Anti-Mitotic Spindle Apparatus Autoantibodies: Prevalence and Disease Association in Chinese Population

Qian Xi,^{1,2} Yongkang Wu,² Lixin Li,² Bei Cai,² Junlong Zhang,² Bin Yang,²
and Lanlan Wang^{2*}

¹Department of Clinical Laboratory, Sichuan Academy of Medical Sciences & Sichuan Provincial People's Hospital, Chengdu, China

²Department of Laboratory Medicine, West China Hospital, Sichuan University, Chengdu, China

Background: Mitotic spindle apparatus (MSA) antibodies are rare findings with undefined clinical significance in clinical research. We aimed at investigating the prevalence and clinical significance of anti-MSA antibodies in Chinese population. **Methods:** Between 2008 and 2013, a total of 180,180 patients were studied for the presence of anti-MSA antibodies. The clinical details and laboratory data of anti-MSA-positive patients were retrospectively collected and analyzed. **Results:** Of the 180,180 patients tested, 68,640 patients presented with positive antinuclear antibodies (ANAs, 38.10%), but only 32 patients with positive anti-MSA antibodies (0.018%). Diagnoses were established in 22 of 32 patients: 16 connective tissue diseases (CTDs), mainly Sjogren syndrome (SS, 5/16), rheumatoid arthritis

(RA, 4/16), and systemic lupus erythematosus (SLE, 3/16), and 6 nonautoimmune conditions. The most frequent clinical symptoms of the anti-MSA-positive patients were arthralgia and eyes and mouth drying. Additionally, 70% of anti-MSA antibodies were not associated with other ANAs, however, when associated, the most frequent ANA was anti-SSA. **Conclusions:** Anti-MSA antibodies have a low prevalence and female gender predominance. Anti-MSA antibodies are primarily associated with CTDs, mainly SS, RA, and SLE. The presence of anti-MSA antibodies might be the unique serological markers of the CTDs, especially when anti-SSA, SSB, and dsDNA antibodies are negative, or the level of RF is low. J. Clin. Lab. Anal. 30:702–708, 2016. © 2016 Wiley Periodicals, Inc.

Key words: connective tissue disease; mitotic spindle apparatus; rheumatoid arthritis; Sjogren syndrome; systemic lupus erythematosus

INTRODUCTION

During the past 50 years, autoantibodies of many nuclear and cytoplasmic proteins, such as antinuclear antibodies (ANAs), have been described in the sera of patients with diverse conditions. ANAs, involved in many aspects of cell functions, including protein synthesis, DNA replication, and mitosis, are the hallmark of the diagnosis, classification, and disease activity monitoring of almost all systemic autoimmune rheumatic diseases (1–3). Inflammation, immune hyperstimulation, and any procedure that is associated with tissue destruction might stimulate the generation of ANAs (4). ANA testing, revealing several well-established fluorescence patterns, is now routine practice in the diagnosis and management of diseases with an underlying autoimmune pathogenesis. Anti-mitotic spindle apparatus (anti-MSA) antibodies,

described by McCarty et al. in 1981 (5), are infrequently detected in ANA routine IIF (indirect immunofluorescence) screening of human pathological sera (6). MSA, a unique microtubule or protein structure involved in mitosis and postmitotic nuclear reorganization (7, 8), is composed of two main antigens, the nuclear mitotic

Grant sponsor: National Natural Science Foundation of China; Grant numbers: 81202354 and 81301496.

*Correspondence to: Lanlan Wang, Department of Laboratory Medicine, West China Hospital, Sichuan University, No. 37 Guoxue Alley, Wuhou, Chengdu 610041, China. E-mail: wanglanlanhxvip@163.com

Received 20 September 2015; Accepted 9 December 2015

DOI 10.1002/jcla.21925

Published online in Wiley Online Library (wileyonlinelibrary.com).

apparatus protein (NuMA1) and spindle kinesin-like protein HsEg5 (NUMA2) (9–11).

The presence of circulating ANAs is a hallmark of many autoimmune diseases or clinical symptoms and can be a useful tool in the investigation of pathogenic mechanism of diseases and the nature and function of target cellular constituents. So, the exploration of autoantibodies is extremely significant to disease diagnosis and prognosis, and is of great importance to the clinical and scientific research.

