Erhöhte ANA, wie weiter?

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ANA without specific symptoms

Patient characteristics	Symptoms	Lab results
• Female patient • Born 1986 • No underlying disease • None smoker • IT specialist	• Generalized pain • Headaches • Fatigue • Depression	• ANA 1:320 • CRP and BSR normal

Avoid ANA testing in patients with no or unspecific symptoms

Probability of increased ANA in the general population: 2,5%

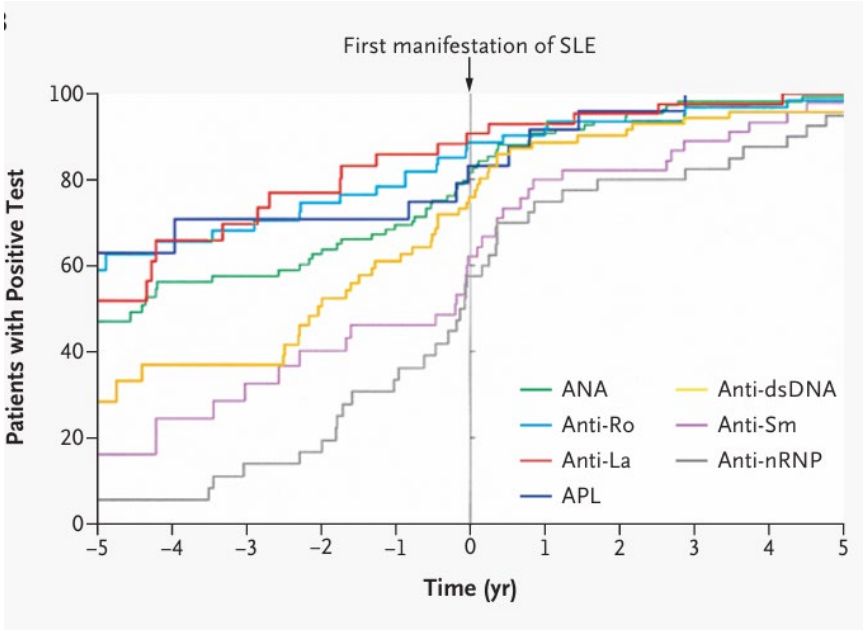
Switzerland: 250 000 positive ANA tests (10 million inhabitants)

Prevalence of SLE: 0.1% = 10 000 positive ANA

At least 200 000 positive ANA without clinical relevance

Grygiel-Górniak et al, Reumatologia 2018; 56, 4: 243–248

Avoid ANA testing in patients with no or unspecific symptoms - despite it is an early preclincal phenomenon in CTDs



At least 200 000 positive ANA without clinical relevance in Switzerland

Arbuckle et al, N Engl J Med. 2003 Oct 16;349(16):1526-33

ANA without specific symptoms

Patient characteristics	Symptoms	Lab results
• Female patient • Born 1986 • No underlying disease • None smoker • IT specialist	• Generalized pain • Headaches • Fatigue • Depression • Confirmed Sicca	• ANA 1:320 • CRP and BSR normal

ANA testing in patients with confirmed symptoms associated with CTDs does make sense

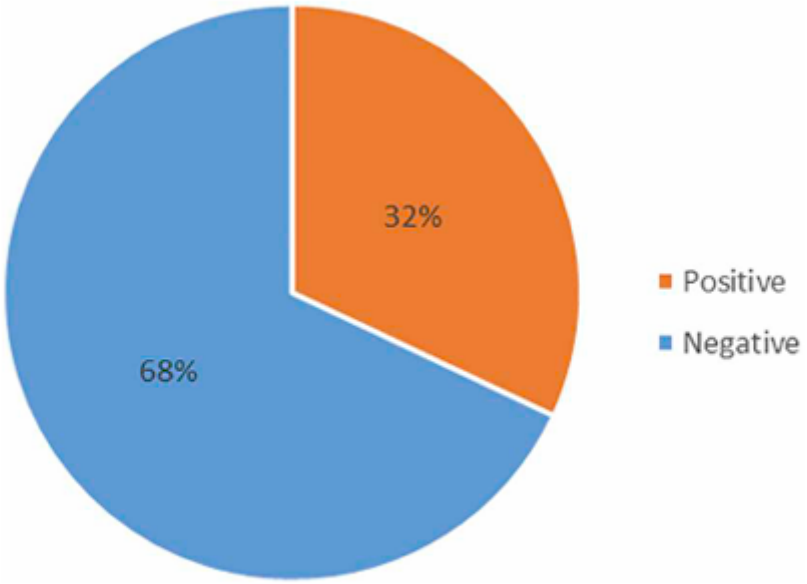


Fig. 1. About a third of FM patients tested positive for either early or classic SS antibodies when evaluated for all biomarkers.

Applbaum et al, Autoimmun Rev. 2019 Feb;18(2):199-202

Another example: Patients with Raynaud-Syndrome

1. **Labor: Autoantikörper (Antinukleäre Antikörper, dann Sklerodermie Antikörper)**

