Systemic Lupus Erythematosus
An Insight

Dr Lal PathLabs

Introduction

- **Systemic lupus erythematosus, often abbreviated as SLE or lupus**
- **Systemic autoimmune disease in which the body's immune system mistakenly attacks healthy tissue.**
- **SLE most often harms the heart, joints, skin, lungs, blood vessels, liver, kidneys, and nervous system.**
- **The course of the disease is unpredictable, with periods of illness (called *flares*) alternating with remissions.**

History

- ✓ Lupus means Wolf in latin
- ✓ 1846- Ferdinand von Hebra used the term Lupus Erythematosus
- ✓ Moriz Kaposi first recognised Lupus as Systemic disease
- ✓ 1948 – Malcolm Hargraves discovers the lupus erythematosus (LE) cell.
- ✓ 1957 – The first anti-DNA antibody was identified.

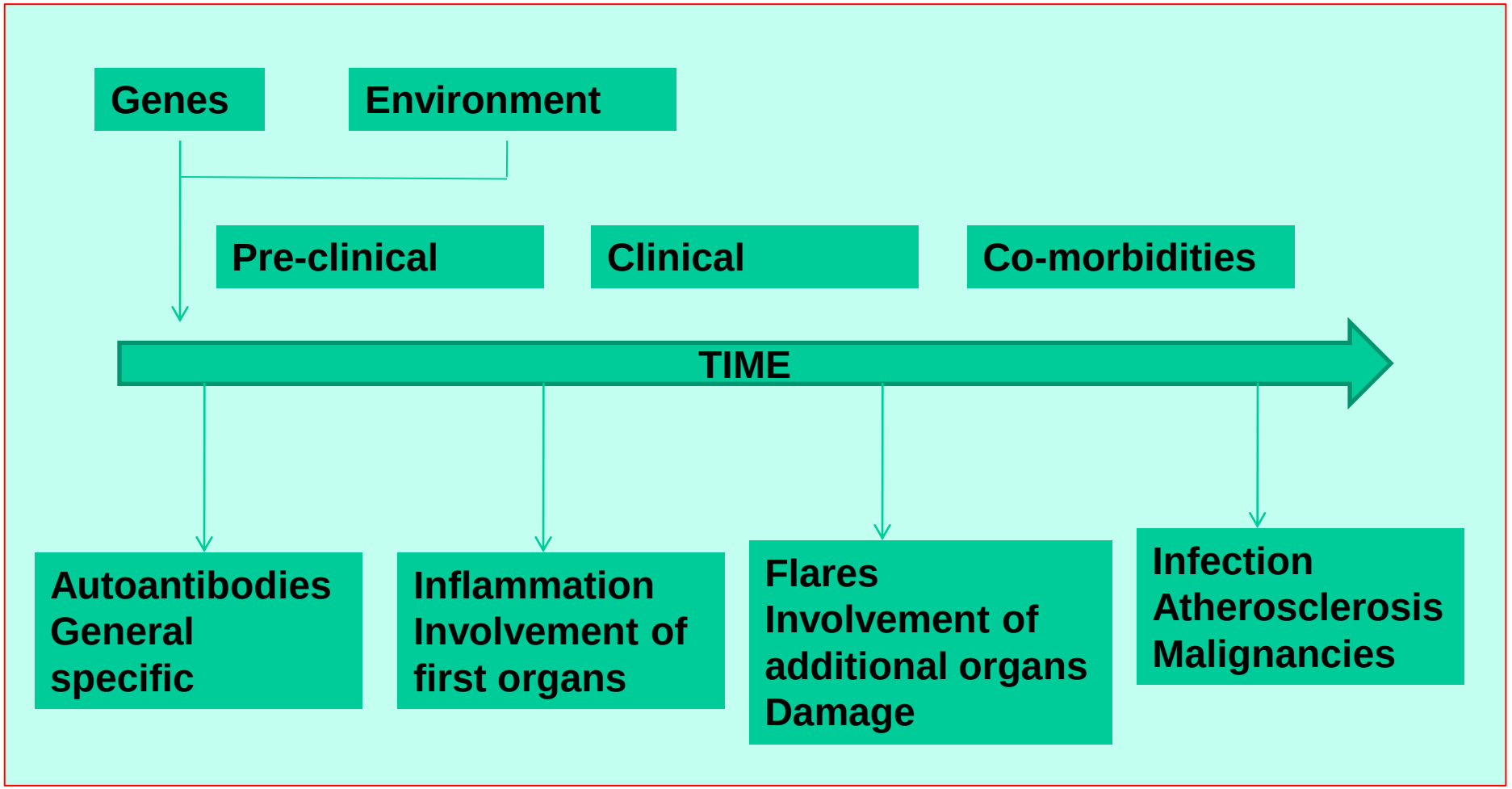
Prevalence

- **Global rates of disease varies from 20 to 70 per 100,000.**
- **The disease is more common in women (♀ : ♂ = 9:1), especially in women of ages 15 to 35 years (child-bearing age group).**
- **More common in those of African-American or Caribbean descent.**
- **Appears to be more common in urban than in rural areas.**

- **The incidence of Lupus has nearly doubled in the last 40 years.**

- **Childhood SLE usually presents between the ages of 3 and 15, with girls to boys ratio 4:1 with typical skin manifestations being a butterfly-shaped rash on the face and photosensitivity.**

Natural History and course



Aetiology

- Genetic factors
- Epigenetic effects
- Environmental factors
- Hormonal factors

Genetic factors

- Siblings of SLE patients are approximately 30 times more likely to develop SLE compared with individuals without an affected sibling.
- HLA-DR2
- HLA-DR3
- HLA-DQB1
- These are involved in mediating production of antibodies to ds-DNA.