Circulating anti-MSA antibodies have been reported to be associated with a wide spectrum of pathological conditions, mainly connective tissue diseases (CTDs) (12), infections (13–16), malignancy (17), and primary biliary cirrhosis (18). However, at the moment few studies have clearly stated the prevalence and clinical correlation of anti-MSA antibodies, especially in Chinese population. Hence, in the present study, we set out to investigate the prevalence and clinical significance of anti-MSA antibodies to determine if they are related to some specific diseases, disease manifestations, or clinical indexes and to guide clinical decision, therapy, and further research through retrospective study.

MATERIALS AND METHODS

Subjects and Samples

A total of 180,180 sera were evaluated for ANAs in the clinical medicine laboratory of West China Hospital Sichuan University between June 2008 and June 2013. We retrospectively collected and analyzed clinical details and laboratory data of patients with positive anti-MSA antibodies. Our study passed the ethical review of hospital.

Methods

ANA test was performed by IIF on HEp-2 cells (EU-ROIMMUN IF), using serial dilutions commencing at 1:100. ANA profiles, including anti-SSA, SSB, U1RNP, Sm, Scl-70, Jo-1, and Rib antibodies, were detected by immunoblot assay (EURO Blot Master).

Anti-double-stranded DNA (anti-dsDNA), anti-keratin antibody (AKA), and anti-neutrophil cytoplasmic antibody (ANCA) were performed by IIF (EU-ROIMMUN), using serial dilutions commencing at 1:10. The clinical indexes of biochemistry, immunity, and hematology are the routine clinical examinations.

Statistical Analysis

Categorical variables were compared using Fisher's exact test, and continuous variables using the *t*-test. Probability (*P*) values less than 0.05 were considered

statistically significant. All analyses were performed using SPSS17.0.

RESULTS

The Main Characteristics of the Study Population

A total of 180,180 patients were tested for ANAs between June 2008 and June 2013, among which 680,640 patients were discovered to have positive ANAs (38.10%), while only 32 patients was found to present with positive anti-MSA antibodies (0.018%), whose mean age were 50.9 ± 13.7 years. The ANA median titer of these anti-MSA-positive patients was 438.63 ± 3.49 . Besides, 70% of anti-MSA antibodies always independently existed without association with other ANAs, while 30% were associated with other autoantibodies, among which the most frequent was anti-SSA antibody (Table 1). Besides, rheumatoid factor (RF) was present in high concentration (>100 KU/l) in 4 of 24 patients tested. The main features of the population are presented in Table 1. However, of the 32 patients with positive anti-MSA antibodies, only 22 (including 19 women (86.4%) and 3 men (13.6%)) had definitive diagnoses (Table 2). In addition, only 14 of the 22 patients with definitive diagnoses had detailed clinical data.

Anti-MSA Antibodies and Disorders

First, we analyzed the association of anti-MSA antibodies with autoimmune diseases. In our study, 16 of 22 patients with definitive diagnoses (72.7%, 16/22) suffered from autoimmune diseases, which were Sjogren syndrome (SS) in 5 patients (31.2%, 5/16), rheumatoid arthritis (RA) in 4 patients (25.0%, 4/16), systemic lupus erythematosus (SLE) in 3 patients (18.7%, 3/16), undifferentiated CTD (UCTD) in 3 patients (18.7%, 3/16), and dermatomyositis in 1 patient (6.2%, 1/16). In the 16 patients with autoimmune diseases, some other ANAs were also observed in addition to the anti-MSA antibodies, among which anti-SSA was detected positive in 4 of 16 (25%, 4/16) patients, anti-SSB in 1 (6.2%, 1/16), anti-RNP in 1 (6.2%, 1/16), and anti-AKA in 1 (6.2%, 1/16). However, two of the five patients with SS and one of the three patients with SLE were found to have positive anti-MSA antibodies without other ANA antibodies. In addition, of the 11 patients (with autoimmune diseases) tested, only 3 patients were found to have high concentration of RF (more than 100 KU/l). Among the four RA patients, three were found to have negative ANA antibodies and concentration of RF less than 30 KU/l, but they were observed to have positive anti-MSA antibodies.

