

Rheumatoid arthritis

Laboratory investigations:

1. Rheumatoid factor

- Antibodies against the Fc portion of IgG.
- Usually measured by rate nephelometry.
- Part of diagnostic criteria for rheumatoid arthritis.

• 80% specific.

• Also positive in:

- viral infections or persistent infections e.g. osteomyelitis, TB.
- other rheumatic diseases
- healthy people, especially the elderly.

3. Anti-CCP antibodies

(anti-cyclic citrullinated peptide antibodies)

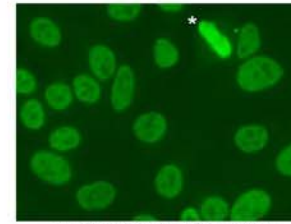
- Antibodies to various citrullinated proteins associated with rheumatoid arthritis e.g. fibrinogen, vimentin.
- Peptidyl arginine deiminase (PAD) enzymes up-regulated in inflamed joint deiminate arginine to citrulline.
- Measured by ELISA, using a commercial preparation of cyclic citrullinated peptides to mimic in vivo targets.
- Specific for rheumatoid arthritis.
- Useful when rheumatoid factor does not correlate with clinical picture.
- Presence of both rheumatoid factor and anti-CCP antibodies highly specific for rheumatoid arthritis and may predict aggressive disease.
- May be useful for prediction of rheumatoid arthritis in the future.

Symptoms:

- Arthritis (inflammation) in small joints e.g. of hands and feet
- Early morning stiffness lasting at least 1h.
- Soft tissue swelling and rheumatoid nodules
- Can also affect blood vessels (small vessel vasculitis)
 - heart (pericarditis)
 - lungs (interstitial fibrosis)
 - eyes (scleritis)

2. CRP

- Acute phase protein produced by the liver.
- Increased in bacterial infections and in inflammation, including in rheumatoid arthritis.
- Concentration in active rheumatoid arthritis typically 30-80mg/L.



Homogeneous staining in nuclei (excluding nucleoli), positive staining in chromosomes of dividing cells*.

Symptoms:

- Highly variable.
- Classically arthritis, mouth ulcers, photosensitive butterfly rash across the cheeks.
- May also have neurological, renal, haematological symptoms.
- Important to monitor renal function.

Laboratory investigations:

- Measure C3 and C4 concentrations, and antinuclear antibodies.
 - **Low C4 concentration**
- Indicates active immune complex disease (due to activation of classical pathway – see Immune Deficiency poster). C2 and C4 deficiency increase risk of developing SLE (rare)
- Positive **antinuclear antibodies**
- Homogeneous pattern and anti-dsDNA antibodies
- OR
- Speckled pattern and anti-Sm antibodies
- CRP not raised due to SLE but will increase with infection

RHEUMATOLOGY

Systemic lupus erythematosus

A systemic rheumatological immune complex disease. Thought to be due to defective clearance of apoptotic cell debris. Also associated with Sjogren's syndrome and anti-phospholipid syndrome.

Antinuclear antibodies

- Includes antibodies to **double stranded DNA (dsDNA)** and **extractable nuclear antigens (ENA)**.
- Detected with a screening assay using indirect immunofluorescence on HEp2 (human epithelioma) cells.
- HEp2 cells have large nuclei and lots of dividing cells, allowing clear interpretation of staining pattern due to patient IgG binding to cellular antigens.
- Appropriate follow-on tests (ELISAs) are done, based on the pattern of staining in nuclei and chromosomes of dividing cells.
- Many patterns are recognized but not all are clinically relevant.

Anti-dsDNA antibodies

- **Pattern = homogeneous, chromosomes staining in dividing cells.**
- Present in 60% of patients with SLE.
- An increase in concentration may precede a relapse.
- Antibodies to ssDNA give the same pattern, may give false positive results on dsDNA ELISA and are found in SLE but also viral infections. Not clinically specific.
- **Crithidia** test (indirect immunofluorescence on Crithidia, a flagellate protozoan) is used to distinguish between antibodies to ssDNA and dsDNA where results are queried, but are not sensitive enough to be used as a screening test.

Anti-ENA antibodies

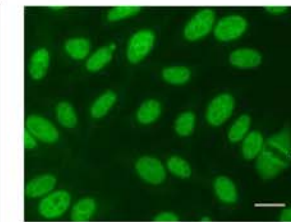
- **Pattern = speckled, chromosomes negative in dividing cells.**
- Clinically relevant ENA include Ro (SSA), La (SSB), Sm and RNP.
- Antibodies to Ro and La are found in SLE and Sjogren's syndrome, and can cross the placenta to cause neonatal heartblock.
- Antibodies to Sm are found in 10% of patients with SLE.
- Antibodies to RNP are found in mixed connective tissue disease.



Crithidia, showing staining in nucleus and kinetophore (two circles)

Sjogren's syndrome

- Sicca syndrome (clinically measurable dry eyes and dry mouth), maybe with arthritis.
- Associated with positive antinuclear antibodies – speckled pattern with anti-Ro and anti-La antibodies.
- Important in pregnancy – these antibodies can cross the placenta and cause neonatal heart block.



Speckled staining pattern in nuclei, no staining in chromosomes of dividing cells*.

Other laboratory investigations

In many rheumatological diseases, immunoglobulins are polyclonally raised. This is a non-specific finding that reflects persistent inflammation. It is of no use in diagnosis, prognosis or monitoring disease.

Antiphospholipid syndrome

- Various symptoms caused by abnormal clotting e.g. deep vein thrombosis, recurrent miscarriages, stroke etc.
- Have raised INR that doesn't correct with normal plasma.
- Associated with antibodies to phospholipids e.g. cardiolipin, and to proteins associated with phospholipids e.g. β_2 -glycoprotein I.
- Also have positive lupus anticoagulant (patient plasma inhibits in vitro coagulation assays).

Systemic sclerosis

- A rare chronic multisystem connective tissue disease.
- Caused by progressive fibrosis of skin and visceral organs.
- Associated with various rare autoantibodies
 - anti-centromere (target = CENP proteins)
 - anti-RNA polymerase (associated with renal crisis)
 - anti-nucleolar (target = fibrillar)in)
 - anti-SCL70 (target = topoisomerase I)
 - others
- Each patient only has one type of autoantibody.
- May have limited or diffuse skin symptoms, and also affects various organs of the body (see also Renal poster)