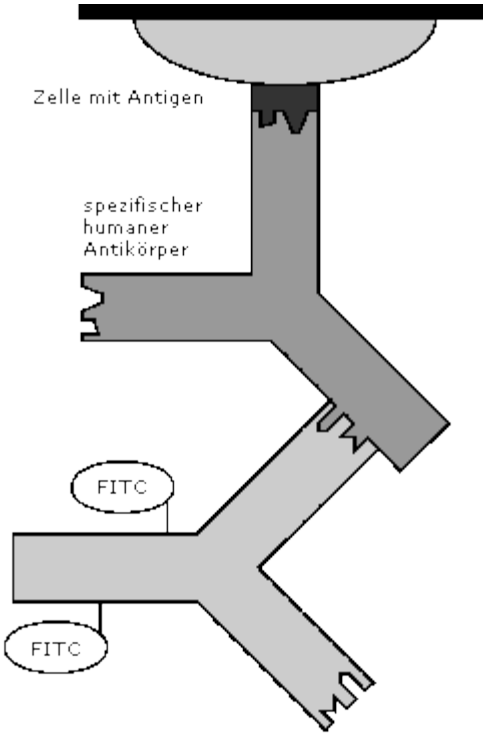
2. **Kapillarmikroskopie**

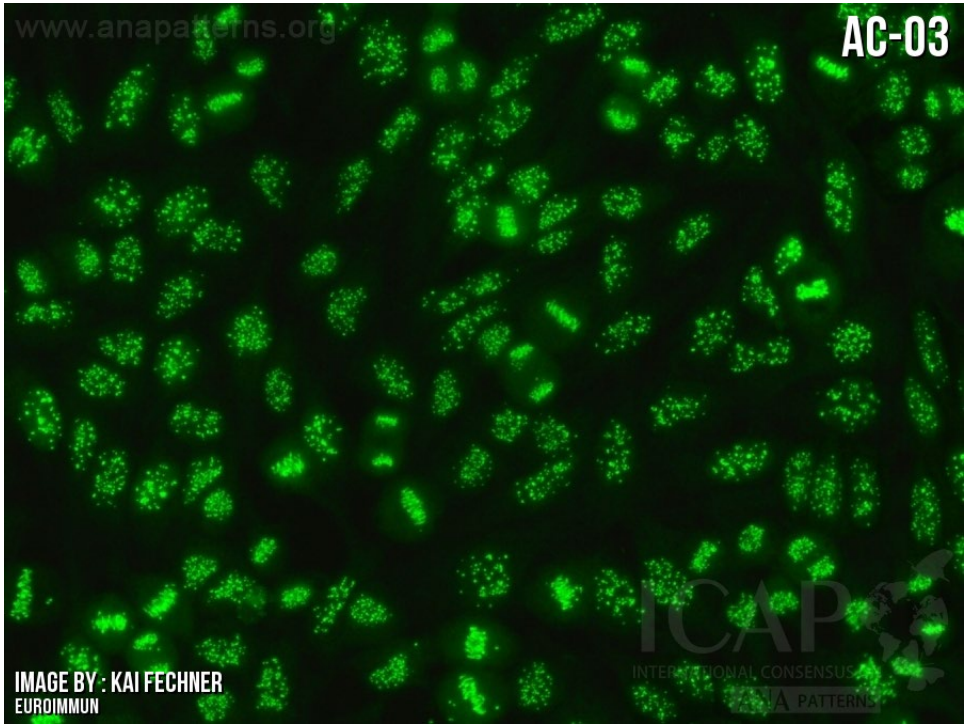
Wenn beide Tests negativ: Kollagenose (Sklerodermie) unwahrscheinlich

Wenn ein Test pathologisch: Wahrscheinlichkeit 25%

Wenn beide Tests pathologisch: Wahrscheinlichkeit 75%

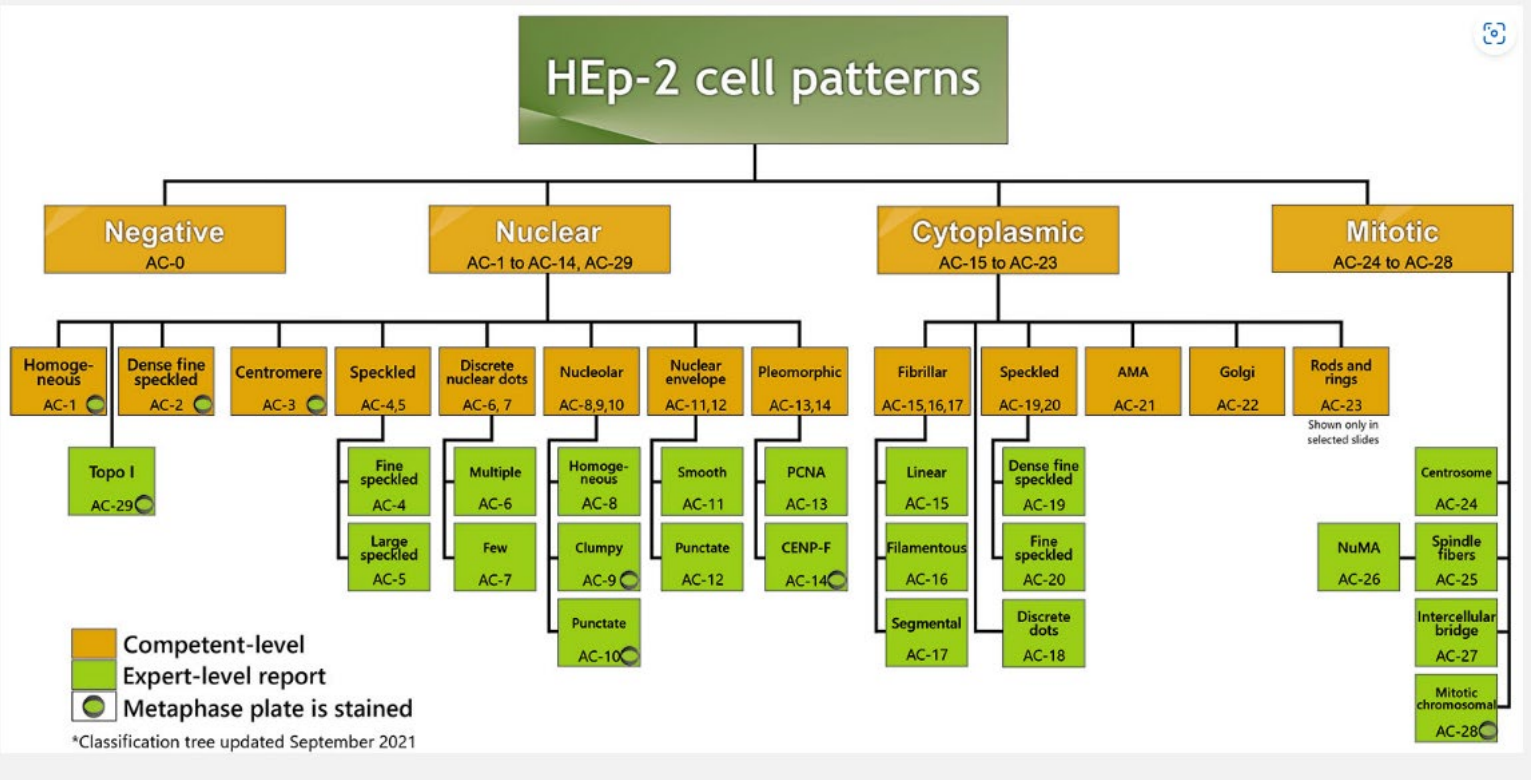
You found positive ANA in a patient with autoimmune symptoms. What next?