Next, we investigated the correlation of anti-MSA antibodies with nonautoimmune diseases. In our study, we

TABLE 1. Main Features of the Study Population

ANA profiles (n/N (%))															
		Age	Female	Median									AKA	ANCA	RF
	N	(years)	(n (%))	titer of	dsDNA	Jo-1	U1RNP	SSA	SSB	Rib	Scl-70	Sm	(n (%))	(n (%))	(n (%))
				ANAs											
Patients	32	50.9 ± 13.7	29 (90.6)	438.63 ± 3.49	0/32 (0)	1/30 (3.3)	1/30 (3.3)	9/30 (30)	2/30 (6.7)	0/30 (0)	0/30 (0)	0/30 (0)	1/10 (10)	0/7 (0)	4/24 (16.7)

found that six patients with positive anti-MSA antibodies (27.3%, 6/22) presented with nonautoimmune diseases, of which one had hypertrophic cardiomyopathy; one had esophageal achalasia; one had pulmonary interstitial fibrosis; one had ephrapostasis after renal transplantation; one had nasopharyngeal carcinoma; and one had hypothyroidism combining hypertension, diabetes, and osteoporosis. No positive antibodies and high concentration of RF were discovered. In addition, no significant differences in age and ANA titer were found between the patients with autoimmune disorders and the patients with nonautoimmune conditions ($P > 0.05$).

In addition, only 14 of 22 patients with definitive diagnoses had detailed clinical data (Table 2). Among them, five patients were observed to have the clinical symptoms of arthralgia (35.71%), four eyes and mouth drying (28.57%), two limited mobility (14.28%), two renal dysfunction (14.28%), two anemia (14.28%), and two splenomegaly (14.28%).

Anti-MSA Antibodies and the Fluorescence Patterns

In our study, the spindle fluorescence pattern was present in every patient with positive anti-MSA antibodies. However, some other fluorescence patterns were also observed in addition to the spindle pattern. Among the 22 anti-MSA-positive patients tested, speckled pattern (59.1%, 13/22; Supplementary Fig. 1) was found more frequently than other fluorescence patterns, including speckled nucleolar pattern (4.5%, 1/22), speckled homogeneous pattern (13.6%, 3/22), speckled cytoplasmic pattern (4.5%, 1/22), homogeneous pattern (9.1%, 2/22), vimentin pattern (4.5%, 1/22), and PCNA pattern (4.5%, 1/22). According to the differences of the fluorescence pattern, we divided the patients into two groups, the speckled spindle pattern group and the other patterns group. The main characteristics of these patients are reported in Table 3.

In the speckled spindle pattern group, 13 patients (who were all females) were included. A definite autoimmune disease was established for ten patients (76.9%), including SS ($n = 4$), RA ($n = 3$), SLE ($n = 2$), and UCTD

($n = 1$). However, nonautoimmune disorders including one esophageal achalasia, one pulmonary interstitial fibrosis, and one hypothyroidism cases were also discovered. Among the 12 patients tested with speckled spindle pattern, ANA antibodies coexisted with anti-MSA antibodies in three patients (25.0%, 3/12), that is, anti-SSA in three patients, anti-SSB in one patient, and anti-RNP in one patient.

In the other patterns group, nine patients were included. Among them, six patients (66.7%, 6/9) presented with autoimmune diseases, including two UCTD, one SS, one RA, one SLE, and one dermatomyositis cases. Among the eight patients tested with speckled spindle pattern, anti-SSA coexisted with anti-MSA antibodies in one patient (12.5%, 1/8). Beyond all that, no significant differences were found in sex, age, disease constitution, ANA titer, and the presence of associated ANA profile antibodies between the speckled spindle pattern and other patterns groups (Table 3). Besides, there were no significant differences in the level of clinical indexes of liver and renal function, immunologic function, and blood routine (Table 4).