Epigenetic effects

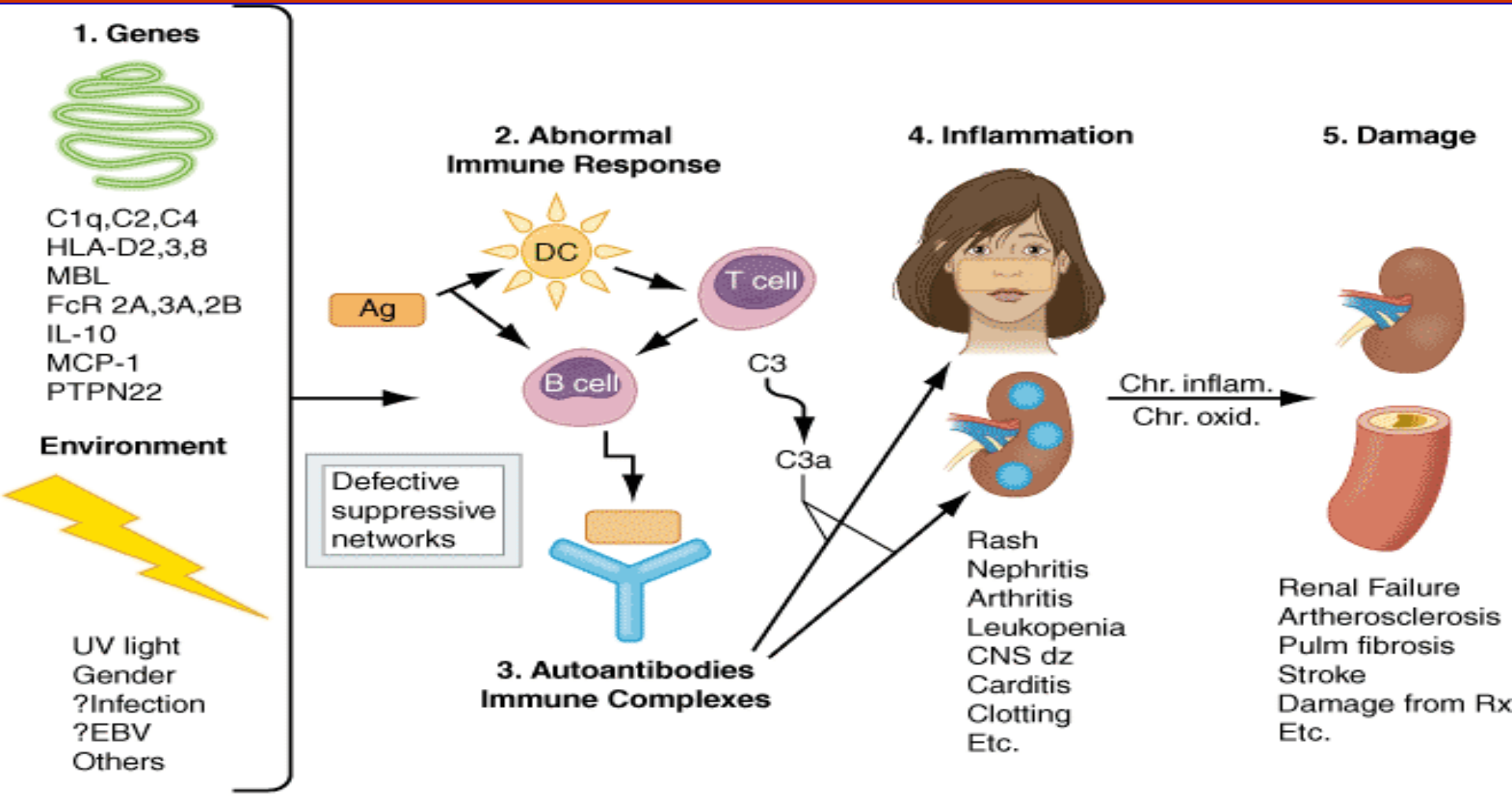
- The risk of SLE may be influenced by epigenetic effects such as DNA methylation and post-translational modifications of histones, which can be either inherited or environmentally modified.

- Epigenetic refers to inherited changes in gene expression caused by mechanisms other than DNA base sequence changes.

Environmental factors

- Ultraviolet light
- Demethylating drugs
- Infectious or endogenous viruses or viral like elements (Epstein-Barr virus)
- Sunlight may exacerbate SLE

Pathogenesis & Pathophysiology of SLE



Source: Fauci AS, Kasper DL, Braunwald E, Hauser SL, Longo DL, Jameson JL, Loscalzo J: *Harrison's Principles of Internal Medicine*, 17th Edition: http://www.accessmedicine.com

- One manifestation of SLE is abnormalities in apoptosis, a type of programmed cell death in which aging or damaged cells are neatly disposed of as a part of normal growth or functioning.
- In SLE, the body's immune system produces antibodies against itself, particularly against proteins in the cell nucleus.
- SLE is triggered by environmental factors that are unknown.

- **The immune system balances between being sensitive to protect against infection, and becoming sensitized to attack the body's own proteins (autoimmunity).**

- **During an immune reaction to a foreign stimulus, (bacteria, virus, or allergen), immune cells that would normally be deactivated due to their affinity for self tissues can be abnormally activated by signaling sequences of antigen-presenting cells.**

- **Thus triggers may include viruses, bacteria, allergens (IgE and other hypersensitivity), and can be aggravated by environmental stimulants such as ultraviolet light and certain drug reactions.**

- **These stimuli begin a reaction that leads to destruction of other cells in the body and exposure of their DNA, histones, and other proteins, particularly parts of the cell nucleus.**

- The body's sensitized B-lymphocyte cells will now produce antibodies against these nuclear-related proteins.
- These antibodies clump into antibody-protein complexes which stick to surfaces and damage blood vessels in critical areas of the body, such as the glomeruli of the kidney; these antibody attacks are the cause of SLE.
- Researchers are now identifying the individual genes, the proteins they produce, and their role in the immune system. Each protein is a link on the autoimmune chain, and researchers are trying to find drugs to break each of those links

- **SLE is a chronic inflammatory disease believed to be a type III hypersensitivity response with potential type II involvement.**

- **Reticulate and stellate acral pigmentation should be considered a possible manifestation of SLE and high titers of anti-cardiolipin antibodies, or a consequence of therapy.**

Types of Lupus

Systemic Lupus Erythematosus (SLE):

- SLE is the most common type of lupus.
- According to the Centres for Disease Control and Prevention, the incidence is difficult to establish due to vague signs & symptoms and onset.
- Ninety percent of newly diagnosed patients are women of child-bearing age. It affects many parts of the body (systemic).

Discoid lupus:

Characterized by a chronic skin rash, such as on the face or scalp.

Subacute Cutaneous Lupus:

Associated with skin lesions on parts of the body that are exposed to sunlight.