anti-centromer (AC-3) Muster

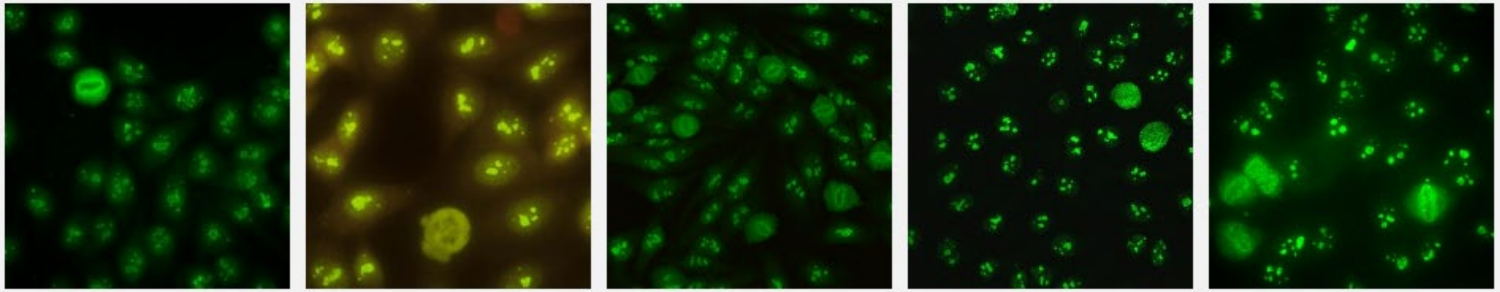
Consensus about ANA patterns: The AC classification



https://www.anapatterns.org/trees-2021.php

Example: The AC 9 pattern

AC-9 - Clumpy nucleolar



Previous Nomenclature	None
Description	Irregular staining of the nucleoli and Cajal bodies with a peri-chromosomal staining at the metaphase plates. e.g. anti-fibrillarin.
Antigen Association	U3-snoRNP/fibrillarin

Clinical Relevance

First level information

About Clinical Relevance & List of Abbreviations

- ▶ Found in patients with SSc (48)
- ▶ If SSc is clinically suspected, it is recommended to perform a follow-up test for anti-U3RNP/fibrillarin antibodies; the antigen is included in disease specific immunoassays (i.e., SSc profile*) (48)
- ▶ If confirmed as anti-U3RNP/fibrillarin reactivity by immunoassay, the clinical association is with diffuse SSc, increased incidence of pulmonary arterial hypertension, skeletal muscle disease, severe cardiac involvement, and gastrointestinal dysmotility (23, 48–50)
- ▶ Among SSc patients, anti-U3RNP/fibrillarin antibodies are most commonly found in African American and Latin American patients (48, 49, 51)

Consistency of results with different methodoloigies is important

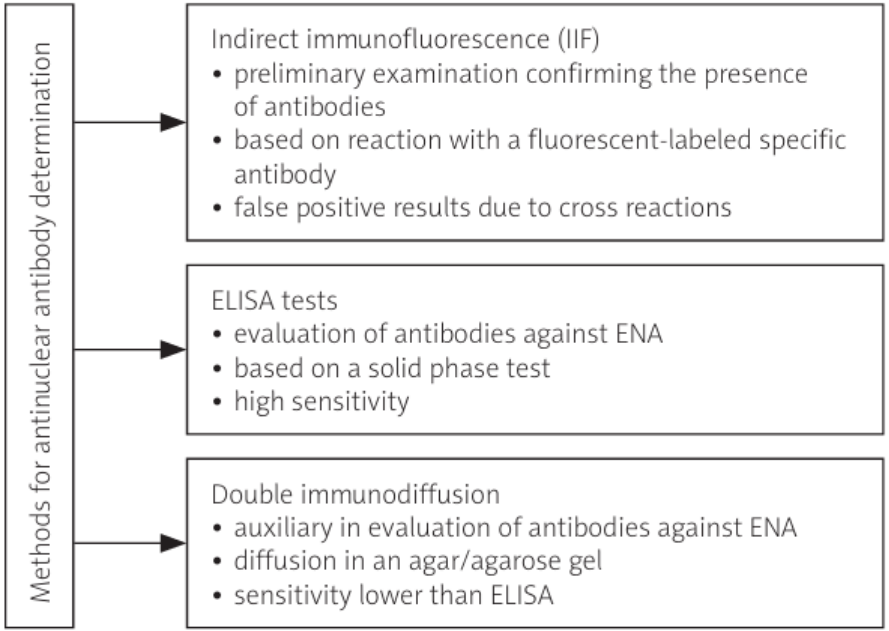
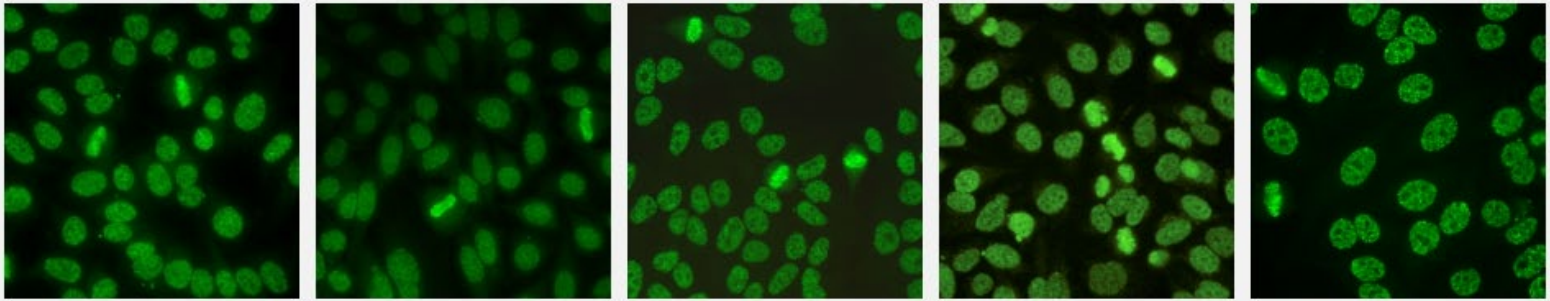


Fig. 1. Methods for antinuclear antibody determination.