DISCUSSION

This is the first study of the clinical significance of anti-MSA antibodies in Chinese population. Anti-MSA antibodies are a rare finding during ANA testing, with an incidence varying between 0.18% (19) and 0.45% (20) in survey populations. Our results showed a lower prevalence (0.018%) of anti-MSA antibodies among the tested samples, and a low frequency (0.047%) in the positive ANA samples, which might be explained by the differences of race and disease distribution. It is reported that around 80% of the people affected with autoimmune diseases are women (21, 22), as women always respond to infection and trauma with increased antibody production and a more T-helper-2-predominant immune response (23, 24). In our study, we found that gender predominance existed with a high proportion of female patients (90.6%), which is consistent with the known higher prevalence of autoimmunity and most autoimmune diseases in women.

TABLE 2. Clinical Characteristics of MSA-Positive Patients

No.	Sex	Age (years)	Clinical symptoms	Anti-SSA	Anti-SSB	Anti-RNP	Other antibodies	RF	Diagnosis
5	F	53	Eyes and mouth drying, arthralgia, limited mobility	Neg	Neg	Neg			SS, OA
9	F	41	Eyes and mouth drying, dizzy	Neg	Neg	Neg		<20	SS
12	F	67	Eyes and mouth drying	++	Neg	Neg		<20	SS
15	F	57		+++	Neg	Neg	AKA+		SS
17	F	45	Vomit, eyes and mouth drying	++	+	Neg		191	SS
6	F	20	Arthralgia	Neg	Neg	Neg		25.6	RA
10	F	44	Arthralgia, limited mobility	Neg	Neg	Neg		<20	RA
14	F	35	Arthralgia	Neg	Neg	Neg		23.1	RA
16	F	47	Arthralgia	Neg	Neg	Neg		114	RA
11	F	35	Anemia, splenomegaly,	+++	Neg	+++			SLE
18	F	51							SLE
19	M	76	Anemia, dry skin, rash, gum bleeding, oral ulcer, renal dysfunction	Neg	Neg	Neg		212	SLE
1	F	28							UCTD
2	F	51		Neg	Neg	Neg		<20	UCTD
21	F	43	Hair loss, conjunctival congestion	Neg	Neg	Neg		<20	UCTD
4	M	59		Neg	Neg	Neg			DM
3	M	50	Chest distress	Neg	Neg	Neg		<20	DCM
7	F	77		Neg	Neg	Neg			Esophageal achalasia
8	F	49		Neg	Neg	Neg		<20	Pulmonary interstitial fibrosis
13	F	50	Fever, renal dysfunction	Neg	Neg	Neg		<20	Nephropostasis after renal transplantation
20	F	60		Neg	Neg	Neg		<20	Hypothyroidism, hypertension, diabetes, osteoporosis
22	F	49	Splenomegaly, dizzy, bloody sputum	Neg	Neg	Neg		<20	NPC

No., the number of the patients with positive anti-MSA; F, female; M, male; Neg, negative; +, the degree of positive; DCM, dilated cardiomyopathy; NPC, nasopharyngeal carcinoma; DM, dermatomyositis; OA, osteoarthritis.

Anti-MSA antibodies are reported to be associated with a wide variety of autoimmune diseases including RA, SS, SLE, MCTD, polyarteritis nodosa, polymyositis, and Hashimoto's thyroiditis (9, 10, 12, 20, 25–27). Our study revealed that 72.7% (16/22) of anti-MSA-positive patients in Chinese had autoimmune CTDs, mostly SS, RA, and SLE. In keeping with the previous reported

studies (12, 26), our results exposed that anti-MSA antibodies were almost not associated with other ANA profiles (70%), while when associated, the most frequent ANA was anti-SSA. In addition, it is also important to point out that anti-MSA antibodies were the unique positive ANAs in two of five anti-MSA-positive SS, three of four anti-MSA-positive RA, and one of three

TABLE 3. Comparison Between the Speckled Spindle Pattern Group and Other Patterns Group

