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Kidney Biopsy Findings in Patients with COVID-19

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ABSTRACT

Background Coronavirus disease 2019 (COVID-19) is thought to cause kidney injury by a variety of mechanisms. To date, pathologic analyses have been limited to patient reports and autopsy series.

Methods We evaluated biopsy samples of native and allograft kidneys from patients with COVID-19 at a single center in New York City between March and June of 2020. We also used immunohistochemistry, *in situ* hybridization, and electron microscopy to examine this tissue for presence of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2).

Results The study group included 17 patients with COVID-19 (12 men, 12 black; median age of 54 years). Sixteen patients had comorbidities, including hypertension, obesity, diabetes, malignancy, or a kidney or heart allograft. Nine patients developed COVID-19 pneumonia. Fifteen patients (88%) presented with AKI; nine had nephrotic-range proteinuria. Among 14 patients with a native kidney biopsy, 5 were diagnosed with collapsing glomerulopathy, 1 was diagnosed with minimal change disease, 2 were diagnosed with membranous glomerulopathy, 1 was diagnosed with crescentic transformation of lupus nephritis, 1 was diagnosed with anti-GBM nephritis, and 4 were diagnosed with isolated acute tubular injury. The three allograft specimens showed grade 2A acute T cell–mediated rejection, cortical infarction, or acute tubular injury. Genotyping of three patients with collapsing glomerulopathy and the patient with minimal change disease revealed that all four patients had *APOL1* high-risk gene variants. We found no definitive evidence of SARS-CoV-2 in kidney cells. Biopsy diagnosis informed treatment and prognosis in all patients.

Conclusions Patients with COVID-19 develop a wide spectrum of glomerular and tubular diseases. Our findings provide evidence against direct viral infection of the kidneys as the major pathomechanism for COVID-19–related kidney injury and implicate cytokine-mediated effects and heightened adaptive immune responses.

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As coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) spreads worldwide, there is growing recognition of potential renal dysfunction in SARS-CoV-2–infected patients.[1–3] Proposed mechanisms of kidney injury range from direct viral infection to effects on the renin-angiotensin-aldosterone system, hemodynamic instability, coagulopathy, and cytokine storm.[4] Correlation with pathologic changes is needed to inform mechanistic hypotheses. To date, most descriptions of kidney pathology in SARS-CoV-2–infected patients are autopsy based and limited by autolysis[5,6] or consist of patient reports of COVID-19–associated collapsing glomerulopathy.[7–10] There is urgent need for biopsy-based series to elucidate the spectrum of kidney pathology in patients with COVID-19 and AKI or proteinuria. Herein, we provide a large kidney biopsy series of SARS-CoV-2–infected patients from the pandemic's New York epicenter.

METHODS

All kidney biopsies from SARS-CoV-2–infected patients accessioned by the Columbia University Irving Medical Center Renal Pathology Laboratory from March 13 to June 1, 2020 were identified. In total, there were 14 native kidney biopsies (including 1 previously published)[10]and 3 kidney allograft specimens. Biopsies originated from patients in six states: New York (5), New Jersey (5),

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Connecticut (3), Pennsylvania (2), Delaware (1), and Indiana (1). Clinical, laboratory, and follow-up data were provided by the submitting nephrologists. Indications for kidney biopsy were recorded as any combination of AKI, AKI superimposed on CKD, nephrotic-range proteinuria, or nephrotic syndrome, as previously described.[11] All kidney biopsies were processed by standard techniques for light microscopy, immunofluorescence, and electron microscopy. A directed search for virions was performed by systematic high-power ultrastructural examination of tubular epithelial and glomerular cells.

Immunohistochemical stain for the S2 subunit for SARS-CoV-2 spike protein was performed on formalin-fixed, paraffin-embedded (FFPE) tissue sections using mouse monoclonal IgG1 antibody from clone 1A9 (catalog no. GTX632604; GeneTex, Irvine, CA). Immunohistochemical stain for nucleocapsid protein was performed on FFPE tissue sections using rabbit monoclonal antibody from clone 001 (catalog no. 40143-R001; Sino Biologic, Beijing, People's Republic of China).

In situ hybridization (ISH) for SARS-CoV-2 RNA was performed manually on FFPE tissue sections using the chromogenic RNAscope 2.5 HD Reagent Kit-RED (catalog no. 322350; Advanced Cell Diagnostics, Newark, CA) and the RNAscope 2.5 HD Duplex Reagent Kit (catalog no. 322430; Advanced Cell Diagnostics) according to the manufacturer's protocols.[12] Two probes specific to the SARS-CoV-2 RNA encoding the spike protein were used, one in each chromogenic channel: V-nCoV2019-S (catalog no. 848561) and V-nCoV2019-S-C2 (catalog no. 848561-C2).[12] ISH by manual RNAscope was performed first in COVID-19–infected lung as positive tissue control and then, in 16 kidneys from COVID-19 biopsies and 2 kidneys from COVID-19–negative controls. The integrity of tissue RNA was demonstrated by *in situ* detection of the proximal tubule anchor gene, *LRP2* (megalin). ISH was also performed by automated platform using a single probe to SARS-CoV-2 RNA-encoding spike protein (RNAscope 2.5 LS Probe V-CoV2019-S, catalog no. 848568; Advanced Cell Diagnostics).

