BMC Infectious Diseases

RESEARCH

Open Access

Study on the dynamic changes in antinuclear antibody spectrum in SARS-CoV-2 infection: a retrospective analysis

Jing Zheng[1†], Ying Zhang[1†], Jing Wang[1], Yijun Chen[1], Guangjun Zhu[1], Yufen Zheng[1], Haixi Yan[1], Jun Li[1*] and Bo Shen[1*]

Abstract

Background Patients with COVID-19 often produce multiple autoantibodies that impact immune function. This study aimed to assess changes in immune status and correlation with SARS-CoV-2 infection by analyzing dynamic shifts in patients' antinuclear antibody (ANA) profiles.

Methods A retrospective analysis was conducted on ANA data and clinical characteristics of 680 patients with novel coronavirus pneumonia admitted to Taizhou Enze Medical Center (Group) between December 7, 2022, and January 31, 2023. The analysis covered three phases: early COVID-19 (within one year before admission, T1), COVID-19 phase (during hospitalization, T2), and late COVID-19 (within one year after discharge, T3). ANA quantification was primarily performed using indirect immunofluorescence, and the magnetic stripe immunofluorescence luminescence method was employed to detect the ANA profile (ENA), including anti-dsDNA, nucleosome, Sm, SS-A/Ro52kD, SS-A/Ro60kD, SS-B/La, PCNA, AMA M2, Scl-70, and Jo-1.

Results During the T2 phase, 680 patients were analyzed. The positive rate of the ANA test was 35%. The proportion of autoimmune diseases (AID) in ANA-positive patients was higher than in ANA-negative patients (22%vs.7%). The ANA-positive group with AID showed higher ANA titers compared to the ANA-positive group without AID. During the follow-up one year before and after SARS-CoV-2 infection, in the T1-T2 group, there were two cases of ANA changing from negative to positive (one with AID, one without AID). The positive intensity of ANA increased by 15.6% and decreased by 20%. In the T2-T3 group, the positive intensity of ANA increased by 3.3% and decreased by 33.3%. Followed up of 7 patients with high ANA titers in T2 phase, among whom 5 cases did not support AID from the perspective of diagnosis and medication, and 2 cases were diagnosed with SLE after being infected with SARS-CoV-2.

†Jing Zheng and Ying Zhang contributed equally to this work and are co-first authors.

*Correspondence:
Jun Li
lij@enzemed.com
Bo Shen
shenb@enzemed.com

Full list of author information is available at the end of the article

Conclusions SARS-CoV-2 infection induces overactivation of the immune system, significantly impacting patients with autoimmune diseases. For patients without autoimmune diseases, ANA produced due to COVID-19 does not persist. Some COVID-19 patients may trigger their own immune system response.

Keywords SARS-CoV-2, Autoantibodies, Autoimmune diseases, Antinuclear antibody, Dynamic changes

Introduction

The novel coronavirus (SARS-CoV-2) infection not only severely impacts the immune system but may also trigger autoimmune-related symptoms in susceptible individuals and the presence of autoantibodies in blood circulation. Some patients have also been diagnosed with multiple autoimmune diseases [1], such as Guillain–Barré syndrome [2], Miller–Fisher syndrome [3], antiphospholipid syndrome (APS) [4], immune thrombocytopenic purpura [5], systemic lupus erythematosus (SLE) [6], and Kawasaki disease [7]. This indicates that SARS-CoV-2 infection can substantially affect the immune system and potentially induce autoimmune diseases in vulnerable populations.

In May 2020, Gazzaruso et al. [8] reported that among 45 patients admitted with COVID-19, 35.6% had antinuclear antibodies (ANA) in their systems. Another study by Vlachoyiannopoulos et al. [9] indicated that 34.5% (10 out of 29) of patients with severe COVID-19 tested positive for ANA, and nearly 70% of SARS-CoV-2 infected individuals exhibited activation of multiple autoimmune responses. The high occurrence of ANA and other autoimmune markers suggests that SARS-CoV-2 infection may trigger autoimmune mechanisms. However, their research lacked serological data before infection, making it unclear whether these ANAs are transient or persistent.

In this study, we found that the proportion of ANA positive detected by in-patients increased significantly during the epidemic period of COVID-19. The high positive rate led to the design of a retrospective study cohort focusing on the detection of ANA for patients with COVID-19, covering a period of one year before and after the outbreak and group COVID-19 patients with ANA positive detection according to whether they had autoimmune basic diseases.

Patients and methods

Patient monitoring and clinical evaluation

This study included patients with COVID-19 who were hospitalized at the Enze Medical Center between December 7, 2022, and January 31, 2023. Inclusion criteria required a confirmed diagnosis of SARS-CoV-2 infection through serological testing or nucleic acid detection of novel coronavirus following the "Recommendations for Diagnosis and Treatment of Novel Coronavirus Infection in China (7th edition)". ANA testing was performed during hospitalization. The final sample consisted of 680 COVID-19 patients. Clinical characteristics and laboratory test results at admission were collected from electronic medical records and are presented in Table 1. This study was approved by the Medical Ethics Committee of Taizhou Hospital, Zhejiang Province, China.

Study design

A total of 680 patients with COVID-19 hospitalized at the Enze Medical Center between December 7, 2022, and January 31, 2023, underwent ANA detection during their stay (T2). Based on the ANA test results, patients were classified into ANA-positive and ANA-negative groups. Differences in baseline data, COVID-19 disease classification, types of autoimmune diseases, and laboratory indicators between the two groups were analyzed. ANA data were collected for 45 patients one year before admission (T1) and for 30 patients one year after discharge (T3). Changes in ANA intensity and laboratory indicators were compared between patients with and without autoimmune diseases in the pre-middle (T1-T2) and post-middle (T2-T3) periods (Fig. 1). The pre-middle group (T1-T2) and post-middle group (T2-T3) of the ANA queue were paired and followed up at each stage of T1, T2, and T3.Autoimmune underlying diseases were diagnosed according to Chinese expert consensus on the diagnosis and treatment of autoimmune diseases. The types of AID we collected include SLE, interstitial pneumonia, RA, ANCA related vasculitis, Gout gout, and Other AID (such as Sjogren's syndrome, systemic sclerosis, Hashimoto's thyroiditis, myasthenia gravis, etc.).The 7 special cases are characterized by high ANA titers without T1 and T3.