Patients	All patients (<i>n</i> = 22)	Speckled spindle pattern group (<i>n</i> = 13)	Other patterns group (<i>n</i> = 9)	<i>t</i>	<i>P</i>
Age	49.4 ± 13.7	48.5 ± 13.3	50.8 ± 15.0	−0.381	0.707
Females (<i>n</i> (%))	19 (86.4)	13 (100)	6 (66.7)		0.055
Autoimmune diseases (<i>n</i> (%))	16/22 (72.7)	10/13 (76.9)	6/9 (66.7)		0.655
SS (<i>n</i> (%))	5 (22.7)	4 (30.8)	1 (11.1)		0.360
RA (<i>n</i> (%))	4 (18.2)	3 (23.1)	1 (11.1)		0.616
SLE (<i>n</i> (%))	3 (13.6)	2 (15.4)	1 (11.1)		1
UCTD (<i>n</i> (%))	3 (13.6)	1 (7.7)	2 (22.2)		0.544
Dermatomyositis (<i>n</i> (%))	1 (4.5)	0 (0)	1 (11.1)		0.409
Nonautoimmune diseases (<i>n</i> (%))	6/22 (27.3)	3/13 (23.1)	3/9 (33.3)		0.655
Esophageal achalasia (<i>n</i> (%))	1 (4.5)	1 (7.7)	0 (0)		1
Pulmonary interstitial fibrosis (<i>n</i> (%))	1 (4.5)	1 (7.7)	0 (0)		1
Hypothyroidism, hypertension, diabetes, osteoporosis (<i>n</i> (%))	1 (4.5)	1 (7.7)	0 (0)		1
Hypertrophic cardiomyopathy (<i>n</i> (%))	1 (4.5)	0 (0)	1 (11.1)		0.409
Nephropostasis after renal transplantation (<i>n</i> (%))	1 (4.5)	0 (0)	1 (11.1)		0.409
Nasopharyngeal carcinoma (<i>n</i> (%))	1 (4.5)	0 (0)	1 (11.1)		0.409
Geometric mean titer of ANA	564.47 ± 3.56	770.19 ± 3.83	360.33 ± 2.91		0.17
Associated ANA profile (<i>n</i> (%))	4/20 (20)	3/12 (25)	1/8 (12.5)		0.619
Positive anti-RNP (<i>n</i> (%))	1/20 (5)	1/12 (8.3)	0/8 (0)		1
Positive anti-SSA (<i>n</i> (%))	4/20 (20)	3/12 (25)	1/8 (12.5)		0.619
Positive anti-SSB (<i>n</i> (%))	1/20 (5)	1/12 (8.3)	0/8 (0)		1

The comparison was between the speckled spindle pattern group and other patterns group. Data were analyzed by Fisher's exact tests except for age, using *t*-tests.

anti-MSA-positive SLE patients. Taken together, these results display that the anti-MSA antibodies may be the potential serological marker of SS, RA, and SLE, especially when anti-SSA, SSB, and dsDNA antibodies are negative, or the level of RF is low.

Apart from autoimmune disorders, anti-MSA antibodies can be also found associated with nonautoimmune conditions, which were osteoarthritis, dilated cardiomyopathy, cancer (such as melanoma and adenocarcinoma), and infection (5, 13–16, 28). In our series, one serum from an esophageal achalasia patient, without arthritis or other features to suggest autoimmune disease, was recognized anti-MSA. We also found one patient of ephrapostasis after renal transplantation also had positive anti-MSA antibodies, which might be associated with transplantation immunity. To the best of our knowledge, these have never been mentioned before, while the mechanism still remains unknown. It might be the result of the generation of antibodies against various autoantigens, including virus, tumor suppression genes (P53), proliferation associated antigens (i.e., CENP-F), cancer antigens (MAGE etc.) (12, 17). As the results of repeated ANA tests were not available in any of these patients, it would be of interest to determine whether anti-MSA antibodies disappeared after treatment or were just transient, and whether it may serve as a serological indicator of these nonautoimmune conditions.

Besides, we also collected the detailed clinical data of 14 anti-MSA-positive patients, and found that the most

frequent clinical symptoms were arthralgia, followed by eyes and mouth drying. It suggested that the presence of anti-MSA might be associated with the constitution of diseases mentioned above, such as RA, SS, and SLE, which were involved in joint, eyes, and mouth. It is further confirmed that the presence of anti-MSA antibodies was associated with CTDs, especially for SS, RA, and SLE.