Grygiel-Górniak et al, Reumatologia 2018; 56, 4: 243–248

The exception of DFS70 antibodies

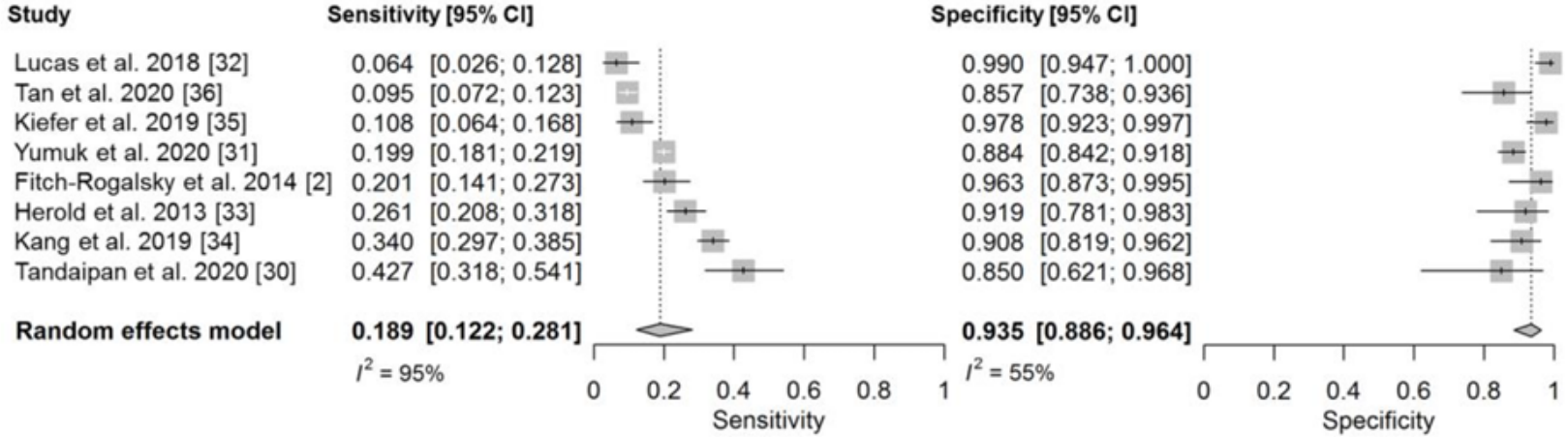
AC-2 - Nuclear dense fine speckled



Previous Nomenclature	None
Description	Speckled pattern distributed throughout the interphase nucleus with characteristic heterogeneity in the size, brightness and distribution of the speckles. Throughout the interphase nucleus, there are some denser and looser areas of speckles (very characteristic feature). The metaphase plate depicts strong speckled pattern with some coarse speckles standing out.
Antigen Association	DFS70/LEDGF

https://www.anapatterns.org/view_pattern.php?pattern=2

Anti-DFS 70 antibodes are negatively associated with autoimmune diseases



Cheng et al, Diagnostics (Basel). 2021 Sep 1;11(9):1592.

We have now confirmed ANA in a patient with autoimmune symptoms What next?

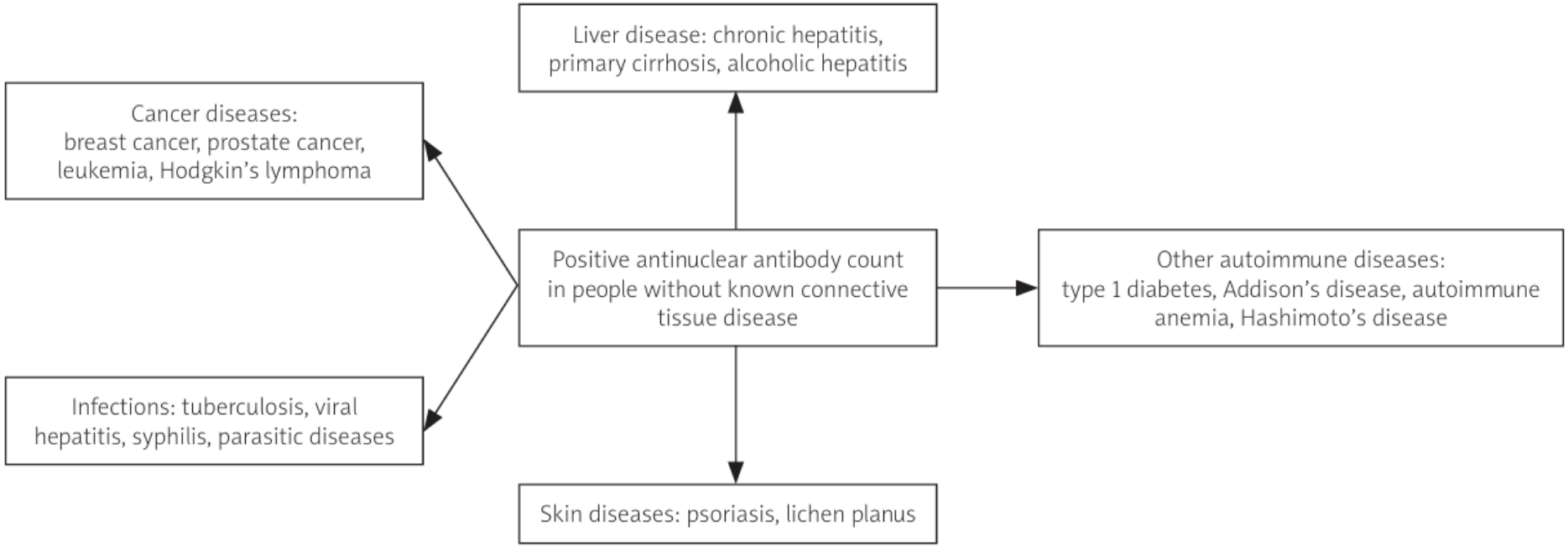


Fig. 2. The occurrence of antinuclear antibodies in patients without a diagnosed connective tissue disease.

Grygiel-Górniak et al, Reumatologia 2018; 56, 4: 243–248

53-jähriger Patient:

Zuweisung mit V.a. Anti-Ku-positive Polymyositis

Anamnese

- Knieschmerzen rechts seit April 2021
- Im Verlauf auch Schmerzen des rechten Oberschenkels
- Rechter Oberschenkel seither auch dünner und schwächer
- Rasches Ermüden der Oberarme, z.B., wenn er im Bett lese
- Ständig müde, müsse tagsüber schlafen gehen

- <u>Systemanamnese:</u>
- Seit 2 Wochen wiederholt Temperatur > 38°C
- Gewichtsverlust von 2 kg seit Beginn der Beschwerden

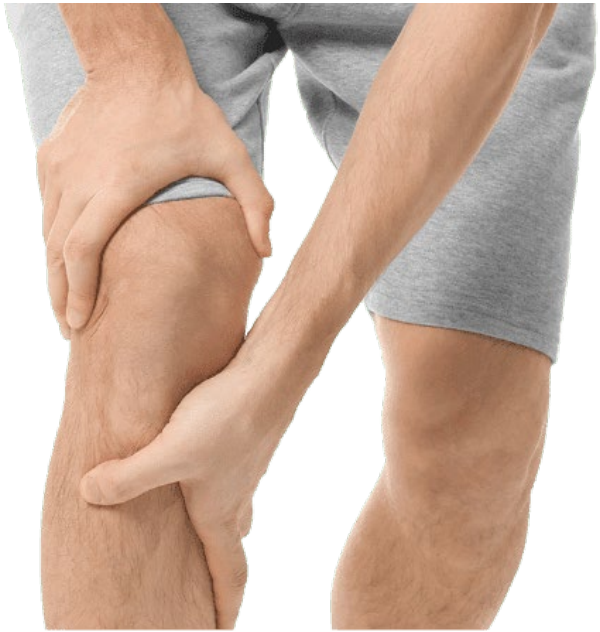


Bild: apmsbaytown.com

Status bei Eintritt

- Vitalparameter unauffällig
- Knie rechts: minim überwärmt, suprapatellär wenig Erguss
- Oberschenkel rechts: Umfang M. quadriceps femoris suprapatellär 45 cm (links: 46 cm)
- Ansonsten Normalbefund