Detection of ANAs in serum samples

ANAs were quantified primarily through indirect immunofluorescence, with a dilution of 1:100 considered positive. In cases of ANA positivity, magnetic stripe immunofluorescence was used to evaluate the ANA spectrum (ENA), which included anti-dsDNA, nucleosome, histone, CENuB, Sm, PO, SS-A/Ro52kD, SS-A/Ro60kD, SS-B/La, PCNA, AMA M2, Scl-70, J0-1, PM Scl, and U1snRNP (IgG). There was standardization across the pre-COVID, during-COVID, and post-COVID ANA measurements. And in all phases, there were the same assay platforms(Fully Automated Multi Immunoassay Analyzer (Mclia-800)(Lizhu)) and the same commercial kits(ANA-IgG detection kit (Indirect Immunofluorescence Method)(EUROIMMUN, Germany);

Table 1 Clinical characteristics and laboratory test results of patients (T2).($n=680$)

Characteristic	ANA + ($n=235$)	ANA - ($n=445$)	*P*
Sex - no. (%)			
Male	115 (48.9)	280 (62.9)	0.000
Female	120 (51.1)	165 (37.1)	
Age - yr.			
Median (IQR)	71.0 (59.0–80.0)	68.0 (56.0–78.0)	0.089
BMI, kg/m^2			
Total number	153	286	
Median (IQR)	22.6 (19.8–24.8)	23.4(21.3–26.0)	0.003
Onset to admission, days			
Median (IQR)	7.0 (3.0–10.0)	7.0 (3.0–10.0)	0.575
Onset to sampling time, days			
Median (IQR)	8.0 (4.0–11.0)	8.0 (4.0–11.0)	0.933
Length of stay, days			
Median (IQR)	9.0 (6.0–13.0)	9.0 (6.0–14.0)	0.307
COVID-19 typing - no. (%)			
Mild/asymptomatic	71 (30.2)	148 (33.3)	0.299
Moderate	98 (41.7)	189 (42.5)	
Severe	51 (21.7)	93 (20.9)	
Critical	15 (6.4)	15 (3.3)	
Outcome - no. (%)			
discharged with medical advice.	222 (94.5)	426 (93.0)	0.804
death	13 (5.5)	19 (7.0)	
Symptoms-no.(%)			
Cough	144 (61.3)	284 (63.8)	0.514
Sputum	123 (52.3)	233 (52.4)	0.996
Fever (temperature ≥ 37.3 ℃)	111 (47.2)	216 (48.5)	0.746
Chest tightness	82 (34.9)	126 (28.3)	0.077
Fatigue	40 (17.0)	103 (23.1)	0.062
Comorbidity- no. (%)			
Hypertension	102 (43.4)	211 (47.4)	0.318
Diabetes	47 (20.0)	116 (26.1)	0.078
Cardiovascular disease	43 (18.3)	56 (12.6)	0.045
Lung disease	34 (14.5)	55 (12.4)	0.438
Cerebral infarction	29 (12.3)	46 (10.3)	0.428
Nephropathy	29 (12.3)	64 (14.4)	0.461
Therioma	27 (11.5)	25 (5.6)	0.006
Autoimmunity disease- no. (%)			
Total number	51	31	
SLE	20 (39.2)	0 (0)	0.000
Interstitial pneumonia	14 (27.5)	4(12.9)	0.000
RA	8(15.7)	6(19.4)	0.669
AAV	6(11.8)	0(0)	0.047
Gout	2 (3.9)	11 (35.5)	0.000
Others	9 (17.6)	10 (32.3)	0.128

IQR Interquartile range, *SLE* Systemic lupus erythematosus, *RA* Rheumatoid arthritis, *AAV* Anti-Neutrophil Cytoplasmic Antibodies-associated vasculitis

ANA spectrum detection kit (Magnetic Stripe Code Immunofluorescence Luminescence method)(Zhuhai Lizhu Reagent Co., Ltd.)) used. Meanwhile, before each batch of tests, internal quality control (both high and low values) is conducted.

Definition

The positive intensity of the anti-nuclear antibody (T-ANA) was calculated as the sum of ANA-positive and ENA-positive intensities. ANA positivity was scored based on the observed positive intensity: no fluorescence was rated as 0, weakly positive fluorescence as 1, positive fluorescence intensity at 1:100 as 2, at 1:320 as 3, at 1:1000 as 4, at 1:3200 as 5, and 1:10000 as 6.

The ENA positive intensity was determined by the presence of positive results. No positive result was rated as 0. For positive results, the intensity was accumulated based on the number of positives. As per the instructions, a detection index of ≥ 1 was considered positive, and for dsDNA, a value of ≥ 100 was considered positive.

Statistical analysis

Descriptive statistical methods were used to analyze the data. Categorical variables were analyzed using frequency, while quantitative continuous variables were expressed as median and interquartile range (IQR). Fisher's exact test or the Mann–Whitney U-test was used to compare the groups. Continuity-corrected McNemar test and paired t-test were applied to evaluate the positive and negative levels of autoantibodies, respectively. A significance level of $p < 0.05$ was set for all tests. Statistical analyses were performed using SPSS 25.0 and R software version 4.0.2.

Results

First, we identified the prevalence and characteristics of ANA in patients with COVID-19, finding that 34.6% (235 of 680) tested positive for ANA. We compared the clinical characteristics (Table 1) and laboratory indicators (Fig. 2) of the ANA-positive (235 cases) and ANA-negative (445 cases) groups. Table 1 shows significant sex differences between the groups ($p = 0.000$). The man-to-woman ratio in the ANA-positive group was approximately equal (115 men, 48.9%; 120 women, 51.1%), whereas the ANA-negative group had a higher proportion of men (280 men, 62.9%; 165 women, 37.1%). BMI differed significantly between the groups ($p = 0.003$), with the ANA-positive group having a lower BMI than the ANA-negative group (22.5 ± 3.8 vs. 23.7 ± 3.8). The proportion of patients with malignant tumors was higher in the ANA-positive group compared to the ANA-negative group (11.5% vs. 5.6%, $p = 0.006$). However, there was no significant difference in age between the two groups ($p > 0.0500$), likely because the hospitalized population