However, in our study, we found the spindle pattern, as one kind of fluorescence pattern, was not presented alone in the anti-MSA-positive sera, but always accompanied by other fluorescence patterns, mainly speckled pattern (59.1%). It is reported that the nuclear speckled pattern is strongly associated with anti-Sm, Ro/SSA, La/SSB, and anti-U1snRNP antibodies (29–31). On the contrary, anti-Ro/SSA or anti-La/SSB antibodies are associated with SS (32), and anti-dsDNA or anti-Sm antibodies are associated with SLE (6, 33), so the nuclear speckled pattern might be associated with SS and SLE. Our study, in accordance with this conclusion, showed that 76.9% (10/13) of anti-MSA-positive patients with speckled pattern had CTDs, mostly SS (30.8%), RA (23.1%), and SLE (15.4%), while only 25% of these patients had other positive ANAs, mostly SSA, SSB, and RNP. So we speculate that some of these speckled antibodies, accompanying with spindle antibodies, could be anti-SSA, SSB, and RNP antibodies, the other might be some molecules or antibodies not detected by routine IIF, or some new autoantibodies that had not been found before. But the hypothesis remains to be confirmed.

TABLE 4. Comparison of the Level of Clinical Serum Indexes Between the Speckled Spindle Pattern Group and Other Patterns Group

Patients	All patients (<i>n</i> = 22)	Speckled spindle pattern group (<i>n</i> = 13)	Other patterns group (<i>n</i> = 9)	<i>t</i>	<i>P</i>
Liver function					
TP	66.28 ± 19.11	63.93 ± 23.39	70.98 ± 2.45	-0.660	0.521
ALB	42.76 ± 4.53	43.93 ± 3.18	41.26 ± 5.76	1.187	0.255
AST	24.75 ± 11.00	26.40 ± 11.63	23.28 ± 9.97	0.576	0.573
ALT	22.19 ± 12.37	24.44 ± 14.04	19.29 ± 10.11	0.819	0.427
ALP	72.62 ± 23.58	74.00 ± 29.96	70.86 ± 13.66	0.256	0.802
GGT	26.12 ± 17.60	28.10 ± 19.64	23.29 ± 15.20	0.543	0.595
Renal function					
BUN	5.79 ± 2.72	5.21 ± 1.54	6.62 ± 3.85	-1.052	0.309
CREA	79.28 ± 38.66	66.04 ± 14.51	98.20 ± 54.41	-1.803	0.092
CYS-C	1.14 ± 0.53	1.02 ± 0.23	1.32 ± 0.78	-1.167	0.262
Immunologic function					
C3	0.84 ± 0.22	0.87 ± 0.23	0.79 ± 0.21	0.728	0.477
C4	0.18 ± 0.064	0.20 ± 0.075	0.17 ± 0.051	0.687	0.502
IgA	2076.15 ± 959.08	2221.43 ± 1096.71	1,906.67 ± 836.80	0.573	0.578
IgG	15.52 ± 4.10	15.64 ± 3.95	15.37 ± 4.64	0.114	0.911
IgM	1853.08 ± 988.03	1698.71 ± 1121.90	2033.17 ± 871.89	-0.592	0.566
CD3	64.85 ± 11.27	65.03 ± 9.25	64.64 ± 14.51	0.055	0.958
CD4	31.92 ± 7.57	33.10 ± 10.07	30.50 ± 3.43	0.547	0.598
CD8	28.41 ± 10.68	26.98 ± 7.21	30.12 ± 14.62	-0.465	0.653
Blood routine					
HGB	124.45 ± 20.29	124.83 ± 23.78	123.88 ± 15.10	0.101	0.921
RBC	3.81 ± 1.29 × 10 ⁹	4.24 ± 0.61	3.57 ± 1.23	1.624	0.122
WBC	6.74 ± 3.39 × 10 ⁶	6.82 ± 3.34	6.13 ± 3.26	0.441	0.665
PLT	165.65 ± 98.90	185.58 ± 111.93	137.68 ± 62.66	1.095	0.288

The comparison was between the speckled spindle pattern group and other patterns group. All of the data were analyzed by *t*-tests.