Diagnostik: Labor

- CRP 29 mg/l, BSR 75 mm/h
- CK und Myoglobin normwertig
- ANA 1:640 (nuklear fein granulär, AC-4)
- Anti-SS-A / Anti-SS-B: negativ
- Anti-Ku (70 und 80): positiv (110)
- Erregerdiagnostik negativ (inkl. Blutkulturen, Quantiferontest, bakterielle Breitspektrum-PCR, Borrelien- und Brucella-Serologie, T. pallidum)

AUTOANTIKÖRPER			
SLE, MCTD, Sjögren-Synd…			
Antinukleäre AK	Titer	<1:320	* 1:640 (1)
ANA:Zytoplasma	Titer	<1:320	negativ
Anti-U1-snRNP (70)	U/ml	<10	0
Anti-SS-A (Ro/52+60kD)	E/ml	<10	0
Anti-SS-B (La,Ha)	E/ml	<10	0
Systemsklerose			
Anti-PM Scl 100		negativ	negativ
Anti-Ku (70 und 80)		negativ	** positiv
Anti-Ku 70, 80 quant		<15	* 110
Anti-Synthetase-Syndrom			
Anti-JO 1		neg.	negativ
Anti-PL7 (Synthetase)		neg.	negativ
Anti-PL12 (Synthetase)		neg.	negativ
Anti-EJ (Synthetase)		neg.	negativ
Myositis			
Anti-SRP		neg.	negativ
Anti-Mi2		neg.	negativ
Anti-MDA-5		neg.	negativ
Anti-TIF1 GAMMA		neg.	negativ
Anti-HMGCR		neg.	negativ
Anti-RO52		neg.	negativ
Anti-SAE1/2		neg.	negativ
Anti-NXP2		neg.	negativ (2)
Virale Infektionen			
HBs-Antigen		neg.	negativ
HBs-Antigen (qual.)	Quot.	<1.0	0.31 (3)
Anti-HBs		neg.	positiv (4)
Anti-HBs (quant)	IE/l	<10	* >1000
Anti-HBc-IgG/M		neg.	negativ
Anti-HBc-IgG/M quant	Quot.	>1.0	2.23 (5)
Anti-HCV (Screening)		neg.	negativ (6)
Anti-HCV-IgG (quant)	Quot.	<1.0	0.03 (7)
SARS-CoV-2 RNA (PCR)		negativ	
HIV und HTLV			
HIV Ag/Ak Comboscreen		neg.	negativ (8)
HIV Ag/Ak Combo quant	Quot.	<1.0	0.18
ZYTOKINE			
sIL-2 Rez. Blut	pg/ml	<477.0	

Legende: * Pathologischer Wert

Diagnostik: MRI

- Physiotherapeutisches Assesment der Muskelkraft und –ausdauer:

  Keine relevante Beeinträchtigung

- MRI Ganzkörper:

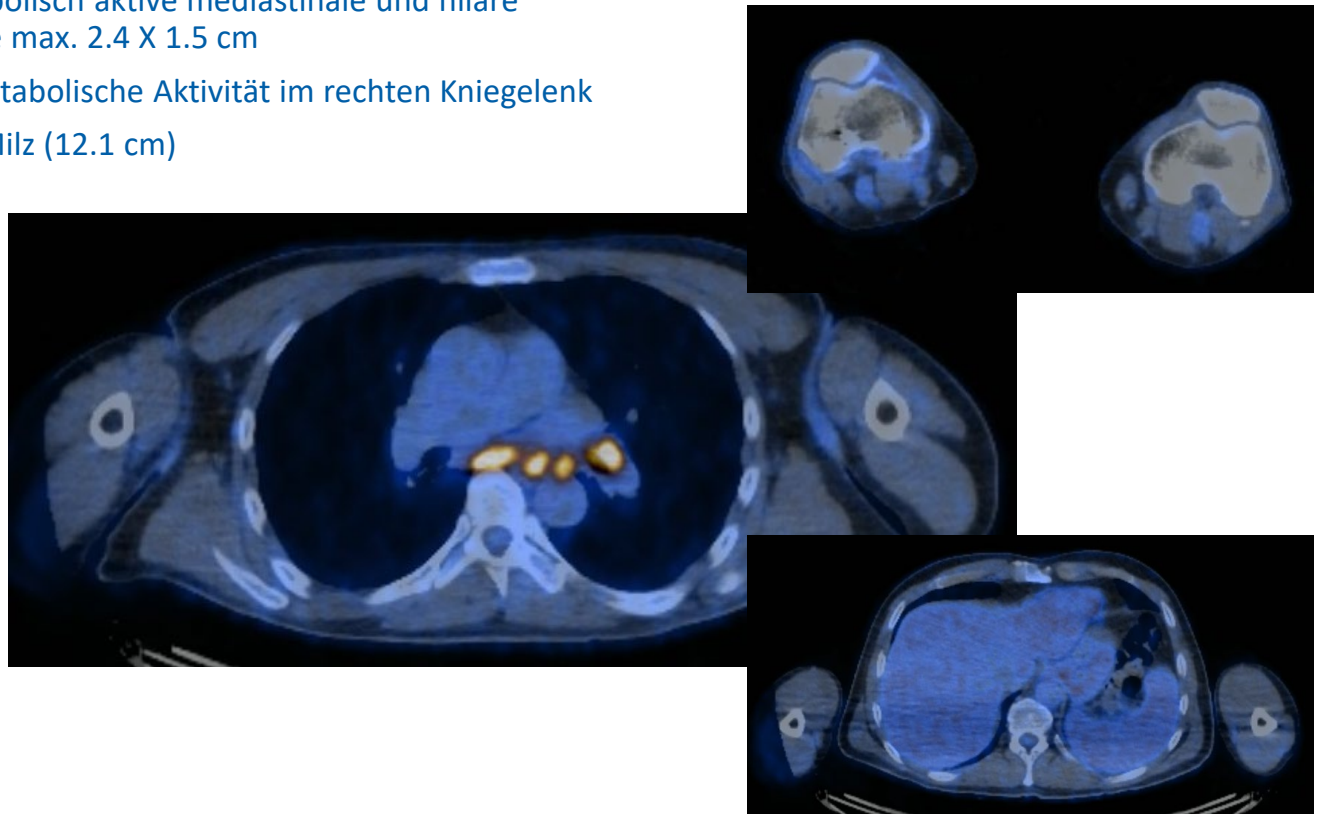
  Keine Hinweise auf Myositis oder Myopathie