In three patients with collapsing glomerulopathy and one patient with minimal change disease, DNA was extracted from the FFPE kidney tissue, and *APOL1* genotyping was performed by Sanger sequencing of PCR fragments encompassing the *APOL1* risk alleles, as previously described.[10,13]

This study was approved by the institutional review board of Columbia University Irving Medical Center (protocol no. AAAT0009 [M00Y01]).

RESULTS

The study group of 17 patients included 12 men, with median age of 54 years (range, 22–72 years) (Tables 1 and 2). Racial demographics included 12 blacks, 3 whites, 1 Asian, and 1 Hispanic. Fourteen patients underwent native kidney biopsies, and three had allograft specimens (including two biopsies and one allograft nephrectomy). Sixteen patients had one or more comorbidities, including 11 with hypertension, 8 with obesity, 3 with diabetes, 3 with history of malignancy (2 prostate and 1 cervix), 4 with solid organ transplants (3 kidney and 1 heart), 1 with SLE, and 1 with untreated hepatitis C infection. Two were former smokers, but none had known preexisting lung disease. In addition to four patients with solid organ transplants, one was receiving immunosuppression for SLE.

Eight patients had mild COVID-19 without pneumonia, including one asymptomatic patient and another with predominantly gastrointestinal symptoms. The other nine had imaging-confirmed COVID-19 pneumonia, including two immunosuppressed patients (with SLE and kidney transplant) who required intubation. Baseline serum creatinine was normal in eight patients. Fifteen patients (88%) presented with AKI, including four with AKI superimposed on CKD. Nine patients (53%) had nephrotic-range proteinuria, including six (35%) with new-onset nephrotic syndrome.

Significance Statement

The mechanisms underlying coronavirus disease 2019 (COVID-19)–associated kidney injury are unknown, and morphologic correlates are few and limited to patient reports or autopsy series. The authors' evaluation of a biopsy series of 14 native and 3 allograft kidneys from patients with COVID-19 who developed AKI or nephrotic-range proteinuria found diverse glomerular and tubular diseases. These included collapsing glomerulopathy and minimal change disease (both of which occurred in patients with high-risk *APOL1* gene variants), membranous glomerulopathy, anti-GBM nephritis, acute tubular injury, exacerbation of preexisting autoimmune GN, and allograft rejection. They found no definitive evidence of SARS-CoV-2 in the samples by *in situ* hybridization, immunohistochemistry and electron microscopy, arguing against direct viral infection of the kidney as the major pathomechanism. Instead, the findings implicate cytokine-mediated effects and heightened adaptive immune responses. The kidney biopsy findings informed treatment and prognosis.

At presentation, the study group had median serum creatinine of 5.7 mg/dl (range, 0.8–20 mg/dl), including seven patients who required dialysis. Median urine protein-creatinine ratio was 7.8 g/g (range, 0.2–21 g/g), and median serum albumin was 2.9 g/dl (range, <1.5–4.5 g/dl). Six patients had microhematuria (more than five red blood cells per high-power field), including two with indwelling urinary catheters. Four patients had peripheral leukocytosis (including two with concurrent lymphopenia), and three had leukopenia. Positive serologies included two patients with antinuclear antibodies (including one with double-stranded DNA antibody), one patient with antiglomerular basement membrane (anti-GBM) antibody, and one with hepatitis C antibody. Inflammatory markers were abnormal in all patients tested, including elevated ferritin (n=13), C-reactive protein (n=10), erythrocyte sedimentation rate (n=9), lactate dehydrogenase (n=8), IL-6 (n=5), and IL-2 receptor (n=1).