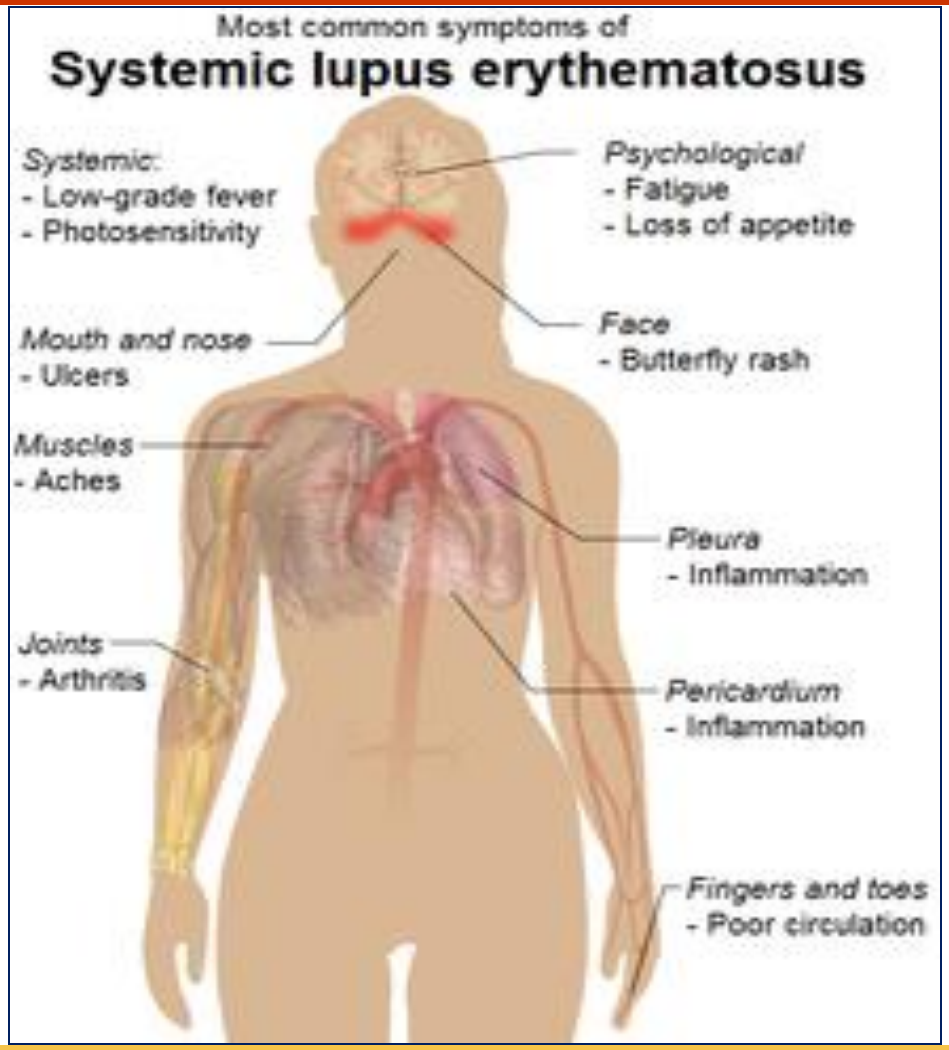
Drug-induced lupus:

- A form of lupus that can be caused by certain medications, such as some anti-seizure, high blood pressure, and anti-thyroid medications.
- The most common medications known to cause drug-induced lupus are the antibiotic isoniazid, used to treat and prevent tuberculosis, hydralazine, used to treat hypertension, and procainamide, used to treat abnormal heart rhythms.
- Symptoms tend to occur after taking the medication for several months and usually resolve once the medication is stopped.

Neonatal lupus:

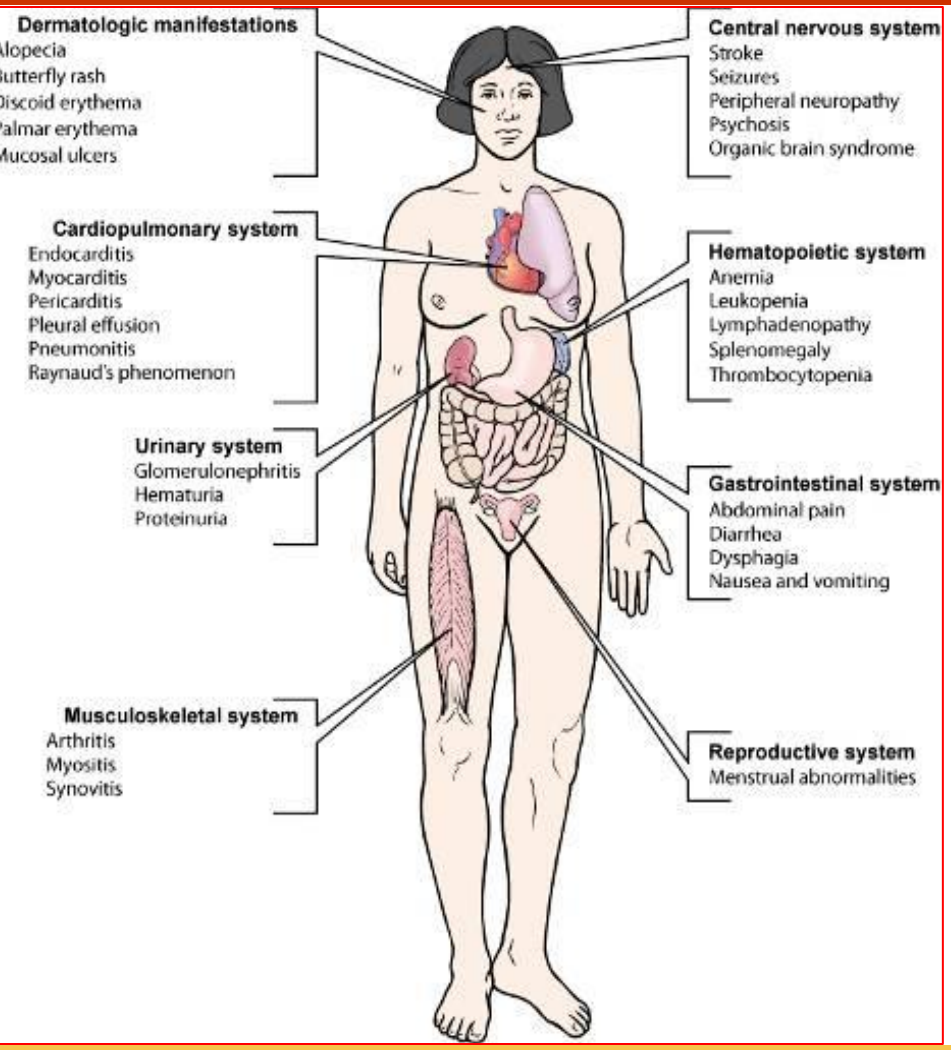
- A rare form of lupus that affects newborns and that is characterized by a skin rash, liver problems, and low blood counts at birth.
- These usually resolve over several months. Newborns who have neonatal lupus may be born to women who have SLE, Sjögren syndrome, or no particular disease, but it is thought that it may be triggered in part by certain autoantibodies in the mother's blood (anti-SSA and anti-SSB).
- Women known to have these autoantibodies may be monitored more closely during their pregnancy.