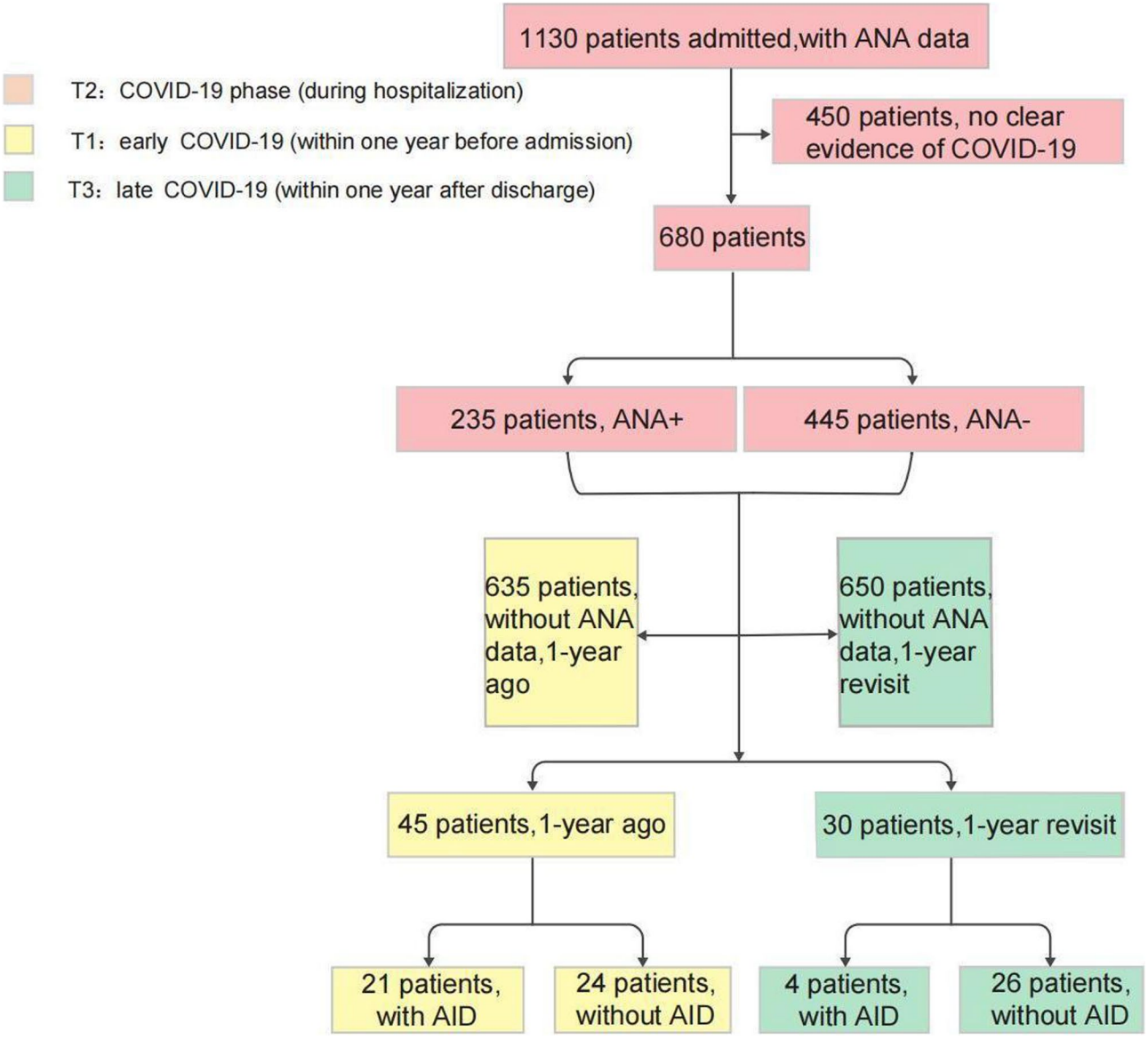

Fig. 1 Flowchart of the cohorts. ANA antinuclear antibodies, COVID-19 Coronavirus Disease 2019, AID Autoimmune Disease

was mostly elderly (average age > 65 years). There was no significant difference in the severity of COVID-19 between the two groups ($p > 0.0500$), with most patients having moderate severity. However, the proportion of critical patients was higher in the ANA-positive group (6.4% vs. 3.3%). The prognosis did not differ significantly between the groups ($p > 0.0500$), although the ANA-negative group had a higher death rate (7.0% vs. 5.5%).

To understand the impact of autoimmune diseases on ANA detection, we listed the common types of autoimmune diseases. We found that the ANA-positive group had a higher percentage of patients with autoimmune diseases compared to the ANA-negative group (22% vs. 7%). Specifically, the ANA-positive group predominantly consisted of patients with SLE, interstitial lung disease, and ANCA-related vasculitis, with these conditions being significantly more common than in the ANA-negative group. In contrast, gout was more prevalent in the ANA-negative group compared to the ANA-positive group (32.3% vs. 17.6%).

Secondly, we divided the ANA-positive group and ANA-negative group into four subgroups based on their own basic autoimmune disease (with AID) and no basic autoimmune disease (without AID) and compared the differences in laboratory test indicators related to COVID-19 (Fig. 2). We observed that the ANA-positive group with AID had the highest levels of IL-2 and IFN-γ and the lowest levels of IL-6 among the four subgroups, with significant differences ($p < 0.05$) compared to the ANA-positive group without AID and the ANA-negative

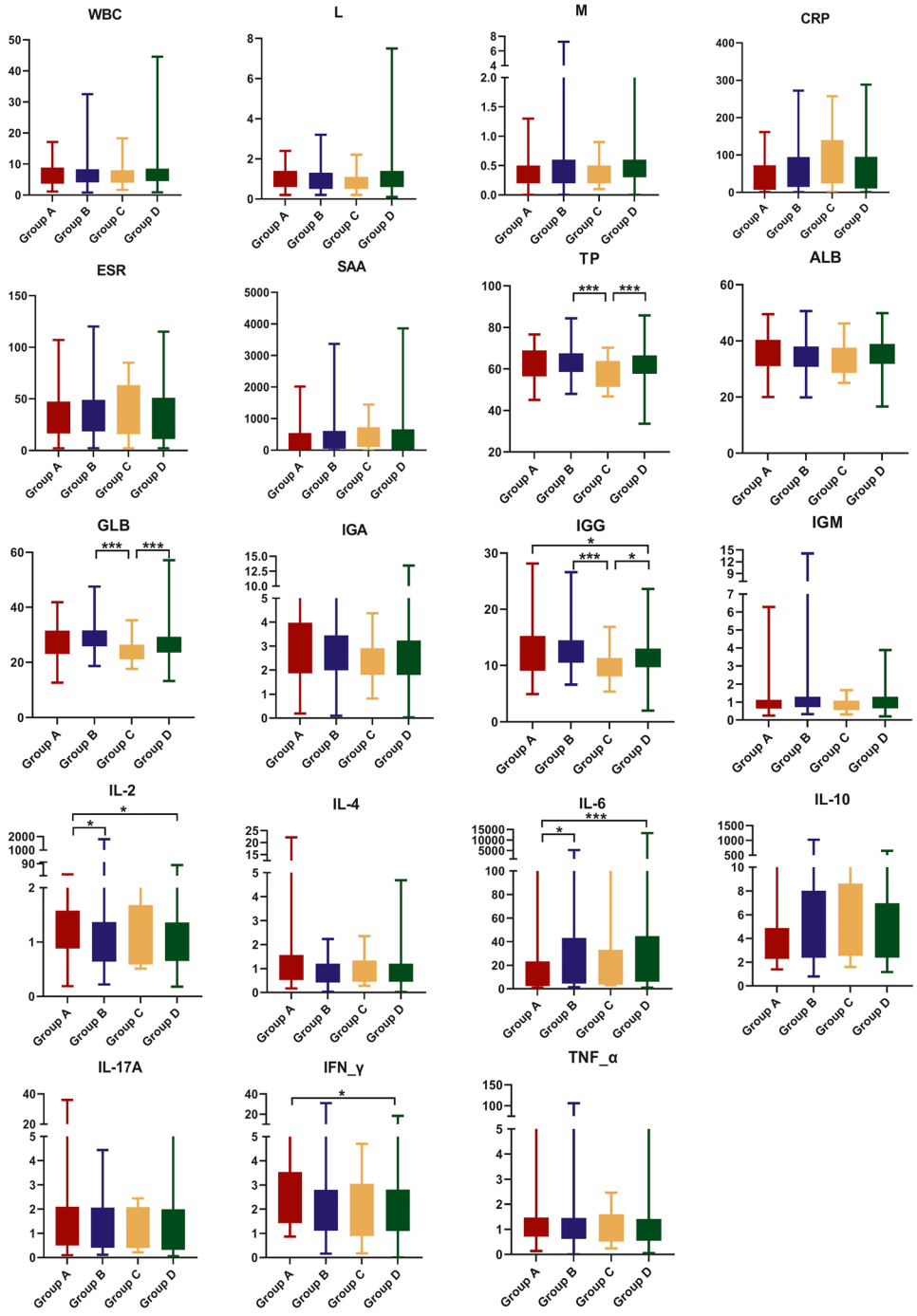

Fig. 2 Laboratory test results of patients (T2).($n = 680$) GROUP A：ANA+With AID；GROUP B：ANA+Without AID；GROUP C：ANA-With AID；GROUP D：ANA-Without AID;*：p <0.05,***：p <0.001.Medians and interquartile ranges (in parentheses) are specified for continuous variables, with p-values obtained by the Mann–Whitney U test. SAA：Serum Amyloid A；IL-2：interleukin-2；IL-4：interleukin-4；IL-6：interleukin-6；IL-10：interleukin-10；IL-17A：interleukin-17A；IFN_γ：interferon-γ；TNF_α：tumor necrosis factor-α

group without AID. The levels of total protein (TP) and globulin (GLB) were the lowest in the ANA-positive group with AID, and the distribution trend of the three immunoglobulins was consistent. Significant differences ($p < 0.05$) were observed compared with the ANA-positive group without AID and the ANA-negative group without AID. The highest level of GLB in the ANA-positive group without AID may be a transient increase unrelated to autoimmune diseases. The median values of IL-6 and C-reactive protein (CRP) in the ANA-positive group with AID were the lowest, while TP was the highest.

