Autoantibodies related to systemic autoimmune rheumatic diseases in severely ill patients with COVID-19

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is the cause of COVID-19, which has affected more than 6 million people worldwide causing more than 400 000 deaths. The disease affects predominantly the upper and lower respiratory tracts causing severe pulmonary disease which often evolves to a multiorgan systemic disease.[1] This is evidenced by thromboembolic lesions of the heart and lungs, pulmonary haemorrhage, muscle weakness, hyperbilirubinaemia and lymphopenia suggesting that COVID-19 affects epithelial barriers, endothelial cells, coagulation, fibrinolysis and the immune system.[2] In patients who are severely ill, innate immune hyperactivity causes a cytokine storm which disturbs microcirculation resulting in shock and acute respiratory distress syndrome.[3] Systemic disease perpetuation may be due to the virus itself, infecting cells via ACE2 receptor or, following the cytokine storm, due to autoinflammatory and/or autoimmune mechanisms.[4]

Earlier studies reported that certain autoantibodies such as anti-cardiolipin (a-CL), anti-β2GPI and lupus anticoagulant might associate with the thromboembolic complications occurring in many patients with COVID-19.[5] These findings, along with the observation that certain clinical features of the infection can mimic those observed in systemic autoimmune diseases, prompted us to investigate serum autoantibodies in these patients and their clinical associations.

We tested the sera of 29 unselected severely ill patients with COVID-19 with positive SARS-CoV-2 PCR, admitted to the intensive care unit of Evangelismos Hospital, Athens, Greece (table 1). Twenty-one patients were male (72.4%) and eight were female (27.6%) with an average age of 64.2 years (range 43–85). Patients were hospitalised for an average of 25.6 days. Testing for anti-SARS-CoV-2 IgG antibodies revealed that 28/29 patients were positive with an average index of 8.54 (Euroimmun, Luebeck, Germany, positive cut-off >1.1). All patients were tested for the following autoantibodies using techniques routinely used in our laboratory: antinuclear antibodies (ANA), anti-neutrophil cytoplasmic antibodies (ANCA,immunofluorescence), antibodies to extractable nuclear antigens (ENA, immunoblot), a-dsDNA, a-CL (IgG/IgM), a-β2GPI (IgG/IgM) and anti-cyclic citrullinated peptide (CCP,ELISA). We found that 10 patients were positive for antinuclear antibodies (34.5%), 2 were positive for p-ANCA (6.9%), 2 were positive for c-ANCA (6.9%, one with a high 1/640 titre), 7 were positive for a-CL antibodies (24.1%, 4 IgG, 3 IgG+IgM), 10 were positive for anti-β2GPI antibodies (34.5%, 2 IgG, 5 IgM, 3 IgG+IgM) and 1 was positive for anti-CCP antibodies (3.5%). ANCA positivity was further investigated with ELISA, but no known specificities, including proteinase 3 or myeloperoxidase, were identified. This has been previously reported for acute viral hepatitis E.[6] None of the patients had a history of systemic autoimmune rheumatic disease. Overall, 20/29 patients with COVID-19 (68.7%) were positive for any kind of systemic autoantibody. Notable laboratory findings include: anaemia (44.9%), significant lymphopenia (62.1%), elevated lactic acid dehydrogenase levels (96.6%), increased fibrinogen levels (100%), elevated creatine kinase levels (41.4%) and increased C-reactive protein levels (89.6%). The average sequential organ failure assessment score of our patients was 7.89 (SD 2.44, range 2–11). Patients, with the exception of hydroxychloroquine (28/29 treated), were not treated with any immunomodulatory drugs.

The small sample size did not allow procurement of any statistically significant clinicolaboratory associations. Despite this and the lack of preinfection serological data, the presence of several systemic autoimmune reactivities in almost 70% of the patients suggests a post-SARS-CoV-2 or para-SARS-CoV-2 infectious autoimmune activation. This is not surprising, as cytokines present in the cytokine storm, for example, interleukin-6, can drive autoinflammatory reactions and autoimmunity, probably via pre-existing natural B cell clones or molecular mimicry. The possible autoimmune mechanism merits further investigation, therefore an autoimmune surveillance in larger cohorts is necessary to investigate possible mechanisms of COVID-19 perpetuation and to inform ongoing convalescence plasma therapeutic trials.[7 8] As several of the reported autoantibodies are disease markers and can be pathogenic, one should be cautious before transferring autoantibodies, along with neutralising antibodies, into severely affected patients.

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Table 1 The main clinical, laboratory and immunological findings are presented