Pathologic evaluation revealed five patients with collapsing glomerulopathy accompanied by acute tubular

Table 1. Clinical findings in patients with COVID-19 who underwent kidney biopsy

Pt	Age	Sex	Race	Comorbidities	IS	Temperature, °C	SpO_2 on RA	BP	Edema	COVID-19 Manifestations	Renal Presentation/ Biopsy Indications
1	46	M	B	OSA, obesity (BMI=44)	N	37.1	94	144/100	Y	Cough, fever, sore throat	AKI, NS
2	62	M	B	HTN, prostate carcinoma	N	37.2	98	126/79	Y	Fever, myalgia, weakness	AKI on CKD, NS
3	62	M	B	HTN, DM, prostate carcinoma	N	36.8	91	122/82	N	Fever, hypoxemia, bilateral perihilar and basilar infiltrates on CXR	AKI, NRP
4	57	M	B	HTN, untreated hepatitis C virus	N	38.1	97	173/92	N	Flu-like symptoms with ground glass opacities and patchy consolidation on CXR	AKI on CKD, NRP
5	61	M	B	HTN, obesity (BMI=31.1)	N	38.5	99	134/79	N	Cough, fever	AKI, NRP
6	25	M	B	Obesity (BMI=32.2)	N	38.5	95	117/79	Y	Cough, fever, myalgia, infiltrates on CXR	AKI, NS
7	43	F	B	DM, HLD, streptococcal infection, obesity (BMI=52.5)	N	37.6	96	107/67	N	Cough, fever, sore throat, weakness, patchy LUL infiltrates on CXR	AKI
8	28	M	B	None	N	38.6	96 (on O_2)	143/62	N	Cough, fever, hypoxemia, bilateral infiltrates on CXR, elevated troponin and CPK	AKI
9	67	M	W	HTN, gout, history of tobacco use, obesity (BMI=34.9)	N	36.5	95	135/75	N	Cough, diarrhea, lethargy, multifocal infiltrates on CXR	AKI on CKD
10	51	M	B	HTN, DSA+ OHTx for NICM 1 yr ago, atrial fibrillation, CVA, BPH, HLD	Y	36.8	96	117/81	N	Predominantly nausea, vomiting, abdominal pain, with cough	AKI on CKD
11	72	M	W	HTN, DM, HLD, gout, spinal stenosis, atrial fibrillation	N	36.8	97 (on O_2)	125/73	Y	Cough, pleural effusion on CT	NS
12	70	F	B	HTN, CAD, PVD, cervical carcinoma, GERD, HLD, obesity (BMI=39.4)	N	38.1	92	118/58	Y	Cough, fever, shortness of breath	AKI, NRP
13	27	F	A	SLE with class 2 lupus nephritis	Y	36.9	93	130/80	Y	Cough, fever, shortness of breath, hypoxemia, bilateral infiltrates on CXR, required intubation and ICU admission	AKI, NS
14	48	F	B	GERD, history of tobacco use, obesity	N	37.6	98	185/80	N	Cough, myalgia, infiltrates on CXR	AKI
15	54	M	W	ESKD secondary to IgAN s/p DSA+ LURTx 1 mo ago, HTN, obesity (BMI=30.7)	Y	36.8	99	116/75	N	Asymptomatic	AKI
16	22	M	B	ESKD likely secondary to PLA2R+ MGN s/p DDRTx 2 years ago, HTN	Y	37.7	95	178/127	N	Cough, fever, bilateral infiltrates on CXR, required intubation and ICU admission	CKD
17	54	F	H	ESKD secondary to PCKD s/p DDRTx 2 months ago, HTN	Y	98.8	100	104/70	N	Fever, dry throat	AKI

BP is in millimeters of Hg. Pt, patient; IS, immunosuppression at presentation; SpO_2, oxygen saturation (percentage); RA, room air; M, man; B, black; OSA, obstructive sleep apnea; BMI, body mass index; N, no; Y, yes; NS, nephrotic syndrome; HTN, hypertension; AKI on CKD, AKI superimposed on CKD; DM, diabetes mellitus; CXR, chest x-ray; NRP, nephrotic-range proteinuria; F, woman; HLD, hyperlipidemia; LUL, left upper lobe; O_2, oxygen; CPK, creatine phosphokinase; W, white; DSA, donor-specific antibody; OHTx, orthotopic heart transplantation; NICM, nonischemic cardiomyopathy; CVA, cerebrovascular accident; BPH, benign prostatic hyperplasia; CT, computed tomography; CAD, coronary artery disease; PVD, peripheral vascular disease; GERD, gastroesophageal reflux disease; A, Asian; ICU, intensive care unit; IgAN, IgA nephropathy; s/p, status post; LURTx, living unrelated renal transplantation; MGN, membranous glomerulopathy; DDRTx, deceased donor renal transplantation; H, Hispanic; PCKD, polycystic kidney disease.