Signs and Symptoms



- Ranges from a relatively mild disorder to rapidly progressing, affecting many body systems
- Most commonly affects the skin/muscles, lining of lungs, heart, nervous tissue, and kidneys

General S/S



Suggestive S/S

- **Fever,**
- **Malaise,**
- **Joint pains,**
- **Muscle aches,**
- **Fatigue**

Skin

- ✓ **Discoid lupus may exhibit thick, red scaly patches on the skin.**

- ✓ **Subacute cutaneous lupus manifests as red, scaly patches of skin but with distinct edges.**

- ✓ **Acute cutaneous lupus manifests as a rash. Some have the classic malar rash (or *butterfly rash*) associated with the disease. This rash occurs in 30 to 60% of people.**

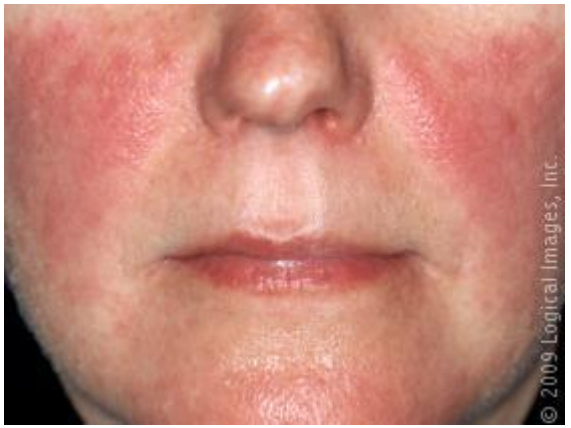


Table 1:Classification of LE associated Skin lesions

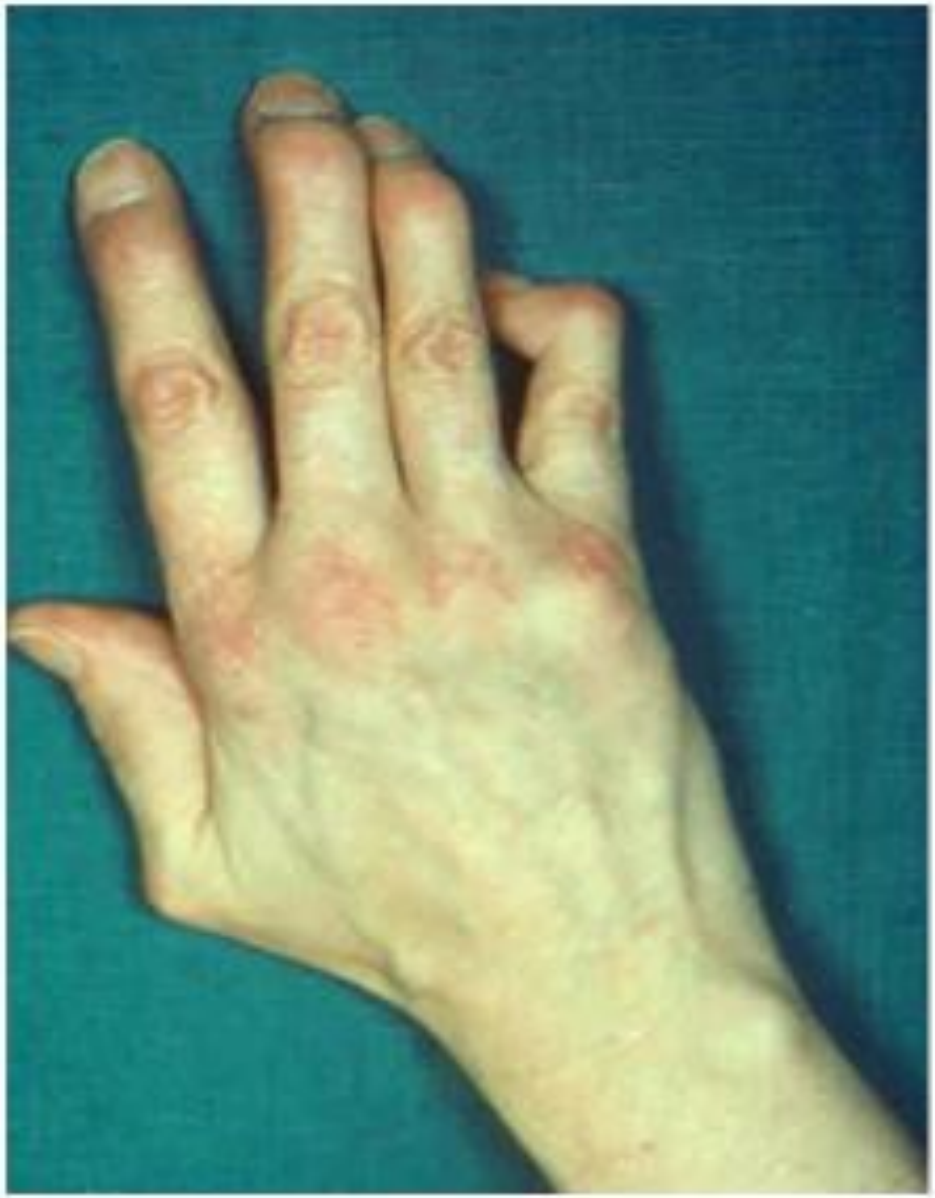
LE specific skin lesions	LE non-specific skin lesions
Acute cutaneous LE Localised	Cutaneous vascular disease vasculitis
Generalised Subacute cutaneous LE	Leucocytoclastic Palpable purpura Urticarial vasculitis
Annular papulosquamous (Psoariasiform)	Polyarteritis nodosa like Papulonodular mucinosis Dego's disease like
Chronic cutaneous LE Classical LE (DLE) Localised	Atrophy blanche-like Livedo reticularis Thrombophlebitis
Generalised Hypertrophic (verrucous) DLELupus panniculitis (profundus)	Reynaud's phenomenon Erythromelalgia LE non specific bullous lesions
Mucosal LE Lupus tumidus Chilblains lupus	Epidermolysis bullosa acquisita Dermatitis herpetiformis like bullous LE Pemphigus erythematosus

