected for SARS-CoV-2 virions, and they are not located within a cell membrane as described. They also lack viral nucleocapsids and are not within vesicles. Since the possibility of confusing SARS-CoV-2 with other subcellular structures is known,[1,2] we ask the authors whether they have other data to support the identification of the particles as SARS-CoV-2.

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THE AUTHORS REPLY: We share the interest of Burel-Vandenbos et al. in the potential involvement of pericytes in the pathobiology of Covid-19. Pericytes have been implicated in several processes relevant to Covid-19, including blood-flow regulation and angiogenesis.[1] The experimental challenge is the rarity of pericytes in the lungs and the lack of exclusive markers for reproducible histologic identification. The α smooth-muscle actin (α-SMA)–positive cells in patients with Covid-19 may reflect not only pericytes but also parenchymal[2] and mesothelial-derived[3] myofibroblasts, which have previously been implicated in lung repair, remodeling, and intussusceptive angiogenesis.

Som et al. show striking changes in the lung of a patient with Covid-19. In their previous correspondence,[4] the authors showed obstructed airspaces and associated vasoconstriction consistent with intact vasoregulation. Here, the images show hypoperfusion of apparently unobstructed airways. Potentially catastrophic for this patient, these findings are consistent with our observations of vasodysregulation in patients with Covid-19. We note that the marked vasoconstriction and vascular tortuosity described

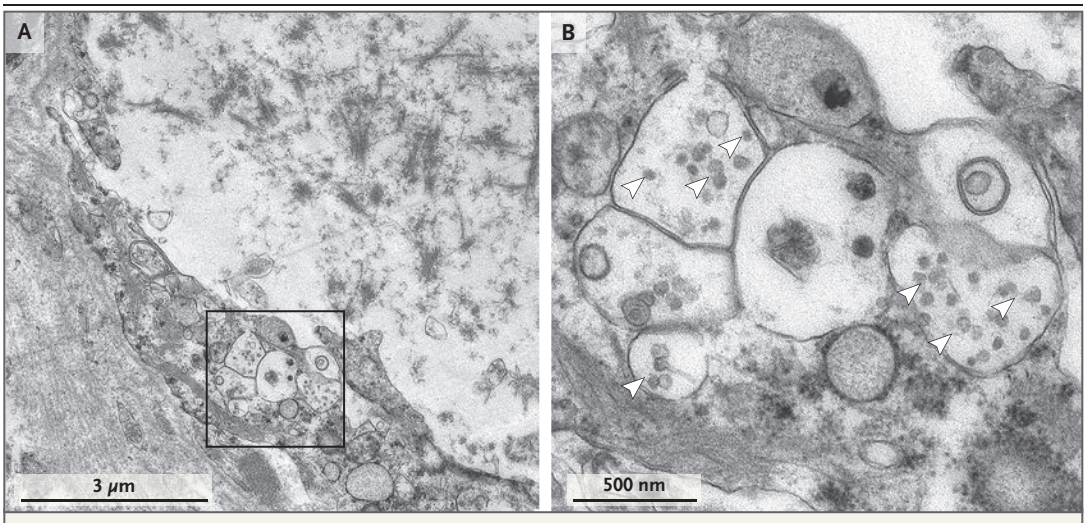

Figure 1. Viruslike Particles within the Endothelial Cells of a Lung from a Patient with Covid-19.

Shown is a transmission electron micrograph of viruslike particles within the endothelial cells of a lung (after aldehyde fixation) from a patient with Covid-19. In Panel B (an enlarged view of the boxed area in Panel A), numerous particles in the size range of 60 nm to 150 nm are visible (arrowheads). (Images prepared in collaboration with G. Griffiths, J. Wohlmann, and N. Roos, University of Oslo.)

by Som et al. are illustrated by the microCT three-dimensional reconstructions included in the Supplementary Appendix of our article (available at NEJM.org).

Scholkmann and Nicholls remind us of the potential variability of calibration in transmission electron microscopy and of the care necessary in interpreting absolute size in planar transmission electron micrographs — especially when comparing images of RNA viruses obtained in glutaraldehyde-fixed tissue and tissue culture. The image in our article was chosen to highlight the dramatic ultrastructural changes in the endothelium. In Figure 1, a transmission electron micrograph of the endothelium from a patient with Covid-19, we show numerous endothelial viruslike particles, ranging in size from 60 nm to 150 nm — that is, a size range consistent with SARS-CoV-2 virus particles. To confirm the presence of SARS-CoV-2 virus in the lungs in our study, we measured viral RNAs in the three study groups and found high levels of SARS-CoV-2 RNA (Table S1 in the Supplementary Appendix of this letter, available at NEJM.org).

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Since publication of their article, the authors report no further potential conflict of interest.

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