Next, to explore the differences in specific autoantibodies produced by the body after SARS-CoV-2 infection in individuals with basic autoimmune diseases, we assessed the distribution of each specific ENA in the ANA-positive group at T2 (Fig. 3-A and -B). Figure A shows that in the Without AID group, ANA was mainly low titer, but the proportion of high titer was higher than that in the With AID group, which may be related to the stronger overreaction of the autoimmune system of COVID-19 patients without AID to SARS-CoV-2. In Figure B, significant differences ($p < 0.05$) in ENA types between the two groups were observed. Among them, the fluorescence intensity of Sm, SSA, SSA-Ro52, nRNP, and P0 in the With AID group was higher than that in the Without AID group. The fluorescence intensity of double-stranded DNA (dsDNA) was higher than that of the Without AID group, with a significant difference between the two groups ($p < 0.001$).

Subsequently, we further studied the dynamic changes in the number and titer of ANAs in this cohort of patients during the one-year course before and after SARS-CoV-2 infection. Due to variations in the number of patients with ANA detection data in each stage, we divided the patients into the T1-T2 group and the T2-T3 group of COVID-19 and compared the dynamic changes in the spectrum of antinuclear antibodies before and after SARS-CoV-2 infection in each group. Among the 45 patients in the T1-T2 group, 2 cases showed a conversion from ANA-negative to positive (1 with AID and 1 without AID). Among the 30 patients in the T2-T3 group, there was no change in the ANA positivity rate at T3 compared to T2 (Table 2).

Simultaneously, we constructed shock plots illustrating the dynamic changes in ANAs for the two groups (Fig. 4): in the T1-T2 group change chart (A), the direction of changes from early to middle stages can be observed, with 2 cases of conversion from ANA-negative to positive accounting for 4.4%, an increase in ANA positivity intensity accounting for 15.6%, and a decrease in ANA positivity intensity accounting for 20%. In the T2-T3 group change chart (B), the direction of changes from middle to late stages can be seen, with no conversion from ANA-negative to positive or positive to negative. Patients with sustained positive ANA accounted for 43.3%, but the intensity of ANA positivity increased by 3.3%, and it decreased by 33.3%, with all patients with

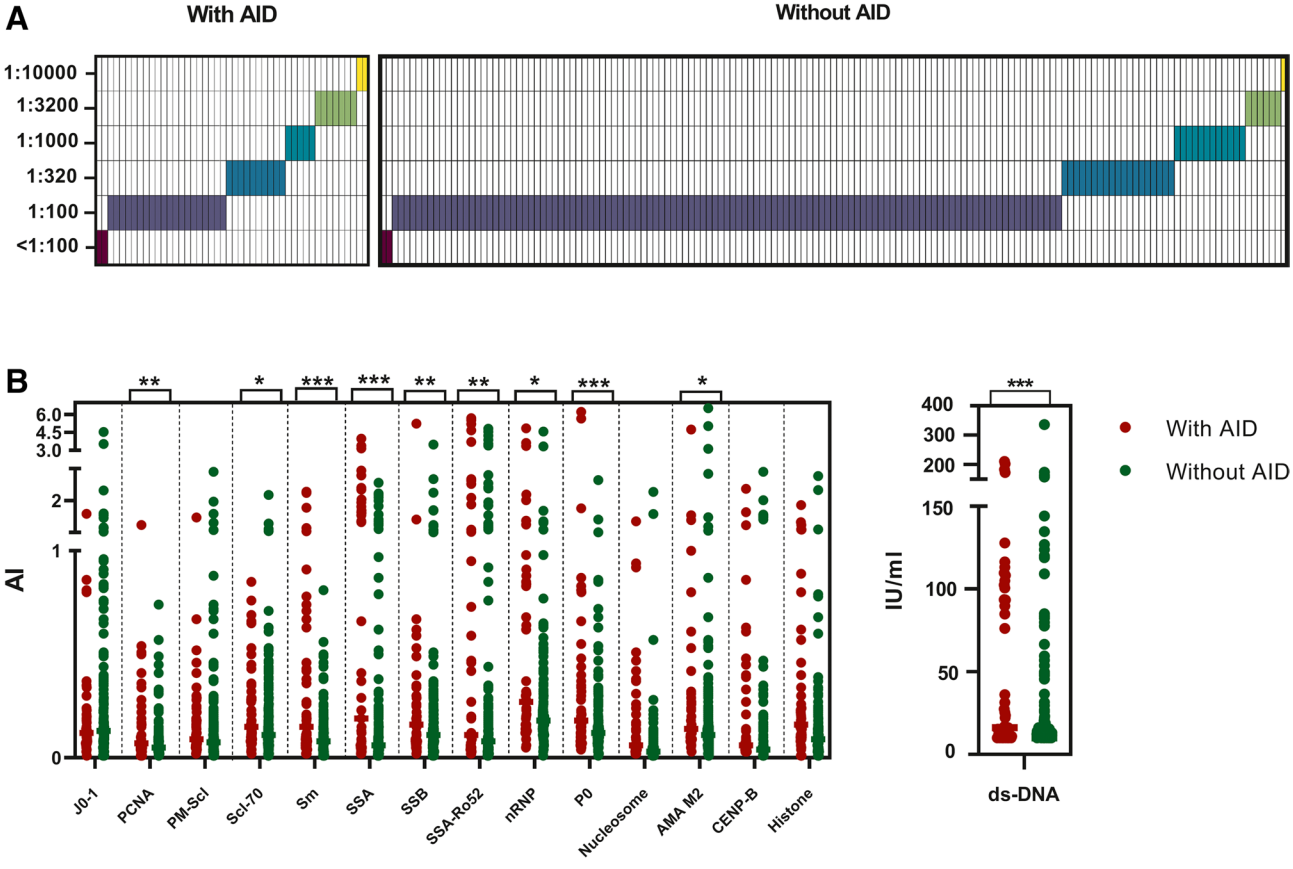

Fig. 3 **A** distribution of ANA titer in the ANA-positive group at T2 patients with and without autoimmune diseases. **B** Distribution of each specific ENA in the ANA-positive group at T2 patients with and without autoimmune diseases. Note: With AID: ANA-positive group with autoimmune diseases; Without AID: ANA-positive group without autoimmune diseases; *: $p < 0.05$,** : $p < 0.01$,*** : $p < 0.001$

Table 2 Comparison of ANA positivity rates in T1, T2, and T3 patients with and without autoimmune diseases