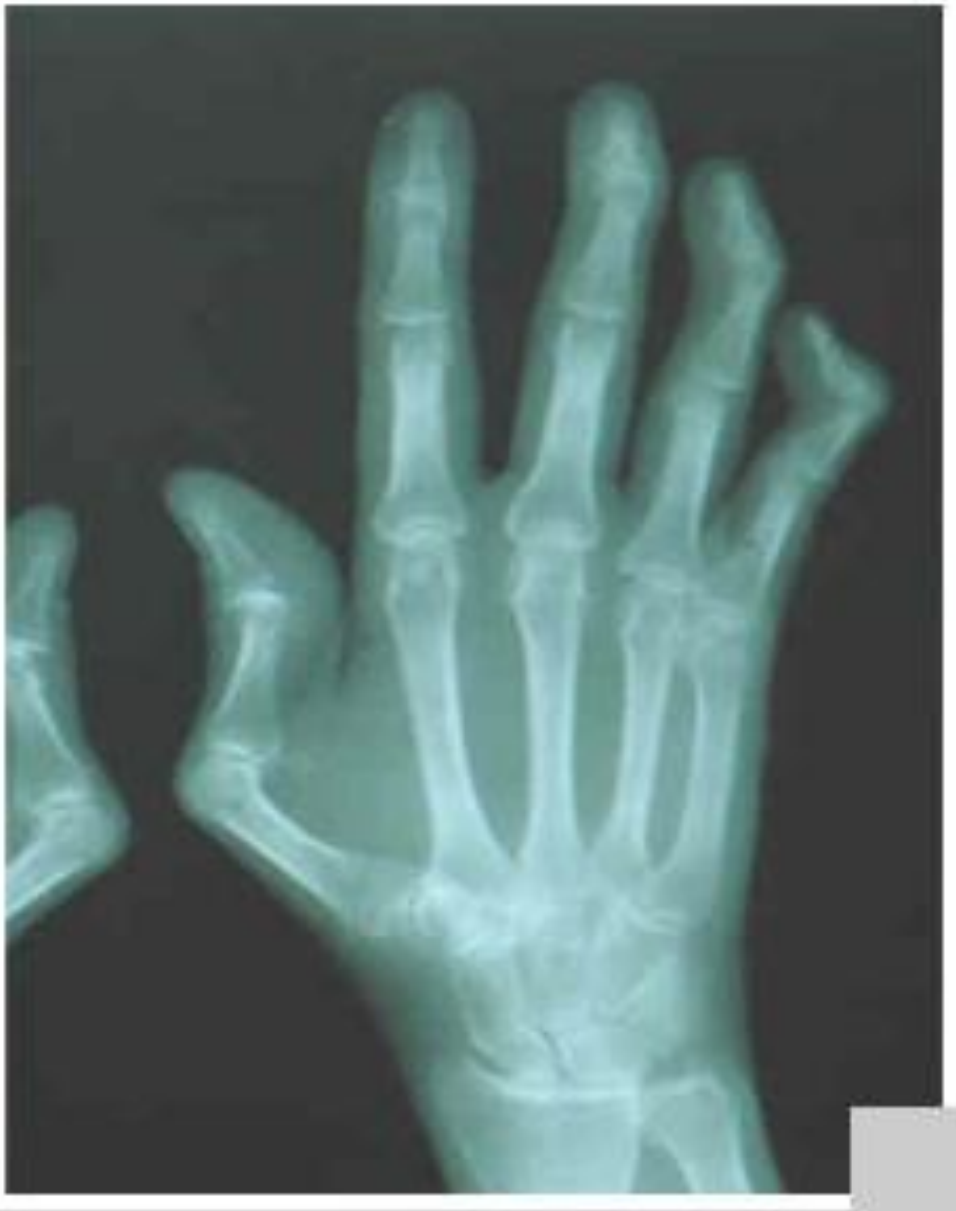
Table 1:Classification of LE associated Skin lesions

LE specific skin lesions	LE non-specific skin lesions
	Porphyria cutanea tarda Urticaria Vasculopathy
	Anetoderma/cutis laxa Acanthosis nigricans Periungual telengiectasia
	Erythema multiforme Leg ulcers Lichen planus
	Alopecia (non scarring) Lupus hair Telogen effluvium
	Alopecia areata Sclerodactyly Rheumatoid nodules
	Calcinosis cutis

Muscles and bones

- The most common symptom is joint pain with prelidiction towards the small joints of hand and wrist.
- The Lupus Foundation of America estimates more than 90 percent of those affected will experience joint and/or muscle pain at some time during the course of their illness.
- Unlike rheumatoid arthritis, lupus arthritis is less disabling and usually does not cause severe destruction of the joints.





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- **People with SLE are at particular risk of developing osteoarticular tuberculosis.**

- **A possible association between rheumatoid arthritis and SLE has been suggested, and SLE may be associated with an increased risk of bone fractures in relatively young women.**

Blood

- **Anemia is common in children with SLE.**
- **Low platelet and white blood cell counts occur due to the disease or a side effect of pharmacological treatment.**
- **People with SLE may have an association with antiphospholipid antibody syndrome (a thrombotic disorder), wherein autoantibodies to phospholipids are present in their serum.**

- **Abnormalities associated with antiphospholipid antibody syndrome include a paradoxical prolonged partial thromboplastin time and a positive test for antiphospholipid antibodies; the combination usually termed as "lupus anticoagulant-positive".**

- **Another autoantibody finding in SLE is the anticardiolipin antibody.**

Heart

- Inflammation of various parts of the heart, such as inflammation of the fibrous sac surrounding the heart, heart muscle, and inner lining of the heart may be seen.
- The endocarditis of SLE is characteristically noninfective (Libman-Sacks endocarditis)
- Involves mitral valve or tricuspid valve.
- Atherosclerosis also occur more often and advances more rapidly in patients of SLE.

Lungs

Lung and pleural inflammation results in :

- ✓ **Pleuritis**
- ✓ **Pleural effusion**
- ✓ **Lupus pneumonitis**
- ✓ **Chronic diffuse interstitial lung disease**
- ✓ **Pulmonary hypertension**
- ✓ **Pulmonary emboli**
- ✓ **Pulmonary hemorrhage**
- ✓ **Shrinking lung syndrome**

Kidneys

- Painless hematuria or proteinuria may be the only presenting sign of kidney involvement.

- Acute or chronic renal impairment may develop with lupus nephritis, leading to acute or end-stage kidney failure (early recognition and management of SLE has reduced its incidence to <5%)

- **A histological hallmark of SLE is Membranous glomerulonephritis with "wire loop" abnormalities.**

- **This finding is due to immune complex deposition along the glomerular basement membrane, leading to a typical granular appearance in immunofluorescence testing**

Table 2 Classification of Lupus Nephritis (International Society of Nephrology and Renal Pathology Society)

Class I: Minimal Mesangial Lupus Nephritis

Normal glomeruli by light microscopy, but mesangial immune deposits by immunofluorescence.

Class II: Mesangial Proliferative Lupus Nephritis

Purely mesangial hypercellularity of any degree or mesangial matrix expansion by light microscopy, with mesangial immune deposits. A few isolated subepithelial or subendothelial deposits may be visible by immunofluorescence or electron microscopy, but not by light microscopy.

Table 2 Classification of Lupus Nephritis (International Society of Nephrology and Renal Pathology Society)

Class III: Focal Lupus Nephritis

Active or inactive focal, segmental or global endo- or extracapillary glomerulonephritis involving <50% of all glomeruli, typically with focal subendothelial immune deposits, with or without mesangial alterations.