Table 2. Laboratory findings and follow-up information in patients with COVID-19 who underwent kidney biopsy

Pt	Baseline sCr	Renal Function at Presentation					Complete Blood Counts			Other Laboratory Tests	Follow-Up Information		
		sCr	Dialysis	Urine Prot	sAlb	Urine RBC	Hgb	WBC	Plt		Duration, d	Therapy	Outcome
1	1.1	12.5	Y	5.8	3.1	2	16.6	7.7	242	LDH 1504 U/L, ESR>130 mm/h, hsCRP 49.4 mg/L ferritin 1147 ng/ml, IL-6 12 pg/ml, IL-2R 1530 pg/ml	24	Tocilizumab, steroids	Dialysis dependent with sCr 14.2 mg/dl, UPCR 10.2 g/g, repeat COVID-19 negative
2	2	10.7	N	12.1	3.1	0–5	14.2	7.1 (with lymphopenia)	235	ESR 125 mm/h, ferritin 340.3 ng/ml	30	None	sCr 3.8 mg/dl, UACR 5.5 g/g, repeat COVID-19 negative
3	1	11.6	N	19	2.4		11.7	14.4 (with lymphopenia)	355	ESR 130 mm/h, CRP 232 mg/L, ferritin 5000 ng/ml	12	Steroids, hydroxychloroquine, ceftriaxone, doxycycline	sCr 2.3 mg/dl, UPCR 4 g/g, decreased ferritin, ESR, IL-6
4	1.1	4.9	N	6.2	2.5	0	9.8	7	257	elevated ESR, CRP 11.1 mg/L, ferritin 907 ng/ml, elevated IL-6 and D-dimer, CK 3200 U/L, +HCV Ab	11	Azithromycin, ceftriaxone	sCr 4.9 mg/dl, UPCR 4.1 g/g, repeat COVID-19 negative
5	Normal	15	Y	9	2.5		8.3	21	368	ESR 41 mm/h, CRP 229 mg/L, ferritin 2542 ng/ml	5		Dialysis dependent, repeat COVID-19 negative
6	Normal	2.2	N	21	<1.5	3–5	13.9	8.5	408	LDH 590 U/L, CRP 33.4 mg/L, ferritin 374 ng/ml	27	Azithromycin, hydroxychloroquine, steroids	sCr 0.8 mg/dl, UPCR 4.3 g/g, decreased CRP
7	3.5 (at adm)	6.7	Y	1+ on UA		21–50	11.4	14	250	None			
8	2 (at adm)	9	Y	100 mg/dl on UA	2.6	0	10.6	13.1 (with lymphopenia)	320	LDH 3075 U/L, CRP 4.7 mg/L, ferritin >7500 ng/ml, CK 3309 U/L	<30	Tocilizumab, hydroxychloroquine, ceftriaxone, azithromycin	sCr 1.2 mg/dl
9		5.7	Y	300 mg/dl on UA	2.7	>5	12.8	9.2 (with lymphopenia)	454	LDH 309 U/L, CRP 13 mg/L, ESR>100 mm/h, ferritin 924 ng/ml, CK 128 U/L	30	Tocilizumab, hydroxychloroquine, ceftriaxone, azithromycin	Dialysis dependent
10	1.5–1.8	4.8	N	0.5	4.2	3	11.5	3.2	107	LDH 524 U/L, CRP 50.8 mg/L, ESR 33 mm/h, ferritin 2282 ng/ml, IL-6 21.4 pg/ml, +ANA	1	Hydroxychloroquine	sCr 2.5 mg/dl, urine prot 100 mg/dl on UA, repeat COVID-19 positive

Table 2. Continued

Pt	Baseline sCr	Renal Function at Presentation					Complete Blood Counts			Other Laboratory Tests	Follow-Up Information		
		sCr	Dialysis	Urine Prot	sAlb	Urine RBC	Hgb	WBC	Plt		Duration, d	Therapy	Outcome
11	Normal	0.8	N	8.8	1.7	6–15	12.3	14.8	283	LDH 300 U/L, CRP 153 mg/L, ferritin 397 ng/ml	18	Tacrolimus	sCr 1.3 mg/dl, repeat COVID-19 positive
12		2.9	N	6.8	3.0	Present	9	6.1	183	LDH 1019 U/L, ESR 127 mm/h, ferritin 635 ng/ml, +ANA, +anti-dsDNA Ab	35	None	sCr 2.4 mg/dl, UPCR 5–6 g/g, improved edema
13	0.9	2.5	N	9.2	2.0	20–50	8	6.8	182	IL-6 79 pg/ml	6	Steroids	Died from multiorgan failure secondary to COVID-19
14	0.9	20	Y	>300 mg/dl on UA	3.1	>182, with RBC casts	9.5	9.1	167	CRP 143 mg/L, CK 1460 U/L, +anti-GBM Ab	16	PLEX, steroids, cyclophosphamide	Dialysis dependent with sCr 6.7 mg/dl, repeat COVID-19 negative
15	1.7	2.6	N	0.2	4.5	60	8.1	3.9	211	IL-6 6.6 pg/ml	55	Tocilizumab, IVIG, steroids, thymoglobulin	sCr 2 mg/dl, UPCR 0.1 g/g, repeat COVID-19 negative
16	Dialysis dependent	9.4	Y		3.4		4.4	6.1	202	LDH 333 U/L, ESR 79 mm/h, CRP 195.6 mg/L, ferritin 1630 ng/ml	4	Tocilizumab, hydroxychloroquine, piperacillin-tazobactam, azithromycin	Dialysis dependent with sCr 6.7 mg/dl
17	2.5	2.9	N	0.2	4.4	3	10.1	3.3	286	CRP 0.49 ng/ml, ferritin 1677 ng/ml (previously)	7	None	sCr 2.2 mg/dl