	T1-T2(*n* = 45)					T2-T3(*n* = 30)				
		T1		T2			T2		T3	
Total number- no. (%)	*n*	positive	negative	positive	negative	*n*	positive	negative	positive	negative
ANA	**45**	**26(57.8)**	**19(42.2)**	**28 (62.2)**	**17 (37.8)**	**30**	**24(80.0)**	**6(20.0)**	**24(80.0)**	**6(20.0)**
With AID	**21**	**16(76.2)**	**5(23.8)**	**17(81.0)**	**4(19.0)**	**4**	**3(12.5)**	**1(16.7)**	**3(12.5)**	**1(16.7)**
Without AID	**24**	**10(37.5)**	**14(62.5)**	**11(45.8)**	**13(54.2)**	**26**	**21(87.5)**	**5(83.3)**	**21(87.5)**	**5(83.3)**

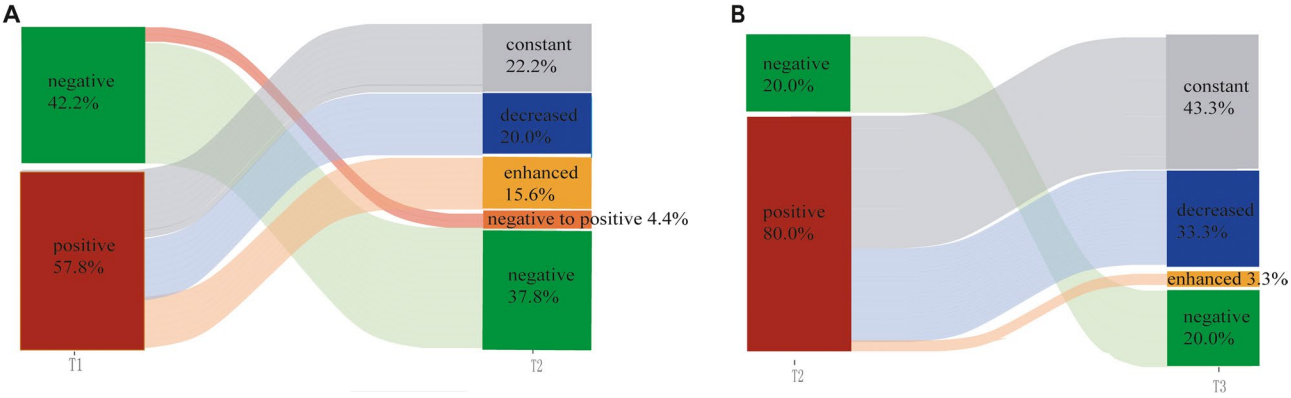

Fig. 4 Plots illustrating the dynamic changes in ANAs for the two groups. **A** T1-T2 group; (**B**) T2-T3 group

decreased intensity in the T3 phase having low titers in the T2 phase.

Meanwhile at the same time, we analyzed the complications of each patient in T1-T2 group and T2-T3 group (Figure S1). It can be seen that hypertension[31] and diabetes[32] accounted for the most complications in both groups. In T1-T2 group, two patients with negative to positive ANA had hypertension, including one with kidney disease, the other with diabetes, cerebral infarction and malignant tumor. In the T2-T3 group, one patient with increased ANA positive intensity also had hypertension.

negative: ANA test negative; positive ˚ ANA test positive ˚˚ enhanced ˚ Enhanced positive intensity of ANA detection ˚˚ decreased ˚ Decreased positive intensity of ANA detection ˚˚ constant ˚ ANA intensity remains unchanged.

Additionally, based on the dynamic changes in ANAs, we analyzed the laboratory indicators of the ANA-enhanced group, ANA-unchanged group, and ANA-weakened group at T1-T2 and T2-T3 (Fig. 5). It can be observed that the white blood cell (WBC) and lymphocyte (L) levels of the ANA-enhanced group at T1-T2 were the lowest, and the CRP level of the ANA-unchanged group was the highest, with a significant difference among the three groups. Due to the retrospective nature of our study and the presence of numerous missing values in the laboratory data at the T3 phase, we did not analyze the data for each group at T2-T3.

We also followed up with 7 patients with high ANA titers and without AID in T2 phase. Among them, 5 patients did not support AID in terms of diagnosis and medication, especially for patient P276, high levels of ANA persisted even after 2.7 years of re-infection with COVID-19. Two other patients were diagnosed with AID (SLE) during T2 phase (Fig. 6).

Discussion

SARS-CoV-2 infection can overactivate the immune system, leading to the production of autoantibodies that impact immune function. Gazzaruso et al. [10] found that 35.6% (16/45) of patients with COVID-19 tested positive for ANA. In a study by Vlachoyianopoulos et al. [8], 34.5% (10/29) of patients with severe COVID-19 were ANA-positive. Zhou et al. [11] detected autoantibodies in 20%–50% of patients with COVID-19 pneumonia. However, the persistence of ANA in these patients post-discharge remains unresolved.

We reviewed the electronic medical records and know that the purpose of detecting ANA before SARS-CoV-2 infection was to monitor, diagnose and differential diagnose the efficacy of patients with AID. Another reason for detecting ANA during SARS-CoV-2 infection period was that some patients had autoimmune related symptoms (such as arthralgia, skin itching, etc.). The reason for detecting ANAs after SARS-CoV-2 infection was to recheck the ANA with high titer in COVID-19 period, and the other reason was that patients had autoimmune symptoms after COVID-19 recovery. So in this study, we retrospectively analyzed 680 patients with COVID-19 and found that the positive detection rate of ANA was as high as 34.6% (235/680), consistent with the