- Synovialpunktion Kniegelenk rechts:

  Zellzahl 1740/µl, einzelne Calciumpyrophosphat-Kristalle, Mikrobiologie negativ (inkl. PCR B. burgdorferii / T. whippleii negativ, Mycobacterium DANN)

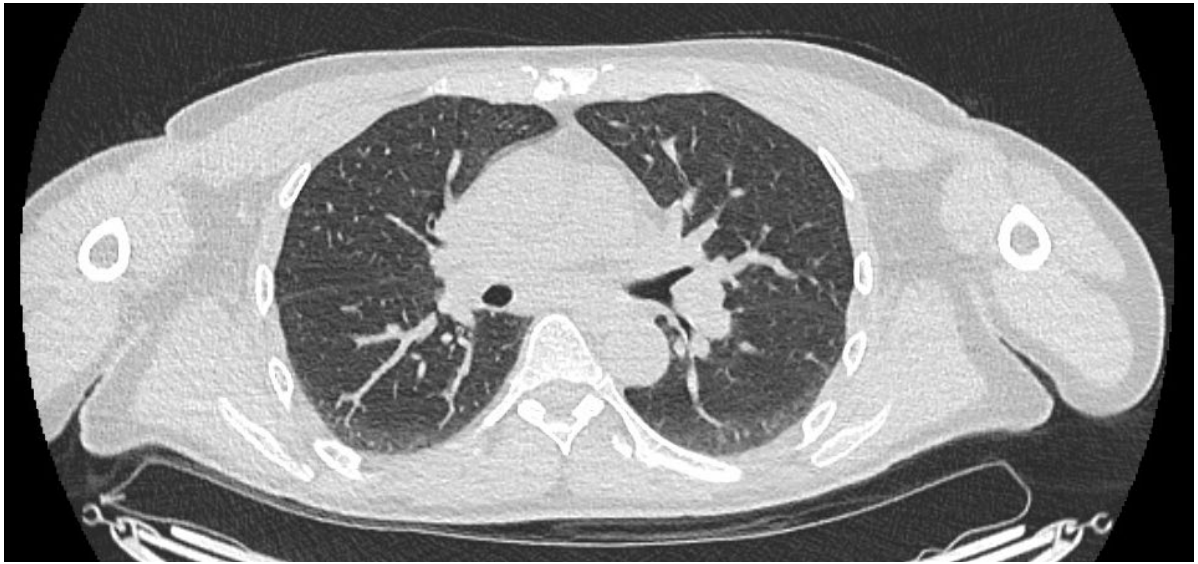
Diagnostik: 18F-FDG-PET-CT

- Mehrere, stark metabolisch aktive mediastinale und hiläre Lymphknoten, Grösse max. 2.4 X 1.5 cm
- Gering vermehrte metabolische Aktivität im rechten Kniegelenk
- Grenzwertig grosse Milz (12.1 cm)



Diagnostik: CT Thorax

- Mediastinale und hiläre Lymphadenopathie mit teils eingeschmolzenen Lymphknoten, primär verdächtig für Sarkoidose Stadium I
- Lungenparenchym unauffällig



Neue Differentialdiagnosen

- **Sarkoidose?**
- **Malignom? (a.e. Lymphom, da kein Primarius im PET-CT)**
- **Infektiös?**

Bild: pixabay.com

Diagnostik: Bronchoskopie

- BAL:

  Grampräparat, Kulturen, M. tuberculosis: negativ

  Pathologie: keine malignen Zellen

- Feinnadelpunktion Lymphknoten:

  Pathologie: Zellen der lymphatischen Reihe, keine malignen Zellen, keine Granulome oder Hodgkin-Zellen

…und jetzt?

- Zytologie nicht wegweisend
- Keine granulomatöse Entzündung als Hinweis für Sarkoidose
- Keine malignen Zellen als Hinweis für Lymphom
- Planung einer erneuten Bronchoskopie mit Materialentnahme mittels Minizange

Überraschende Wende

- Noch vor Durchführung der 2. Bronchoskopie Erhalt des folgenden (zuvor noch ausstehenden) Resultats:

13.10.2021, Beiliegend der Bericht von St.Gallen:

Francisella tularensis IgG (<10 U/ml): >300
Francisella tularensis IgM (<10 U/ml) : 201.7

Diskussion

- Befund mit Klinik passend zu pulmonaler Tularämie
- Erklärt Fehlen von Granulomen und malignen Zeller Zytologie
- PCR für F. tularensis aus Lymphknoten- oder Kniegelenkspunktat leider nicht mehr durchführbar
- Anti-Ku-Antikörper: ohne klinische Relevanz
- Therapie: Doxycyclin 100 mg 1-0-1 für 21 Tage
- Verlauf nach 21 Tagen: Normalisierung der Inflammationsparameter