Pt, patient; sCr, serum creatinine (milligrams per deciliter); urine prot, urine protein-creatinine ratio or 24-h urine protein; sAlb, serum albumin (grams per deciliter); RBC, red blood cell (per high-power field); Hgb, hemoglobin (grams per deciliter); WBC, white blood cell count (10^3 per microliter); Plt, platelet (10^3 per microliter); Y, yes; LDH, lactate dehydrogenase; ESR, erythrocyte sedimentation rate; hsCRP, high-sensitivity C-reactive protein; IL-2R, IL-2 receptor; UPCR, urine protein-creatinine ratio; repeat COVID-19, repeat PCR testing for severe acute respiratory syndrome coronavirus 2; N, no; UACR, urine microalbumin-creatinine ratio; CRP, C-reactive protein; CK, creatine kinase; +, positive; HCV Ab, Ab for hepatitis C virus; adm, admission; UA, urinalysis; ANA, antinuclear antibody; dsDNA, double-stranded DNA; PLEX, plasmapheresis; IVIG, intravenous Ig.

injury (ATI), including one with tubuloreticular inclusions (TRIs) (Figure 1, Table 3). The three patients with collapsing glomerulopathy who consented to genetic studies all had high-risk *APOL1* genotypes (two with G1/G1 and one with G1/G2). One patient had minimal change disease accompanied by ATI and endothelial TRI; this patient also had high-risk *APOL1* genotype (G1/G1). Among these six podocytopathies, three (50%) did not have imaging-confirmed pneumonia. In four patients, the predominant finding was ATI, including one with pigment casts suggesting myoglobinuria. Four patients had immune-mediated glomerular diseases, including two with membranous glomerulopathy (one with positive tissue staining for phospholipase A2 receptor [PLA2R]), one with crescentic lupus nephritis class 4+5, and one with anti-GBM nephritis. The three kidney transplant recipients had grade 2A acute T cell–mediated rejection, cortical infarction, and ATI, respectively.

Electron microscopy was performed on 13 biopsies, including 10 with glomeruli available. Ultrastructural examination demonstrated glomerular endothelial TRI in 6 of 10 cases (60%) and absence of definitive virions within renal cells in all 13 cases. Immunohistochemical stains for the spike and nucleocapsid proteins and automated ISH for SARS-CoV-2 RNA, performed in 16 cases, showed no definitive staining. ISH for SARS-CoV-2 RNA performed manually by RNAscope revealed rare, possibly positive tubular cell staining in 2 of 16 patients, both with diagnosis of ATI (Supplemental Figure 1).

Short-term follow-up was available in 16 patients (median, 16 days; range, 1–55 days) (Table 2). One patient died of multiorgan failure 6 days after biopsy. Three patients received no treatment. Nine patients received treatment directed to COVID-19 (including tocilizumab in five, hydroxychloroquine in six, and azithromycin in five). Apart from RRT, seven received specific treatments for their kidney disease (including steroids in five; tacrolimus in one; and plasmapheresis, steroids, and cyclophosphamide in one). At last follow-up, the study group had median serum creatinine of 2.4 mg/dl (range, 0.8–14.2 mg/dl), including ten patients with decrease in serum creatinine and five who remained dialysis dependent. Among five with collapsing glomerulopathy, one with minimal change disease, and two with membranous glomerulopathy, repeat urine protein level was available in six, of which five had reductions in proteinuria. Repeat inflammatory markers decreased in two patients with available data. Repeat PCR testing for SARS-CoV-2 by nasopharyngeal swab, available in eight patients, converted to negative in six.