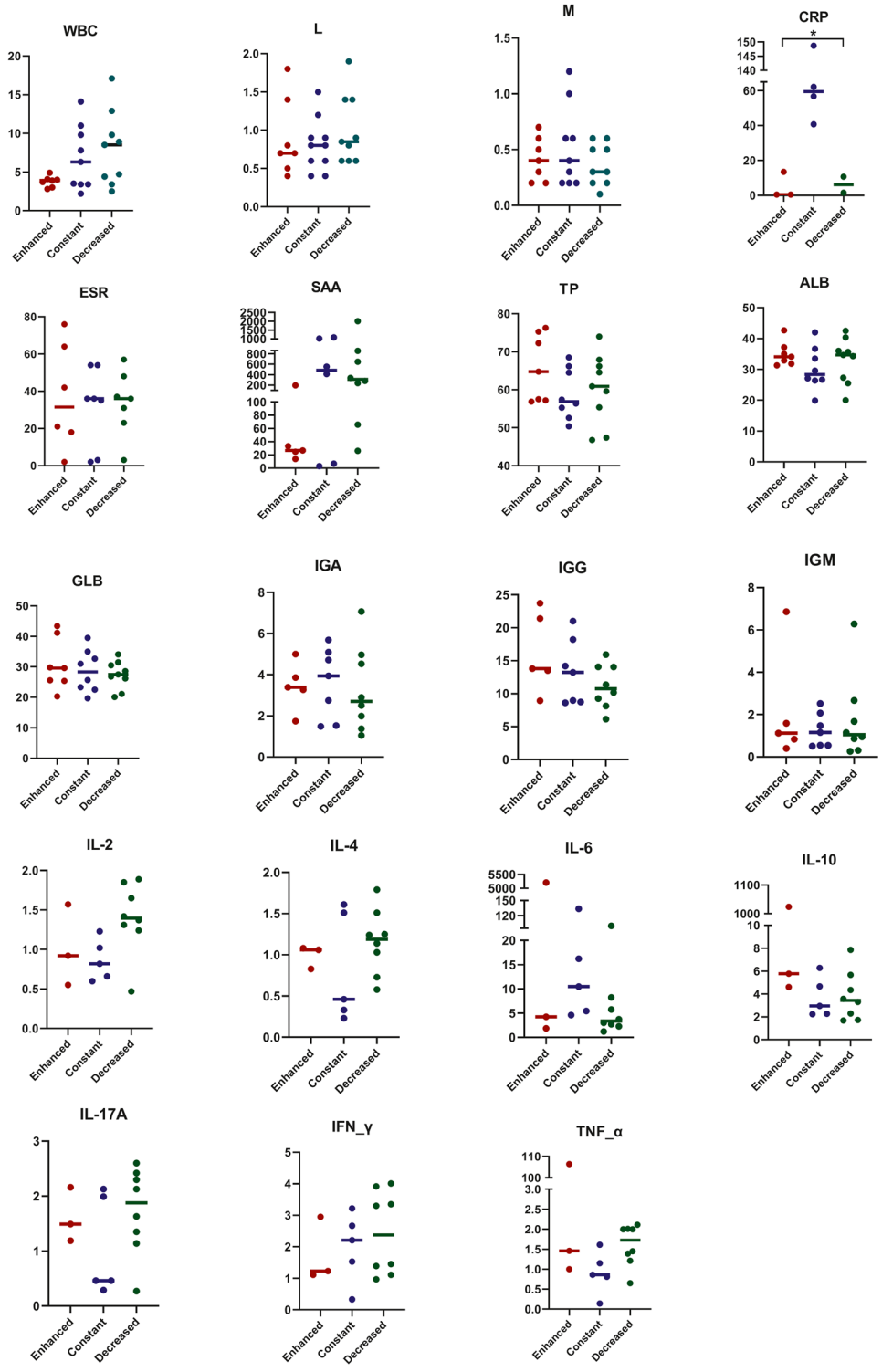

Fig. 5 Differences in laboratory indicators between T1-T2 groups. TP：Total protein；ALB：Albumin；GLB：Globulin；A/G：Ratio of albumin/globulin；IgG：Immunoglobulin G；IgA：Immunoglobulin A；IgM：Immunoglobulin M；*:p <0.05. SAA：Serum Amyloid A

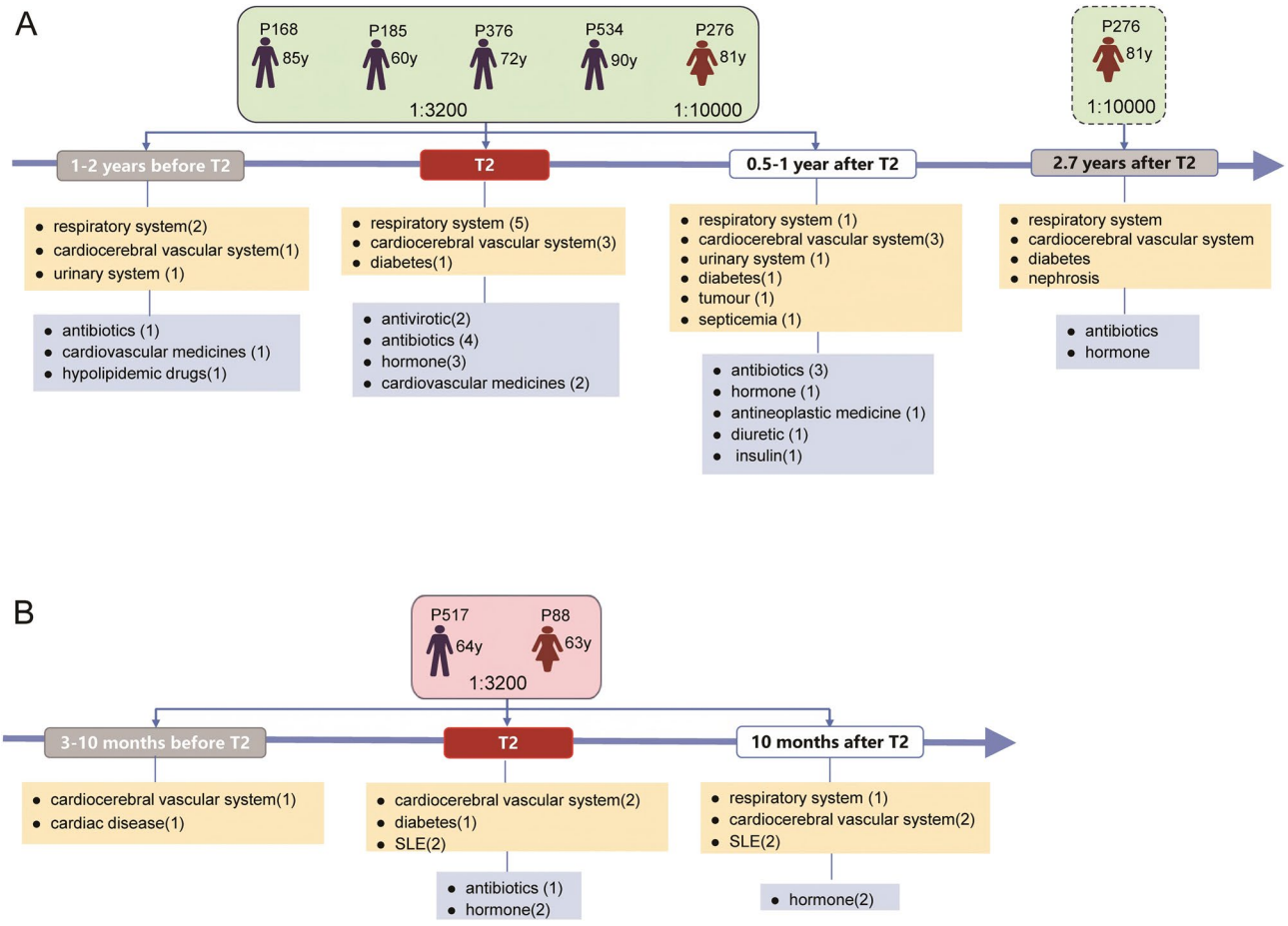

Fig. 6 Timeline of diagnosis and treatment for 7 special cases. **A** Timeline of follow-up for 5 patients with high ANA titers in T2 phase but not diagnosed with AID. **B** Timeline of follow-up for 2 patients with high ANA titers in T2 phase and diagnosed with AID. Green box and Pink box, represent basic information of 7 patients(patient number, gender, age, and titer value).Yellow box, represents clinical history and comorbidities. Blue box, represents the clinical treatment

aforementioned research findings; however, our extensive dataset provides more compelling evidence. Furthermore, we comprehensively evaluated the correlation between SARS-CoV-2 infection and the autoimmune status of the body by analyzing the dynamic changes in the spectrum of antinuclear antibodies in patients before, during, and after SARS-CoV-2 infection.