TP, total protein; ALB, albumin; AST, aspartate aminotransferase; ALT, alamine aminotransferase; ALP, alkaline phosphatase; GGT, gamma-glutamyl transpeptidase; BUN, blood urea nitrogen; CREA, creatinine; CYS-C, cystatin; C3, complement 3; C4, complement 4; IgA, immunoglobulin A; IgG, immunoglobulin G; IgM, immunoglobulin M; CD3, cluster differentiation antigen 3; CD4, cluster differentiation antigen 4; CD8, cluster differentiation antigen 8; HGB, hemoglobin; RBC, red blood cell; WBC, white blood cell; PLT, platelets.

CONCLUSIONS

Anti-MSA antibodies are featured by a low prevalence and female gender predominance, and have a strong association with CTDs, especially SS, RA, and SLE. Isolated presence of anti-MSA antibodies might be a potential unique serological marker of these diseases.

CONFLICT OF INTEREST

The authors declare that they have no competing interests.

Abbreviations

AKA	= anti-keratin antibody
CTD	= connective tissue disease
IIF	= indirect immunofluorescence
MSA	= mitotic spindle apparatus
RA	= rheumatoid arthritis
SLE	= systemic lupus erythematosus
SS	= Sjogren syndrome
UCTD	= undifferentiated connective tissue disease

REFERENCES

- Solomon DH, Kavanaugh AJ, Schur PH. Evidence-based guidelines for the use of immunologic tests: Antinuclear antibody testing. *Arthritis Rheum* 2002;47:434-444.
- Fritzer MJ. Challenges to the use of autoantibodies as predictors of disease onset, diagnosis and outcomes. *Autoimmun Rev* 2008;7:616-620.
- Bertin D, Jourde-Chiche N, Bongrand P, et al. Original approach for automated quantification of antinuclear autoantibodies by indirect immunofluorescence. *Clin Dev Immunol* 2013;2013:182172.
- Samsami Dehaghani A, Karimaghahi N, Parsanezhad ME, et al. Anti-nuclear antibodies in patients with polycystic ovary syndrome before and after laparoscopic electrocauterization. *Iran J Med Sci* 2013;38:187-190.
- McCarty GA, Valencia DW, Fritzler MJ, et al. A unique antinuclear antibody staining only the mitotic-spindle apparatus. *N Engl J Med* 1981;305:703.
- Tan EM, Cohen AS, Fries JF, et al. The 1982 revised criteria for the classification of systemic lupus erythematosus. *Arthritis Rheum* 1982;25:1271-1277.
- Ou Y, Rattner JB. A subset of centrosomal proteins are arranged in a tubular conformation that is reproduced during centrosome duplication. *Cell Motil Cytoskeleton* 2000;47:13-24.
- Kisurina-Evgenieva O, Mack G, Du Q, et al. Multiple mechanisms regulate NuMA dynamics at spindle poles. *J Cell Sci* 2004;117:6391-6400.