DISCUSSION

Impaired renal function is a common complication affecting 5%–37% of hospitalized patients with COVID-19.[1–3] SARS-CoV-2 is postulated to cause AKI by diverse mechanisms, including interaction with its cellular receptor angiotensin-converting enzyme 2 (ACE2), viral immune responses, cytokine storm, hypoxemia, reduced oral intake, circulatory collapse, prothrombotic effects, and multiorgan dysfunction.[14–16] Specifically, SARS-CoV-2 may directly infect the kidney *via* ACE2 widely expressed in proximal tubular cells and podocytes.[5,6,17] Imbalance of the renin-angiotensin-aldosterone system *via* ACE2 also could exert deleterious hemodynamic effects.[15] Viral-induced cytokine storm causes massive release of granulocyte colony–stimulating factor, various interleukins, and IFN,[18–20] which can injure the kidney directly or indirectly *via* effects on other organs, such as heart and skeletal muscle. To date, pathologic analyses of the kidney are few and limited to single patient reports or autopsy series.[5–10] We provide a biopsy-based single-center series exploring kidney pathology in SARS-CoV-2–infected patients.

SARS-CoV-2–infected patients developed diverse glomerular and tubular diseases. The most common glomerular disorder was podocytopathy, including five patients with collapsing glomerulopathy and one patient with minimal change disease; all occurred in black patients (including four with documented *APOL1* high-risk genotype) and presented with nephrotic syndrome or nephrotic-range proteinuria and AKI with associated ATI.[10] These findings enlarge the literature on collapsing glomerulopathy[8–10] in the setting of COVID-19 and provide the first example of minimal change disease with *APOL1* high-risk genotype. Given the closely related association between IFN therapy and both collapsing glomerulopathy and minimal change disease[21,22] as well as the presence of TRI (so-called IFN footprints), the findings support a role for cytokine-mediated podocyte injury in genetically susceptible individuals with SARS-CoV-2 infection.[19] The lack of demonstrable viral particles in the podocytes by all ancillary studies argues against direct glomerular viral infection.

The inflammatory milieu surrounding COVID-19 also may trigger or exacerbate immune-mediated diseases in predisposed patients. Examples include the crescentic transformation of longstanding preexisting class 2 lupus nephritis and development of acute T cell–mediated rejection in a patient with preformed donor-specific antibodies. IFN and granulocyte colony–stimulating factor play an important role in triggering acute rejection[23] or exacerbating immune complex–mediated GN,[24] and both cytokines are known to be elevated in patients with COVID-19.[18–20]

Other glomerular diseases included new onset of anti-GBM nephritis in one patient and membranous glomerulopathy of uncertain duration in two. Pulmonary injury from influenza or other insults has been postulated to precede onset of anti-GBM nephritis by exposing the cryptic target Goodpasture antigen,[25] consisting of distinct epitopes in COL4A3 and COL4A5, in damaged alveolar capillary basement membranes.[26] Conceivably, COVID-19 pneumonia could play a similar priming role. The major target antigen in membranous glomerulopathy, PLA2R, is also expressed in the respiratory tract,[27] suggesting a potential source for antigen presentation to incite or potentiate