Firstly, we found that there were indeed more patients with AID in the ANA-positive group than in the ANA-negative group (22% vs. 7%), with the ANA-positive group mainly consisting of SLE, interstitial lung disease, and ANCA-related vasculitis, with a significantly higher proportion than the ANA-negative group, indicating significant differences. This may be related to the main histopathological feature of COVID-19, which is pulmonary microvascular disease. Patients with COVID-19 have been reported to exhibit evidence of fibrin thrombosis, activated platelets, and intravascular neutrophil traps [1, 2]. Infiltrating neutrophils, monocytes, and macrophages can be observed in other organs outside the lung, including the heart, central nervous system, and liver [2–4]. In addition to cell activation and infiltration, local and systemic complement activation may also lead to microvascular disease. However, since this subset of COVID-19 patients with autoimmune diseases had been treated with immunosuppressive therapy before, the ANA test in the acute phase of COVID-19 was positive, likely because SARS-CoV-2 infection accelerated the occurrence of autoimmunity and produced new autoantibodies [12].

Secondly, the number of confirmed autoantibodies related to SARS-CoV-2 infection has exceeded 20, mainly ANA and antibodies against anticoagulant cascade elements [1, 14, 15],and the exoproteome-directed autoantibodies have diverse efects on immune functionality and associations with clinical outcomes [13].However, differences in ENA were found between the group with autoimmune diseases and the group without autoimmune diseases during the COVID-19 period, with the former dominated by high titers; the low titer in the latter group is mainly related to the fact that SARS-CoV-2 can trigger

an exacerbated immune response [16].Patients without AID, and some patients with AID, suspend taking medicine because they are worried about immunosuppression or lack of availability of drugs [17], and reduce their visits because they are concerned by the spread of COVID-19 [18]. This discontinuity in medical care and non-compliance with medications can worsen autoimmune disease activity, resulting in a heightened immune response and high-titer ANAs in COVID-19 patients with autoimmune diseases. Significant differences in ENA types were observed between the two groups. The fluorescence intensities of Sm, SSA, SSA-Ro52, nRNP, and P0 were higher in with AID group compared to the without AID group, though high titers autoantibodies detected in COVID-19 patients without AID [9]. In the context of acute SARS-CoV-2 infection, our results of this part also indicated that the increase of ANA is still less than that caused by AID. This difference may be related to the specific ENA types present in patients with underlying AID.

Additionally, we monitored the dynamic changes in the number and titer of ANAs in patients before and after COVID-19, over one year. In the group of patients infected in the T1-T2 phase of the study, two individuals showed a conversion from negative to positive ANA status. The increase in ANA-positive intensity in this group was 15.6%, while the decrease in ANA-positive intensity accounted for 20%. There was no conversion from negative to positive ANA status in the T2-T3 group of SARS-CoV-2 infection, but the increase in ANA-positive intensity accounted for 3.3%, and the decrease in ANA-positive intensity accounted for 33.3%. All these decreased patients (10cases) exhibited middle and low titers during the T2 infection stage, verifying that some autoantibodies were temporarily produced by SARS-CoV-2 infection [29]. Furthermore, 9 cases with increased positive intensity suggested that SARS-CoV-2 caused potential de novo autoantibody-production at this time [19, 30].

Analysis of 7 special cases, among which 5 cases did not support AID and their high titer of ANA may be caused by SARS-CoV-2 infection. Especially for patient P276, both instances of high ANA titers of 1:10000 were associated with SARS-CoV-2 infection, which further confirmed this point[20]. Two other patients were diagnosed with AID (SLE) during T2 phase, this also indicates that SARS-CoV-2 had accelerated the occurrence of autoimmunity [12].

Moreover, we found that the proportion of positive intensity of ANA in patients with AID after SARS-CoV-2 infection was higher than in patients without AID. However, the proportion of decline of ANA in COVID-19 patients with AID at the later stage of the disease was higher than in COVID-19 patients without AID, consistent with the research of Vladioyiannopoulos et al., who suggested that novel SARS-CoV-2 infection could overactivate autoimmune systems in patients to produce ANA. For patients without AID, ANA should be a transient result of COVID-19 pathogenesis, not a persistent result [20].

Finally, regarding laboratory indicators, this study found that the ANA-positive group with AID had the highest levels of IL-2 and IFN-γ, and the lowest level of IL-6 compared to other groups. The severity of COVID-19 disease in this group was lower than in other groups, which aligns with the findings of Gong B et al. who suggested that high levels of IL-6 contribute to the severity of COVID-19 [21, 22]. The ANA-positive group with AID had the lowest levels of TP and GLB compared to other groups, with the distribution trend of the three immunoglobulins being consistent. The highest level of serum globulin in the ANA-positive group without AID may be a transient increase unrelated to autoimmune diseases. However, the median levels of IL-6, ESR, CRP, and SAA in the ANA-positive group with AID were the highest, while TP was the lowest, likely related to the abnormal activation of immune cells and the overexpression of inflammatory cytokines and chemical mediators caused by SARS-CoV-2 infection [23–27].

The WBC and L levels in the ANA-enhanced group were the lowest in T1-T2, while the CRP levels were the highest in the ANA-unchanged group, indicating a significant difference among the three groups. Due to the retrospective nature of our study, there were missing values in the T3 phase laboratory data; therefore, we did not analyze the data for each group of T2-T3. The level of IgG antibody in the ANA-enhanced group is the highest, and the level of IgA antibody in the ANA-weakened group is the lowest. This observation indirectly confirms the conclusion of Ampudia et al. that the level of anti-SARS-CoV-2 IgG antibody is very high from the acute attack stage of COVID-19 to the late stage of New Corona, while the level of IgA antibody recovers to the baseline level in the late stage of COVID-19 [28].

This study offers the advantage of observing and analyzing the dynamic data of the ANA spectrum in the same patient before, during, and after SARS-CoV-2 infection, and presenting the diagnosis, complications and treatment information of the special cases. Deeply discussed the complications and diagnostic basis of special cases provide a theoretical basis for evaluating the changes and correlation of immune status before and after SARS CoV-2 infection.

However, this is the limitation of our study. As this is a paired retrospective study, among the patients who underwent ANA testing during the COVID-19 period, those who had undergone ANA testing before the pandemic were relatively fewer, and the same situation occurred after the pandemic, which led to the lack of datas that we included in the pre COVID-19 period (T1)

and the late COVID-19 period (T3), and could not display the dynamic trend more completely.

In summary, the clinical significance of ANA detection in patients with COVID-19 requires further investigation. Particularly, for patients without AID, ANA production should be temporary during COVID-19 and dissipate within weeks, not persist. Conversely, in patients with AID, SARS-CoV-2 infection may induce overactivation of the immune system, leading to ANA production or enhancement. Whether this phenomenon can interact with and exacerbate the treatment of autoimmune diseases warrants further study.

Conclusion

SARS-CoV-2 can trigger exacerbated immune responses resulting in the production of autoantibodies, including ANA. The intensity of ANA positivity is more pronounced in patients with preexisting AID, such as those with COVID-19. However, in COVID-19 patients without underlying AID, ANA production is likely to be temporary rather than persistent.

Abbreviations

ANA	Antinuclear antibody
ENA	ANA profile
AID	Autoimmune diseases

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12879-025-11848-9.

Supplementary Material 1.

Acknowledgements

We thank Dr. Xuefen Li of the Laboratory Department of First Affiliated Hospital of Zhejiang University for her guidance.