Class III (A): Active lesions—focal proliferative lupus nephritis

Class III (A/C): Active and chronic lesions—focal proliferative and sclerosing lupus nephritis

Class III (C): Chronic inactive lesions with glomerular scars—focal sclerosing lupus nephritis

Table 2 Classification of Lupus Nephritis (International Society of Nephrology and Renal Pathology Society)

Class IV: Diffuse Lupus Nephritis

Active or inactive diffuse, segmental or global endo- or extracapillary glomerulonephritis involving 50% of all glomeruli, typically with diffuse subendothelial immune deposits, with or without mesangial alterations. This class is divided into diffuse segmental (IV-S) lupus nephritis when 50% of the involved glomeruli have segmental lesions, and diffuse global (IV-G) lupus nephritis when 50% of the involved glomeruli have global lesions. Segmental is defined as a glomerular lesion that involves less than one-half of the glomerular tuft. This class includes cases with diffuse wire loop deposits but with little or no glomerular proliferation

Class IV-S (A): Active lesions—diffuse segmental proliferative lupus nephritis

Class IV-G (A): Active lesions—diffuse global proliferative lupus nephritis

Class IV-S (A/C): Active and chronic lesions—diffuse segmental proliferative and sclerosing lupus nephritis

Class IV-G (A/C): Active and chronic lesions—diffuse global proliferative and sclerosing lupus nephritis

Class IV-S (C): Chronic inactive lesions with scars—diffuse segmental sclerosing lupus nephritis

Class IV-G (C): Chronic inactive lesions with scars—diffuse global sclerosing lupus nephritis

Table 2 Classification of Lupus Nephritis (International Society of Nephrology and Renal Pathology Society)

Class V: Membranous Lupus Nephritis

Global or segmental subepithelial immune deposits or their morphologic sequelae by light microscopy and by immunofluorescence or electron microscopy, with or without mesangial alterations. Class V lupus nephritis may occur in combination with class III or IV, in which case both will be diagnosed. Class V lupus nephritis may show advanced sclerosis.

Class VI: Advanced Sclerotic Lupus Nephritis

90% of glomeruli globally sclerosed without residual activity.

Neuropsychiatric

- **Neuropsychiatric syndromes result when the central or peripheral nervous systems are affected.**

- **The American College of Rheumatology defines 19 neuropsychiatric syndromes in SLE.**

- **Most difficult challenge to diagnose neuropsychiatric syndromes with SLE as it may involve different patterns of S/S which may be mistaken for S/S of infectious disease or stroke**

Common neuropsychiatric manifestation of SLE

- Headache (specific lupus headache)
- cognitive dysfunction,
- mood disorder,
- cerebrovascular disease,
- seizures,
- polyneuropathy,
- anxiety disorder,
- psychosis.

Rare neuropsychiatric manifestation of SLE

- **Intracranial hypertension syndrome:**
  Characterized by an elevated intracranial pressure, papilledema, and headache with occasional abducens nerve paresis, absence of a space-occupying lesion or ventricular enlargement, and normal cerebrospinal fluid chemical and hematologicalconstituents

Other rare neuropsychiatric manifestation of SLE

- ✓ Acute confusional state
- ✓ Guillain-Barré syndrome
- ✓ Aseptic meningitis
- ✓ Autonomic disorder
- ✓ Demyelinating syndrome
- ✓ Mononeuropathy
- ✓ Myelopathy, Cranial neuropathy & plexopathy
- ✓ Movement disorder (more specifically chorea)
- ✓ Myasthenia gravis

Neurological

- The neural manifestation of lupus is known as neuropsychiatric SLE (NPSLE). One aspect of this disease is severe damage to the epithelial cells of the blood–brain barrier.

- Headaches, depression, seizures, cognitive dysfunction, mood disorder, cerebrovascular disease, polyneuropathy, anxiety disorder, psychosis, and in some extreme cases, personality disorders are seen.

Reproductive

- **Increased rate of fetal death *in utero* and spontaneous abortion**

- **Pregnancy outcome appears to be worse in people with SLE whose disease flares up during pregnancy**

- **Neonatal lupus: Occurrence of SLE symptoms in an infant born to mother with SLE, commonly presents with a rash resembling DLE, sometimes presents with systemic abnormalities such as heart block or hepatomegaly and spleenomegaly. Neonatal lupus is usually benign and self-limiting.**

SLE and Pregnancy

- Rate of fetal loss is increased (approximately two- to threefold) in women with SLE.
- Fetal demise is higher in mothers with high disease activity, antiphospholipid antibodies, and/or active nephritis.
- An additional potential problem for the fetus is the presence of antibodies to Ro, sometimes associated with neonatal lupus consisting of rash and congenital heart block.
- The latter can be life-threatening; therefore, the presence of anti-Ro requires vigilant monitoring of fetal heart rates with prompt intervention

Table 3 : Clinical Manifestations of SLE and Prevalence over the entire course of disease

Manifestation	Prevalence %
Systemic: Fatigue, malaise, fever, anorexia, weight loss	95
Musculoskeletal	95
Arthralgias/myalgias	95
Nonerosive polyarthritis	60
Hand deformities	10
Myopathy/myositis	25/5
Ischemic necrosis of bone	15
Cutaneous	80
Photosensitivity	70
Malar rash	50
Oral ulcers	40
Alopecia	40
Discoid rash	20
Vasculitis rash	20
Other (e.g., urticaria, subacute cutaneous lupus)	15

Manifestation	Prevalence %
Hematologic	85
Anemia (chronic disease)	70
Leukopenia (<4000/L)	65
Lymphopenia (<1500/L)	50
Thrombocytopenia (100,000/L)	15
Lymphadenopathy	15
Splenomegaly	15
Hemolytic anemia	10
Neurologic	60
Cognitive disorder	50
Mood disorder	40
Headache	25
Seizures	20
Mono-, polyneuropathy	15
Stroke, TIA	10
Acute confusional state or movement disorder	2–5
Aseptic meningitis, myelopathy	<1

Table 3 : Clinical Manifestations of SLE and Prevalence over the entire course of disease

Manifestation	Prevalence %
Cardiopulmonary	60
Pleurisy, pericarditis, effusions	30–50
Myocarditis, endocarditis	10
Lupus pneumonitis	10
Coronary artery disease	10
Interstitial fibrosis	5
Pulmonary hypertension, ARDS, hemorrhage	<5
Shrinking lung syndrome	<5
Renal *	30–50
Proteinuria500 mg/24 h, cellular casts	30–50
Nephrotic syndrome	25
End-stage renal disease	5–10

Table 3 : Clinical Manifestations of SLE and Prevalence over the entire course of disease

Manifestation	Prevalence %
Gastrointestinal	40
Nonspecific (nausea, mild pain, diarrhea)	30
Abnormal liver enzymes	40
Vasculitis	5
Thrombosis	15
Venous	10
Arterial	5
Ocular	15
Sicca syndrome	15
Conjunctivitis, episcleritis	10
Vasculitis	5

Diagnosis of SLE

- The diagnosis of SLE is based on characteristic clinical features and autoantibodies.
- Current criteria for classification are listed in Table 4.
- An algorithm for diagnosis and initial therapy is shown in Figure.