9. Whitehead CM, Winkfein RJ, Fritzler MJ, et al. The spindle kinesin-like protein HsEg5 is an autoantigen in systemic lupus erythematosus. *Arthritis Rheum* 1996;39:1635–1642.
10. Andrade LE, Chan EK, Peebles CL, et al. Two major autoantigen-antibody systems of the mitotic spindle apparatus. *Arthritis Rheum* 1996;39:1643–1653.
11. Grypiotis P, Ruffatti A, Tonello M, et al. [Clinical significance of fluoroscopic patterns specific for the mitotic spindle in patients with rheumatic diseases]. *Reumatismo* 2002;54:232–237.
12. Limaye V, Roberts-Thomson P, Gillis D, et al. The clinical associations of mitotic spindle autoantibodies in a South Australian cohort. *Aust N Z J Med* 1999;29:713–717.
13. Lind K, Hoier-Madsen M, Wiik A. Autoantibodies to the mitotic spindle apparatus in *Mycoplasma pneumoniae* disease. *Infect Immun* 1988;56:714–715.
14. Cimolai N, Mah D, Roland E. Anticentriolar autoantibodies in children with central nervous system manifestations of *Mycoplasma pneumoniae* infection. *J Neurol Neurosurg Psychiatry* 1994;57:638–639.
15. Huidbuchel E, Blaschek M, Seigneurin JM, et al. Anti-organelle and anti-cytoskeletal autoantibodies in the serum of Epstein-Barr virus-infected patients. *Ann Med Interne (Paris)* 1991;142:343–346.
16. Auer-Grumbach P, Achleitner B. Epidemiology and clinical associations of NuMA (nuclear mitotic apparatus protein) autoantibodies. *J Rheumatol* 1994;21:1779–1781.
17. Abu-Shakra M, Buskila D, Ehrenfeld M, et al. Cancer and autoimmunity: Autoimmune and rheumatic features in patients with malignancies. *Ann Rheum Dis* 2001;60:433–441.
18. Hansen BU, Eriksson S, Lindgren S. High prevalence of autoimmune liver disease in patients with multiple nuclear dot, anti-centromere, and mitotic spindle antibodies. *Scand J Gastroenterol* 1991;26:707–713.
19. McCarty GA, Valencia DW, Fritzler MJ. Antibody to the mitotic spindle apparatus: Immunologic characteristics and cytologic studies. *J Rheumatol* 1984;11:213–218.
20. Webb J, Maule P, Wells JV. Antibody to mitotic spindle apparatus. *J Rheumatol* 1985;12:623–624.
21. Gleicher N, Barad DH. Gender as risk factor for autoimmune diseases. *J Autoimmun* 2007;28:1–6.
22. Fairweather D, Frisancho-Kiss S, Rose NR. Sex differences in autoimmune disease from a pathological perspective. *Am J Pathol* 2008;173:600–609.
23. Klein SL. The effects of hormones on sex differences in infection: From genes to behavior. *Neurosci Biobehav Rev* 2000;24:627–638.
24. Lang TJ. Estrogen as an immunomodulator. *Clin Immunol* 2004;113:224–230.
25. Rattner JB, Rao A, Fritzler MJ, et al. CENP-F is a .ca 400 kDa kinetochore protein that exhibits a cell-cycle dependent localization. *Cell Motil Cytoskeleton* 1993;26:214–226.
26. Szalat R, Ghillani-Dalbin P, Jallouli M, et al. Anti-NuMA1 and anti-NuMA2 (anti-HsEg5) antibodies: Clinical and immunological features: A propos of 40 new cases and review of the literature. *Autoimmun Rev* 2010;9:652–656.
27. Bonaci-Nikolic B, Andrejevic S, Bukilica M, et al. Autoantibodies to mitotic apparatus: Association with other autoantibodies and their clinical significance. *J Clin Immunol* 2006;26:438–446.
28. Lleo A, Invernizzi P, Gao B, et al. Definition of human autoimmunity–autoantibodies versus autoimmune disease. *Autoimmun Rev* 2010;9:A259–266.
29. Migliorini P, Baldini C, Rocchi V, et al. Anti-Sm and anti-RNP antibodies. *Autoimmunity* 2005;38:47–54.
30. Tan EM, Feltkamp TE, Smolen JS, et al. Range of antinuclear antibodies in "healthy" individuals. *Arthritis Rheum* 1997;40:1601–1611.
31. Tozzoli R, Bizzaro N, Tonutti E, et al. Guidelines for the laboratory use of autoantibody tests in the diagnosis and monitoring of autoimmune rheumatic diseases. *Am J Clin Pathol* 2002;117:316–324.
32. Vitali C, Bombardieri S, Moutsopoulos HM, et al. Preliminary criteria for the classification of Sjogren's syndrome. Results of a prospective concerted action supported by the European Community. *Arthritis Rheum* 1993;36:340–347.
33. Hochberg MC. Updating the American College of Rheumatology revised criteria for the classification of systemic lupus erythematosus. *Arthritis Rheum* 1997;40:1725.