Patient No	Sex/age	Medical history	Anti-SARS-CoV-2 IgG	COVID-19-related symptoms	SOFA score	Pulmonary imaging	Outcome	Elevated muscle enzymes	Prolonged aPPT	Autoantibodies
1	M/78	None	POS	Dyspnoea	10	Diffuse infiltrates and ground glass	Death	Yes	Yes	a-CL (IgG)
2	M/59	Sleep apnoea	POS	Fever, cough, dyspnoea	3	Diffuse infiltrates and ground glass	Alive	No	No	a-CL (IgG+IgM)
3	M/70	Hypertension	POS	Fever, dry cough, dyspnoea	10	Diffuse infiltrates and ground glass	Alive	No	No	NEG
4	M/46	Dyslipidaemia	POS	Fever	8	Diffuse infiltrates and ground glass	Alive	No	No	NEG
5	M/62	None	POS	Fever, cough, dyspnoea	8	Diffuse infiltrates and ground glass	Alive	No	No	NEG
6	M/54	Smoking	POS	Fever	4	Diffuse infiltrates	Alive	Yes	Yes	ANA 1/320 fine speckled cytoplasmic
7	M/79	None	POS	Fever, cough, dyspnoea	9	Diffuse infiltrates and ground glass	Alive	No	No	NEG
8	M/70	None	POS	Fever, productive cough	11	Diffuse infiltrates and ground glass	Alive	No	Yes	NEG
9	M/71	None	POS	Fever, cough, dyspnoea	9	Diffuse infiltrates and ground glass	Alive	Yes	Yes	p-ANCA 1/20
10	M/61	Coronary artery disease	POS	Fever, cough, dyspnoea	3	Diffuse infiltrates and ground glass	Alive	No	No	ANA 1/320 speckled cytoplasmic, p-ANCA 1/20 a-CL (IgG+IgM)
11	M/64	Hyperthyroid, dyslipidaemia	POS	Fever, cough, dyspnoea	11	Diffuse infiltrates	Alive	Yes	Yes	c-ANCA 1/640
12	M/61	Hypertension, diabetes mellitus	NEG	Fever, cough	10	Diffuse infiltrates	Alive	No	No	NEG
13	M/62	Smoking, arrhythmia	POS	Fever, cough, dyspnoea, diarrhoea	7	Diffuse infiltrates and ground glass	Alive	Yes	Yes	a-CL (IgG+IgM), a-CCP 70 IU
14	F/65	None	POS	Fever, cough, dyspnoea	8	Diffuse infiltrates	Alive	No	No	ANA 1/160 fine speckled nucleolar
15	F/58	Asthma, dyslipidaemia, hypertension, psoriasis, hepatitis	POS	Fever, productive cough, dyspnoea	11	Diffuse infiltrates	Death	Yes	No	a-CL (IgG)
16	F/85	Dyslipidaemia, hypertension	POS	Fever, cough	8	Diffuse infiltrates	In ICU	No	No	ANA 1/160 fine speckled nucleolar, a-β2GPI (IgG+IgM)
17	M/75	Hypertension, G6PD (–), hypothyroidism, renal CA	POS	Fever, cough, dyspnoea	9	Diffuse infiltrates and ground glass	In ICU	Yes	No	ANA 1/320 fine speckled nucleolar, Ro60, a-β2GPI (IgG)
18	F/60	Dyslipidaemia	POS	Fever	9	Diffuse infiltrates and ground glass	Alive	Yes	Yes	NEG
19	F/53	Obesity	POS	Fever, myalgia	10	Diffuse infiltrates	Alive	Yes	Yes	NEG
20	M/61	Hypertension, dyslipidaemia	POS	Fever, cough, dyspnoea	6	Diffuse infiltrates and ground glass	In ICU	Yes	No	ANA 1/320 fine speckled nucleolar, a-β2GPI (IgG)
21	F/56	Hypertension, obesity	POS	Fever, cough, dyspnoea	9	Diffuse infiltrates and ground glass	In ICU	Yes	Yes	a-CL (IgG)
22	M/67	Diabetes, hypertension, dyslipidaemia	POS	Headache, cough, fever, fatigue	5	Diffuse infiltrates and ground glass	Alive	No	Yes	c-ANCA 1/20, a-β2GPI (IgM)
23	F/66	Obesity	POS	Fever, cough	9	Diffuse infiltrates and ground glass	Alive	Yes	No	ANA 1/160 fine speckled nucleolar, a-β2GPI (IgM)

Continued

Table 1 Continued

Patient No	Sex/age	Medical history	Anti-SARS-CoV-2 IgG	COVID-19-related symptoms	SOFA score	Pulmonary imaging	Outcome	Elevated muscle enzymes	Prolonged aPPT	Autoantibodies
24	M/43	None	POS	Fever, cough	8	Diffuse infiltrates and ground glass	Alive	No	No	ANA 1/320 fine speckled nucleolar, 1/160 AMA, a-β2GPI (IgM)
25	M/75	Hypertension	POS	Fever, cough, dyspnoea	9	Diffuse infiltrates and ground glass	Death	No	Yes	ANA 1/320 speckled cytoplasmic, a-CL (IgG), a-β2GPI (IgM)
26	F/82	Hypertension	POS	Fever, cough, dyspnoea	8	Diffuse infiltrates and ground glass	Death	No	No	ANA 1/160 fine speckled nucleolar, 1/160 AMA, a-β2GPI (IgG+IgM)
27	M/55	Hypertension	POS	Fever, cough, fatigue	8	Diffuse infiltrates and ground glass	Alive	No	No	a-β2GPI (IgM)
28	M/64	Hypertension, dyslipidaemia	POS	Fever, dyspnoea	7	Diffuse infiltrates and ground glass	Alive	No	Yes	NEG
29	M/59	None	POS	Fever, diarrhoea	2	Diffuse infiltrates	Alive	No	Yes	a-β2GPI (IgG+IgM)

a-CL, anti-cardiolipin; AMA, anti-mitochondrial antibodies; ANA, antinuclear antibody; ANCA, anti-neutrophil cytoplasmic antibody; aPPT, activated Partial Thromboplastin Time; CCP, cyclic citrullinated peptide; ICU, intensive care unit; NEG, negative; POS, positive; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; SOFA, sequential organ failure assessment.

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