Table 4: Diagnostic Criteria for Systemic Lupus Erythematosus

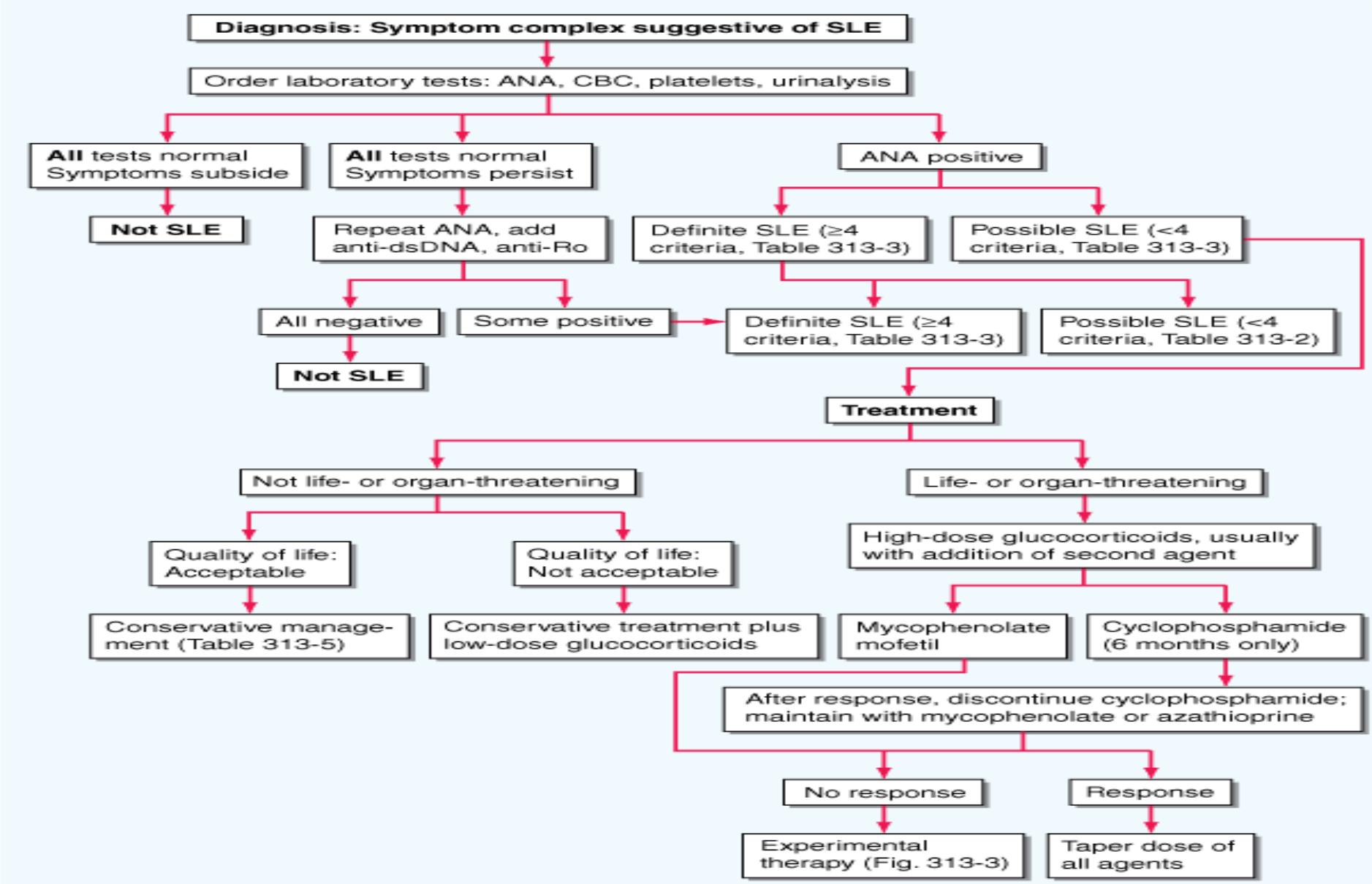
Malar rash	**Fixed erythema, flat or raised, over the malar eminences**
Discoid rash	**Erythematous circular raised patches with adherent keratotic scaling and follicular plugging; atrophic scarring may occur**
Photosensitivity	**Exposure to ultraviolet light causes rash**
Oral ulcers	**Includes oral and nasopharyngeal ulcers, observed by physician**
Arthritis	**Nonerosive arthritis of two or more peripheral joints, with tenderness, swelling, or effusion**
Serositis	**Pleuritis or pericarditis documented by ECG or rub or evidence of effusion**

Table 4: Diagnostic Criteria for Systemic Lupus Erythematosus

Renal disorder	**Proteinuria >0.5 g/dL or 3+, or cellular casts**
Neurologic disorder	**Seizures or psychosis without other causes**
Hematologic disorder	**Hemolytic anemia or leukopenia (<4000/L)**
	or lymphopenia (<1500/L)
	or thrombocytopenia(<100,000/L)
	in the absence of offending drugs

If 4 of these criteria, well documented, are present at any time in a patient's history, the diagnosis is likely to be SLE. Specificity is 95%; sensitivity is 75%.

Figure: Algorithm for diagnosis and initial therapy of SLE



Laboratory Diagnosis

❑ Purpose of Laboratory tests

(1) to establish or rule out the diagnosis

(2) to follow the course of disease, particularly to suggest that a flare is occurring or organ damage is developing

(3) to identify adverse effects of therapies

❑ Serologic testing for SLE:

Antinuclear antibody (ANA) testing and anti-extractable nuclear antigen (anti-ENA) form the mainstay of serologic testing for SLE.

Anti-nuclear antibody (ANA)

- Also known as antinuclear factor or ANF
- ANA are heterogenous group of autoantibodies that bind to contents of the cell nucleus.
- Each of these antibody subtypes binds to different proteins or protein complexes within the nucleus.
- They are directed against ds-DNA, histones, SSA / Ro , SSB / La, Sm, Sm / RNP, Scl-70, Jo-1 & Centromere.
- The common tests used for detecting and quantifying ANAs are indirect immunofluorescence and Enzyme-Linked ImmunoSorbent Assay (ELISA)

- Positive in almost all people with SLE, although can also be positive in those with other autoimmune diseases as it indicates a stimulated immune system.
- 95% sensitive when human cultured cells are used as substrate
- Specificity is low
- Some healthy individuals have positive test
- 10-35% of individuals older than 65 years have ANA
- Family of SLE patients.

- In contrast to the low positive predictive value of ANA testing, a patient with a negative test has less than a 3% chance of having SLE.

- Thus a negative ANA test is useful for excluding the diagnosis of SLE.

- However in the presence of typical features of lupus a negative ANA test does not exclude the diagnosis.