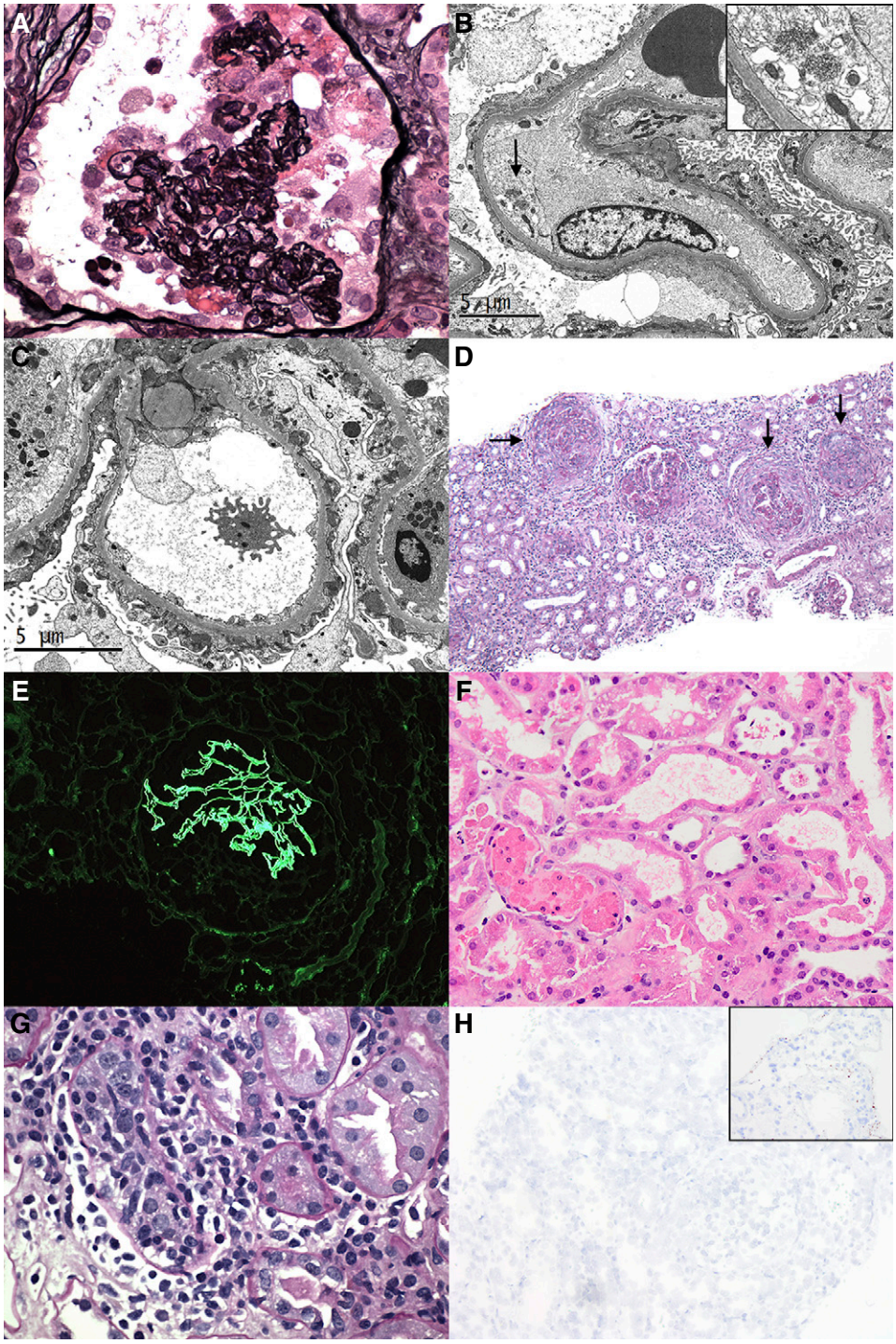

Figure 1. Kidney biopsy findings in patients with COVID-19. (A) Light microscopy demonstrates a lesion of collapsing glomerulopathy characterized by hyperplasia of glomerular epithelial cells and collapse of the underlying glomerular capillaries. Jones methanamine silver. Magnification, ×600. (B) Diffuse foot process effacement and endothelial TRIs (arrow and inset) in a patient with minimal change disease. Electron micrograph. Magnification, ×8000. (C) Subepithelial electron dense deposits in PLA2R-associated membranous glomerulopathy. Electron micrograph. Magnification, ×15,000. (D) Multiple glomeruli with circumferential cellular crescents (arrows) in a patient with class 4+5 lupus nephritis. Periodic acid–Schiff. Magnification, ×100. (E) A glomerulus compressed by a crescent with global linear GBM staining for IgG in a patient with anti-GBM nephritis. Immunofluorescence for IgG. Magnification, ×400. (F) Tubular simplification and focal shedding of degenerating epithelial cells into the tubular lumina in a patient with isolated ATI. Hematoxylin and eosin. Magnification, ×400. (G) Severe lymphocytic tubulitis in a patient with acute T cell–mediated rejection. Periodic acid–Schiff. Magnification, ×600. (H) ISH for the virus by automated method showing undetectable viral RNA in the kidney (inset shows positive lung control). Automated ISH with hematoxylin counterstain. Magnification, ×400.

Table 3. Pathologic findings in patients with COVID-19 who underwent kidney biopsy

Pt	Diagnosis		Light Microscopy									Electron Microscopy		
	Diagnosis	Other Findings	No. Glom	No. GS	No. Collapse	No. Noncollapsed FSGS	Hypercellularity	Microcysts	II	IFTA	VS	FPE	TRI	Viral Particles
1	Collapsing FSGS	ATI	20	0	14	0	N	Y	Focal	Mild	Mild	NA[a]	NA[a]	N
2	Collapsing FSGS	ATI	8	3	2	0	N	Y	Focal	Moderate	Moderate to severe	100	Y	N
3	Collapsing FSGS	ATI	18	4	4	1	N	Y	None	Moderate	Moderate	30	N	N
4	Collapsing FSGS	ATI	10	1	5	0	N	N	Focal	Severe	Mild	NA	NA	NA
5	Collapsing FSGS	ATI	11	3	7	0	N	Y	Focal	Severe	Moderate to severe	90	N	N
6	MCD	ATI	17	0	0	0	N	Y	None	None	None	100	Y	N
7	ATI		15	0	0	0	N	N	Focal	None	Mild	15	Y	N
8	ATI	Pigment casts	22	0	0	0	N	N	None	None	Mild	10	Y	N
9	ATI		2	1	0	0	N	N	None	Mild	Moderate	NA[a]	NA[a]	N
10	ATI		8	0	0	0	N	N	None	None	Moderate	5	N	N
11	MGN	PLA2R stain positive[b]	15	3	0	4	N	N	Focal	Mild	Mild to moderate	100	N	N
12	MGN	PLA2R stain negative[b]	3	2	0	0	N	Y	Focal	Moderate	Moderate to severe	30	Y	N
13	LN class 4+5		35	9	0	0	Mes, Endo, Crescents	N	Diffuse	Mild	Mild	90	Y	N
14	Anti-GBM GN	ATI, RBC casts	32	1	0	0	Crescents	Y	Diffuse	Mild	Moderate	NA[a]	NA[a]	N
15	TCMR grade 2A		11	1	0	0	N	N	Focal	None	Mild	NA	NA	NA
16	Infarction[c]		NA	NA	NA	NA	NA	N	Focal	Severe	NA	NA	NA	NA
17	ATI		20	1	0	0	N	N	None	None	Mild	NA	NA	NA