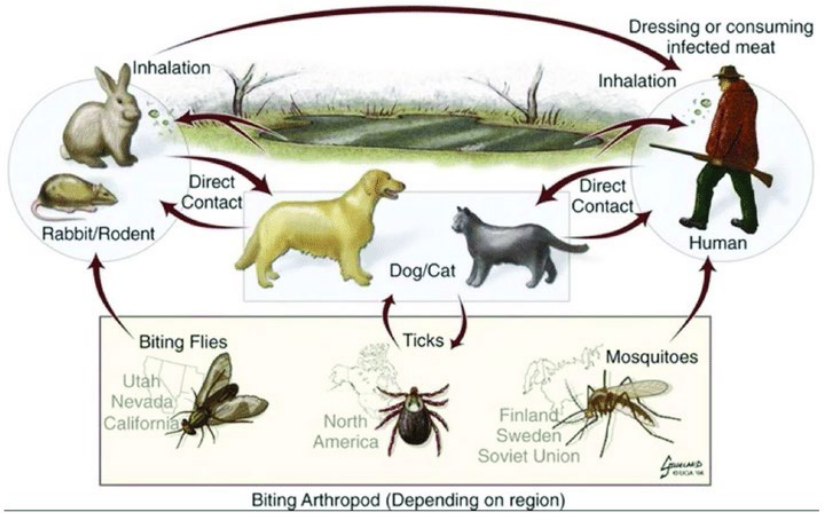


Bild: University of Georgia Research Foundation, Inc

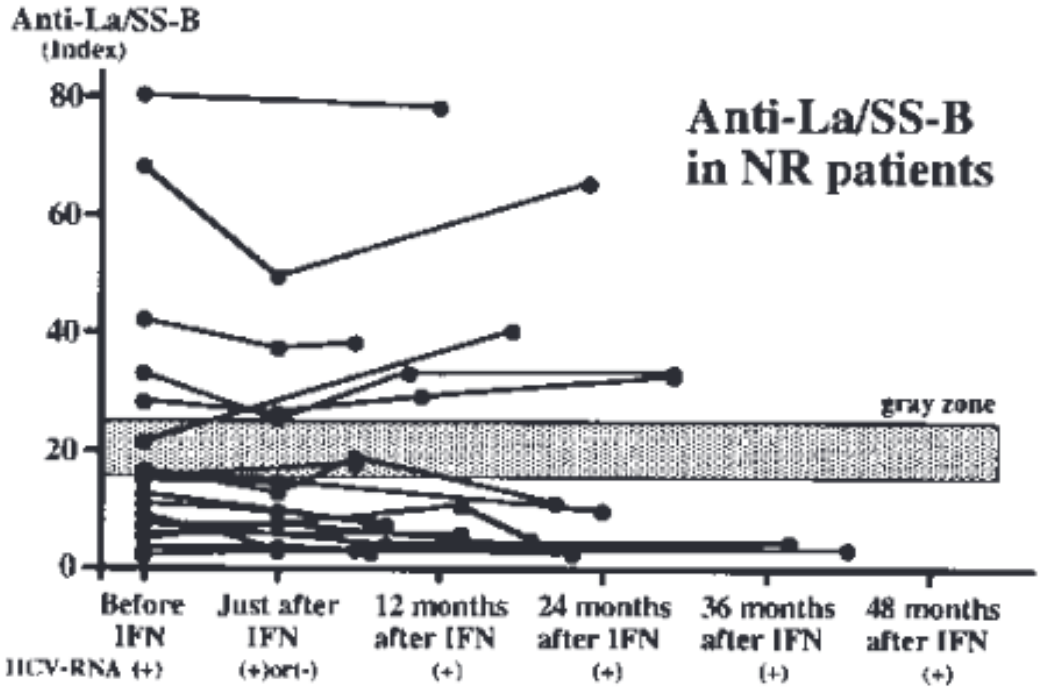
Hepatitis C: ANAs are prevalent and sometimes of high titers

Antibody	Chronic hepatitis C ($n = 44$)	Non-HCV infected liver diseases ($n = 44$)	p
Men/women	24/20	29/15	n.s.
Age (mean ± SD)	53 ± 14	46 ± 13	$p = 0.0175$
Anti-U1 RNP	2	1	n.s.
Anti-Sm	0	0	n.s.
Anti-Ro/SS-A	1	1	n.s.
Anti-La/SS-B	10	3	n.s.
Anti-Scl-70	5	2	n.s.
At least one of anti-ENAs	16 (37%)	7 (16%)	$p = 0.0290$

ENA, extractable nuclear antigens; HCV, hepatitis C virus; n.s., not significant.

Omagari et al, Autoimmunity. 2003 Aug;36(5):269-73.

Hepatitis C: ANAs are prevalent and sometimes of high titers



Omagari et al, Autoimmunity. 2003 Aug;36(5):269-73.

Even if you have confirmed ANA in a patient with autoimmune symptoms, keep your differential diagnosis in mind

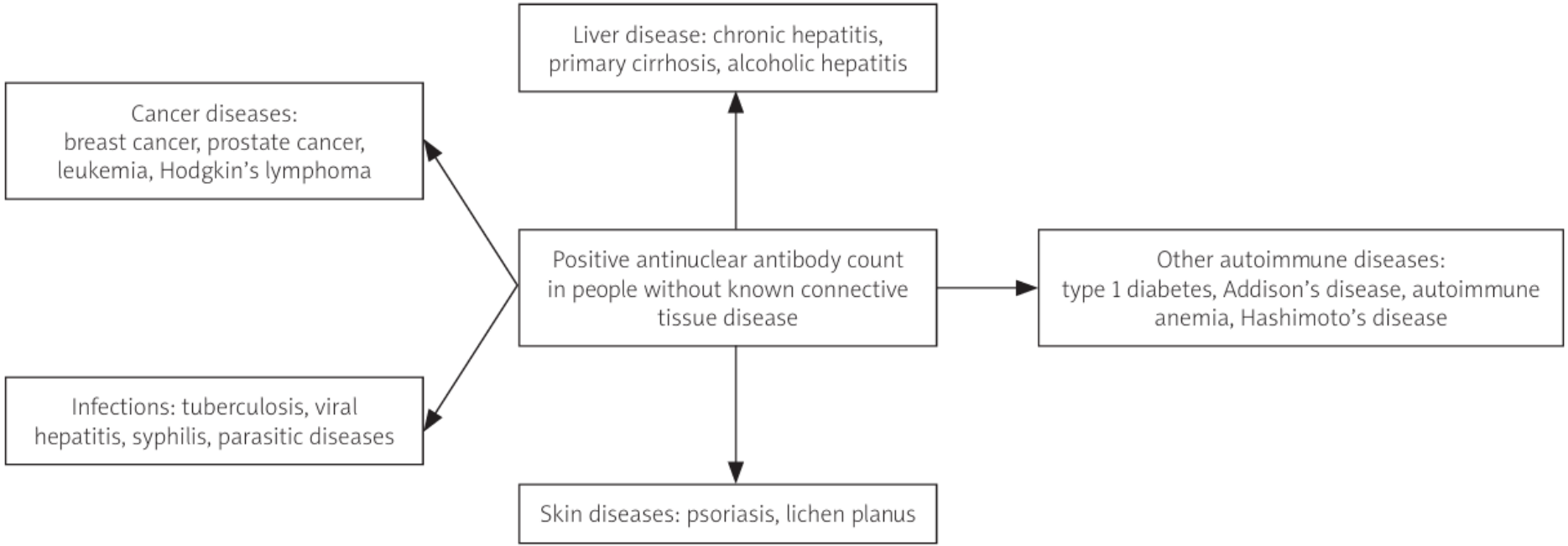


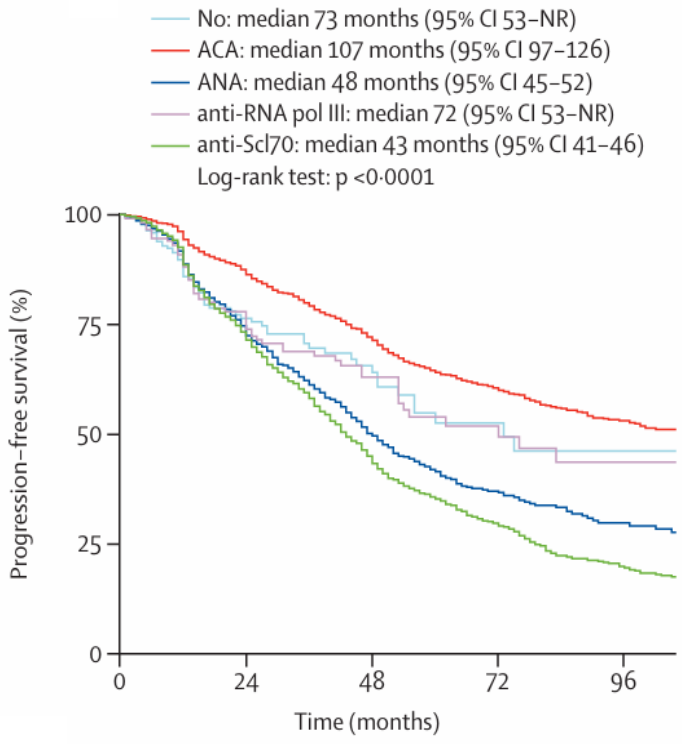
Fig. 2. The occurrence of antinuclear antibodies in patients without a diagnosed connective tissue disease.