Authors' contributions

JZ: Conceptualization, Collected data, Formal analysis, Funding acquisition, Investigation, Methodology, Visualization, Writing original draft.YZ: Collected data, Analyzed the data, Prepared figures and tables, Writing original draft. JW: Data curation, Methodology, Resources.YJC: Data curation, Methodology, Resources.GJZ: Investigation, Methodology.YFZ: Investigation, Methodology. HXY: Conceptualization, Investigation, Methodology.JL: Conceptualization, Investigation, Methodology, Project administration, Supervision. BS: Conceptualization, Investigation, Methodology, Project administration, Supervision.

Funding

The author(s) declare that no financial support was received for the research, authorship, and/or publication of this article.

Data availability

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

The research related to human use has been complied with all the relevant national regulations, institutional policies and in accordance the tenets of the Helsinki Declaration, and has been approved by the Ethics Committee of Taizhou Hospital in Zhejiang Province.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

Author details

[1]Taizhou Hospital of Zhejiang Province Affiliated to Wenzhou Medical University, 150 Ximen Road, Linhai, Taizhou, Zhejiang Province, China

Received: 10 December 2024 / Accepted: 29 September 2025
Published online: 27 October 2025

References

1. Ehrenfeld M, Tincani A, Andreoli L, et al. Covid-19 and autoimmunity [J]. Autoimmun Rev. 2020;19(8):102597.
2. Sedaghat Z, Karimi N. Guillain barre syndrome associated with COVID-19 infection: a case report. J Clin Neurosci. 2020;76:233–5.
3. Manganotti P, Pesavento V, Buoite Stella A, et al. Miller fisher syndrome diagnosis and treatment in a patient with SARS-CoV-2. J Neurovirol. 2020;26(4):605–6.
4. Zhang Y, Cao W, Jiang W, et al. Profile of natural anticoagulant, coagulant factor and anti-phospholipid antibody in critically ill COVID-19 patients [J]. J Thromb Thrombolysis. 2020;50(3):580–6.
5. Tsao HS, Chason HM, Fearon DM. Immune thrombocytopenia (ITP) in a pediatric patient positive for SARS-CoV-2. Pediatrics. 2020;146(2):e20201419.
6. Bonometti R, Sacchi MC, Stobbione P, et al. The first case of systemic lupus erythematosus (SLE) triggered by COVID-19 infection [J]. Eur Rev Med Pharmacol Sci. 2020;24(18):9695–7.
7. Jones VG, Mills M, Suarez D, et al. COVID-19 and Kawasaki disease: novel virus and novel case [J]. Hosp Pediatr. 2020;10(6):537–40.
8. Vlachoyiannopoulos PG, Magira E, Alexopoulos H, et al. Autoantibodies related to systemic autoimmune rheumatic diseases in severely ill patients with COVID-19. Ann Rheum Dis. 2020;79(12):1661–3.
9. Fujii H, Tsuji T, Yuba T, et al. High levels of anti-SSA/Ro antibodies in COVID-19 patients with severe respiratory failure: a case-based review [J]. Clin Rheumatol. 2020;39(11):3171–5.
10. Gazzaruso C, Carlo Stella N, Mariani G, et al. High prevalence of antinuclear antibodies and lupus anticoagulant in patients hospitalized for SARS-CoV2 pneumonia. Clin Rheumatol. 2020;39(7):2095–7.
11. Zhou Y, Han T, Chen J, et al. Clinical and autoimmune characteristics of severe and critical cases of COVID-19. Clin Transl Sci. 2020;13(6):1077–86.
12. Chang SE, Feng A, Meng W, et al. New-onset IgG autoantibodies in hospitalized patients with COVID-19. Nat Commun. 2021;12(1):5417.
13. Wang EY, Mao T, Klein J, et al. Diverse functional autoantibodies in patients with COVID-19. Nature. 2021;595(7866):283–8. https://doi.org/10.1038/s41586-021-03631-y.
14. Dotan A, Muller S, Kanduc D, et al. The SARS-CoV-2 as an instrumental trigger of autoimmunity. Autoimmun Rev. 2021;20(4):102792.
15. Favaloro EJ, Henry BM, Lippi G. COVID-19 and antiphospholipid antibodies: time for a reality check? Semin Thromb Hemost. 2022;48(1):72–92.
16. Pascolini S, Vannini A, Deleonardi G, et al. COVID-19 and immunological dysregulation: can autoantibodies be useful? Clin Transl Sci. 2021;14(2):502–8.
17. Khabbazi A, Kavandi H, Paribanaem R, et al. Adherence to medication in patients with rheumatic diseases during COVID-19 pandemic. Ann Rheum Dis. 2022;81(10):e200.
18. Chuah SL, Teh CL, Wan Mohd Akbar SA, Cheong YK, Sachdev Manjit Singh B. Impact of COVID-19 pandemic on hospitalisation of patients with systemic lupus erythematosus (SLE): report from a tertiary hospital during the peak of the pandemic. Ann Rheum Dis. 2022;81(8):e144.
19. Anaya J-M, Monsalve DM, Rojas M, et al. Latent rheumatic, thyroid and phospholipid autoimmunity in hospitalized patients with COVID-19. J Transl Autoimmun. 2021;4:100091.
20. Shoenfeld Y, Blank M, Abu-Shakra M. et al. The mosaic of autoimmunity: prediction, autoantibodies, and therapy in autoimmune diseases-2008 [J]. Isr Med Assoc J. 2008;10(1): 13–19.

21. Gong B, Huang L, He Y, et al. A genetic variant in IL-6 Lowering its expression is protective for critical patients with COVID-19. Signal Transduct Target Ther. 2022;7(1):112. Published 2022 Apr 2.
22. Liu Y, Sawalha AH, Lu Q. COVID-19 and autoimmune diseases. Curr Opin Rheumatol. 2021;33(2):155–62.
23. Gusev E, Sarapultsev A, Solomatina L, Chereshnev V. SARS-CoV-2-specific immune response and the pathogenesis of COVID-19. Int J Mol Sci. 2022;23(3):1716.
24. Satış H, Özger HS, Aysert Yıldız P, et al. Prognostic value of interleukin-18 and its association with other inflammatory markers and disease severity in COVID-19. Cytokine. 2021;137:155302.
25. Vassallo M, Manni S, Pini P, et al. Patients with covid-19 exhibit different immunological profiles according to their clinical presentation. Int J Infect Dis. 2020;101:174–9.
26. Azar MM, Shin JJ, Kang I, Landry M. Diagnosis of SARS-CoV-2 infection in the setting of the cytokine release syndrome. Expert Rev Mol Diagn. 2020;20(11):1087–97.
27. Sun Y, Dong Y, Wang L, et al. Characteristics and prognostic factors of disease severity in patients with COVID-19: the Beijing experience. J Autoimmun. 2020;112:102473.
28. Acosta-Ampudia Y, Monsalve DM, Rojas M, et al. Persistent autoimmune activation and proinflammatory state in post-Coronavirus disease 2019 syndrome. J Infect Dis. 2022;225(12):2155–62.
29. Son K, Jamil R, Chowdhury A, et al. Circulating anti-nuclear autoantibodies in COVID-19 survivors predict long COVID symptoms. Eur Respir J. 2023;61(1):2200970. https://doi.org/10.1183/13993003.00970-2022.
30. Gracia-Ramos AE, Martin-Nares E, Hernández-Molina G. New onset of autoimmune diseases following COVID-19 diagnosis. Cells. 2021;10(12):3592. https://doi.org/10.3390/cells10123592.

Publisher's Note