Pt, patient; glom, No., number; glomerulus or glomeruli; GS, global sclerosis; II, interstitial inflammation; IFTA, tubular atrophy and interstitial fibrosis; VS, vascular sclerosis; FPE, foot process effacement (percentage); N, no; Y, yes; NA, not available; MCD, minimal change disease; LN, lupus nephritis; Mes, mesangial hypercellularity; Endo, endocapillary hypercellularity; RBC, red blood cell; TCMR, T cell–mediated rejection.

[a]All glomeruli were involved by crescents in the patient with anti-GBM GN, and no glomeruli were available for ultrastructural examination in two patients with native kidney biopsies.

[b]Performed by tissue staining.

[c]Nephrectomy specimen.

anti-PLA2R autoimmune responses. Coincidental associations with COVID-19 cannot be excluded.

The other major disease category was ATI identified in four native and two allograft kidneys, including one with infarction. Four patients had severe COVID-19 pneumonia (including three with hypoxemia), three patients were on immunosuppression as maintenance for solid organ transplants (two kidney and one heart), four patients had exposure to potentially nephrotoxic or nephromodulatory medications (three tacrolimus and one lisinopril), and one patient had rhabdomyolysis with pigment casts. None had evidence of thrombotic microangiopathy. ATI has also been identified as the predominant finding in autopsy series.[6] Etiology is likely to be multifactorial with complex interplay of sepsis, hypoxia, hemodynamic instability, nephrotoxin exposure, and multiorgan complications, such as rhabdomyolysis.[6,14,15]

In an attempt to detect virus in kidney cells, we used five distinct methodologies, namely immunostains for viral spike and nucleocapsid proteins, ISH for viral RNA (by automated platform and manual RNAScope), and ultrastructural examination, all of which failed to reveal definitive viral particles.[28,29] Whether rare, equivocal staining by manual RNAScope represents true positivity, correlating with low viral abundance,[17] or nonspecific staining requires further study. We cannot rule out the possibility that these techniques lack sufficient sensitivity for definitive viral detection, which may require such methodologies as RT-PCR.[17,30] We doubt that such rare and low abundance of virus is sufficient to account for the pathologic changes and favor predominant roles for cytokine-mediated and other systemic effects.[31]

Our series is limited by its descriptive nature. Coincidental associations with COVID-19 cannot be excluded, and detailed pathogenetic mechanisms will require further investigation. Because of lack of consent, *APOL1* genetic testing could not be performed on all black patients with podocytopathy. Clinical and laboratory information provided by referring nephrologists could not be independently verified. Because biopsies were performed for indication and the threshold for kidney biopsy in the setting of COVID-19 varies, our findings may not be generalizable to all patients with COVID-19. Given the recent timing of the pandemic, the follow-up period is necessarily short.

In conclusion, this biopsy series reveals diverse kidney pathology in SARS-CoV-2–infected patients. The findings highlight the potential for viral infection to influence innate or adaptive immune responses that in turn trigger new glomerular diseases (such as podocytopathies and anti-GBM nephritis) or exacerbate preexisting autoimmune or alloimmune conditions (such as lupus nephritis, membranous glomerulopathy, and allograft rejection). ATI is common and likely multifactorial. The lack of definitive virus in kidney cells argues against direct viral infection as the major pathomechanism.

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DISCLOSURES

J. Barasch reports that Columbia University has licenses to Abbott and Bioporto related to biomarkers such as NGAL, including EPO 1 616 184; USPO 7,977,110; EPO 1 616 184; and USPO 7,977,110. Bioporto provides royalties to Columbia University. All other authors have nothing to disclose.

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SUPPLEMENTAL MATERIAL

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Supplemental Figure 1. *In situ* hybridization for SARS-CoV-2 by manual RNAScope.

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