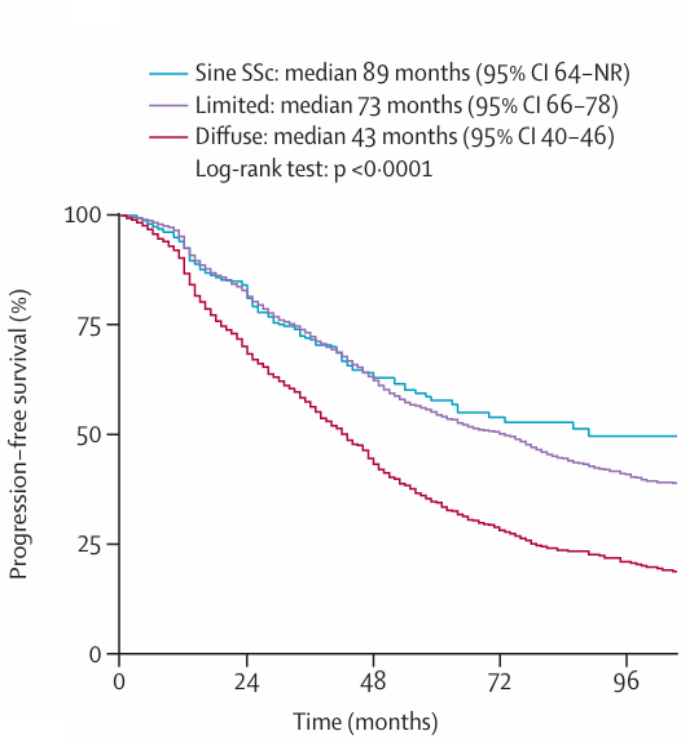
Grygiel-Górniak et al, Reumatologia 2018; 56, 4: 243–248

Antibodies are informative about clinical phenotypes

- SSc with anti-centromere antibodies
- SSc with anti-topoisomerase antibodies
- SSc with anti RNA-Polymerase 3 antibodies

Antibodies are better for risk-stratification in SSc than clinical subclassification





Elhai et al, Lancet Rheumatology 2022, 4(11):e785-e794.

U1-RNP antibodies are associated with a unique clinical phenotype in SSc

	Missing cases n (%)	Whole cohort (n=8391)	Anti-u1RNP positive (n=408)	Anti-u1RNP negative (n=7983)	p value
Demographic					
Age at visit (year), mean±SD	2 (0.0)	55.8±14.0	48.0±14.3	56.2±13.9	**<0.001**
Age at first non-RP (year), mean±SD	1055 (12.6)	46.2±14.4	37.9±13.9	46.6±14.3	**<0.001**
Male sex, n (%)	0 (0.0)	1263 (15.1)	59 (14.5)	1204 (15.1)	0.732
Skin					
Diffuse subset, n (%)	103 (1.2)	2549 (30.8)	126 (31.2)	2423 (30.7)	0.847
MRSS (unit), mean±SD	960 (11.4)	8.5±8.0	7.9±8.5	8.6±7.9	**0.004**
Musculoskeletal					
Joint synovitis, n (%)	85 (1.0)	1139 (13.7)	77 (19.3)	1062 (13.4)	**0.001**
Muscle weakness, n (%)	111 (1.3)	1415 (17.1)	81 (20.4)	1334 (16.9)	0.076
Muscle atrophy, n (%)	146 (1.7)	682 (8.3)	47 (11.8)	635 (8.1)	**0.008**
CK elevation, n (%)	876 (10.4)	615 (8.2)	58 (16.1)	557 (7.8)	**<0.001**

Wu et al, EUSTAR database, unpublished

U1-RNP antibodies are associated with a unique clinical phenotype in SSc

	Missing cases n (%)	Whole cohort (n=8391)	Anti-u1RNP positive (n=408)	Anti-u1RNP negative (n=7983)	p value
Cardiopulmonary					
LVEF <50%, n (%)	2149 (25.6)	175 (2.8)	10 (3.3)	165 (2.8)	0.591
Pulmonary hypertension on Echo, n (%)	1161 (13.8)	1177 (16.3)	83 (24.7)	1094 (15.9)	**<0.001**
Lung fibrosis on chest X-ray or HRCT, n (%)	1002 (11.9)	3146 (42.6)	187 (53.1)	2959 (42.0)	**<0.001**
FVC (% predicted), mean±SD	1614 (19.2)	94.6±22.0	86.9±20.4	95.0±22.0	**<0.001**
FVC <80% predicted, n (%)	1614 (19.2)	1645 (24.3)	110 (33.6)	1535 (23.8)	**<0.001**
DLCO (% predicted), mean±SD	1690 (20.1)	68.2±20.3	63.4±18.9	68.4±20.3	**<0.001**
DLCO <80% predicted, n (%)	1690 (20.1)	4767 (71.1)	247 (81.3)	4520 (70.7)	**<0.001**
Inflammation					
ESR >25mm/h, n (%)	1154 (13.8)	2327 (32.2)	159 (46.4)	2168 (31.4)	**<0.001**
CRP elevation, n (%)	484 (5.8)	1788 (22.6)	101 (26.6)	1687 (22.4)	0.058
Hypocomplementemia, n (%)	1755 (20.9)	496 (7.5)	51 (15.7)	445 (7.1)	**<0.001**
Active disease (VAI≥3), n (%)	0 (0.0)	1672 (19.9)	105 (25.7)	1567 (19.6)	**0.003**

Wu et al, EUSTAR database, unpublished

Conclusions

- Avoid ANA testing in patients with no or unspecific symptoms
- Consistency of ANAs across methodologies
- Never forget differential diagnosis of increased ANAs
- Once autoimmune disease diagnosed antibodies are helping in stratification