Immunoflouresence

- Clinically the most widely used method is indirect immunofluorescence (IF).
- The pattern of fluorescence suggests the type of antibody present in the people's serum.
- Direct immunofluorescence can detect deposits of immunoglobulins and complement proteins in the people's skin.
- When skin not exposed to the sun is tested, a positive direct IF (the so-called lupus band test) is an evidence of systemic lupus erythematosus

- Indirect immunofluorescence is one of the most commonly used tests for ANAs.
- Typically, HEp-2 cells are used as a substrate to detect the antibodies in human serum.
- Microscope slides are coated with HEp-2 cells and the serum is incubated with the cells.
- If antibodies are present then they will bind to the antigens on the cells; in the case of ANAs, the antibodies will bind to the nucleus.

- These can be visualised by adding a fluorescent tagged (usually FITC or rhodopsin B) anti-human antibody that binds to the antibodies.
- The molecule will fluoresce when a specific wavelength of light shines on it, which can be seen under the microscope.
- Depending on the antibody present in the human serum and the localisation of the antigen in the cell, distinct patterns of fluorescence will be seen on the HEp-2 cells

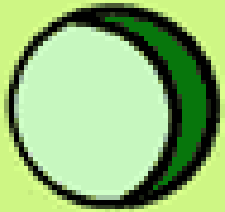
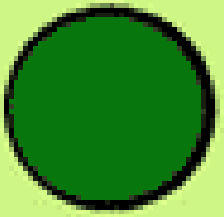
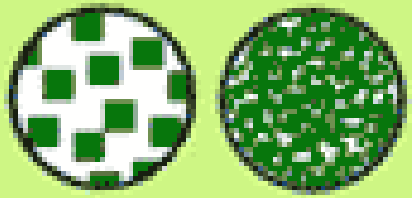
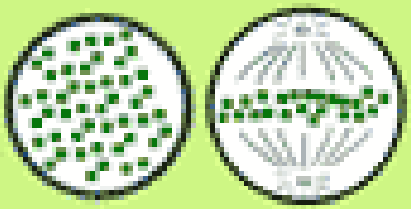
- In immunofluorescence, the level of autoantibodies is reported as a titre. This is the highest dilution of the serum at which autoantibodies are still detectable.
- Positive autoantibody titres at a dilution equal to or greater than 1:160 are usually considered as clinically significant.
- Positive titres of less than 1:160 are present in up to 20% of the healthy population, especially the elderly.
- Although positive titres of 1:160 or higher are strongly associated with autoimmune disorders, they are also found in 5% of healthy individuals.

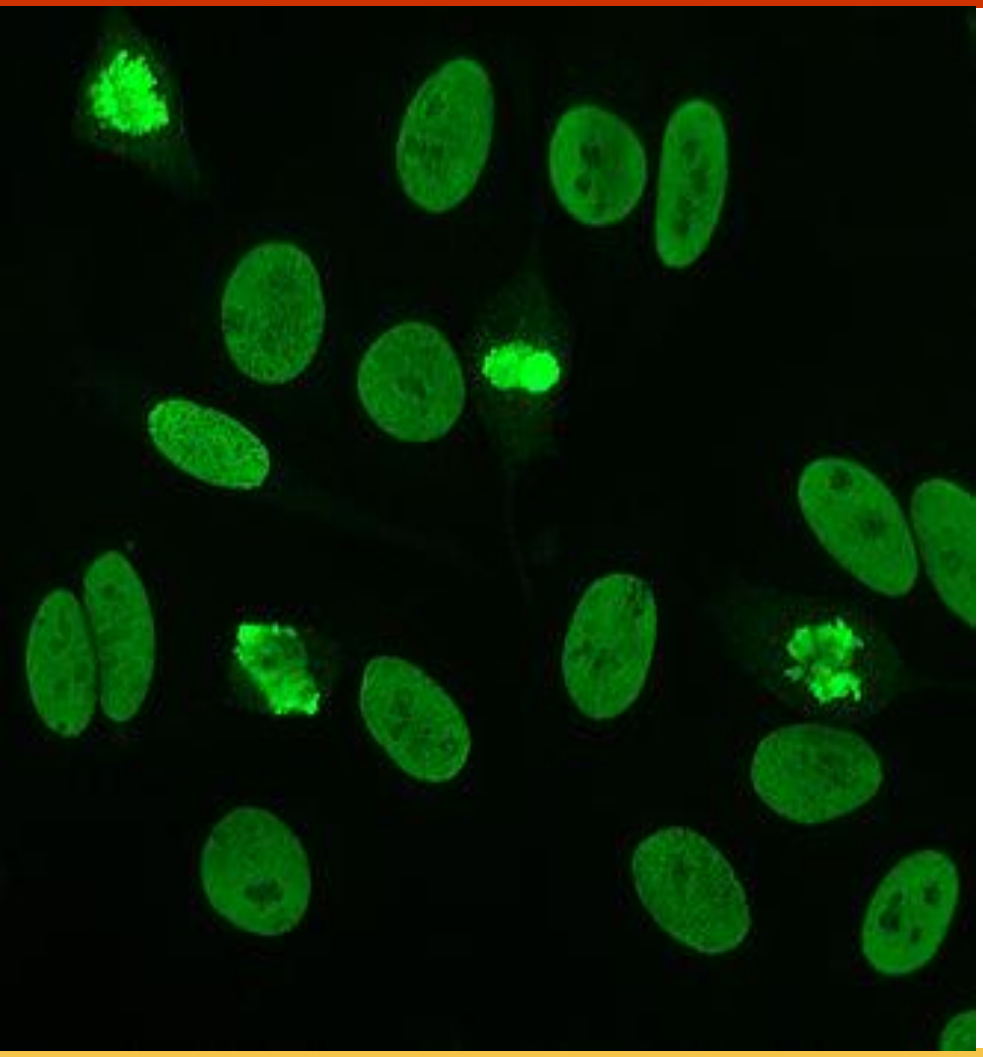
Table: Hep2 pattern with Disease Association

SITE	PATTERN	DISEASE ASSOCIATION
NUCLEAR		
	Homogenous	SLE & other connective tissue disorders, Drug induced SLE
	Peripheral	SLE & other connective tissue disorders
	Speckled Coarse	Mixed Connective Tissue Disorders (MCTD), Scleroderma - Polymyositis overlap syndrome, Raynaud's phenomenon, Psoriasis, Sjogren's syndrome Systemic sclerosis
	Speckled Fine	SLE, Sjogren's syndrome, Scleroderma, Myositis, MCTD
	Nuclear Dots Few	Autoimmune & Viral diseases - Primary Biliary Cirrhosis & Chronic Active Hepatitis, Rarely Collagen vascular diseases
	Nuclear Dots Multiple	Primary Biliary Cirrhosis (>30%)
	Centromere	CREST Syndrome, Progressive Systemic sclerosis

Table contd.

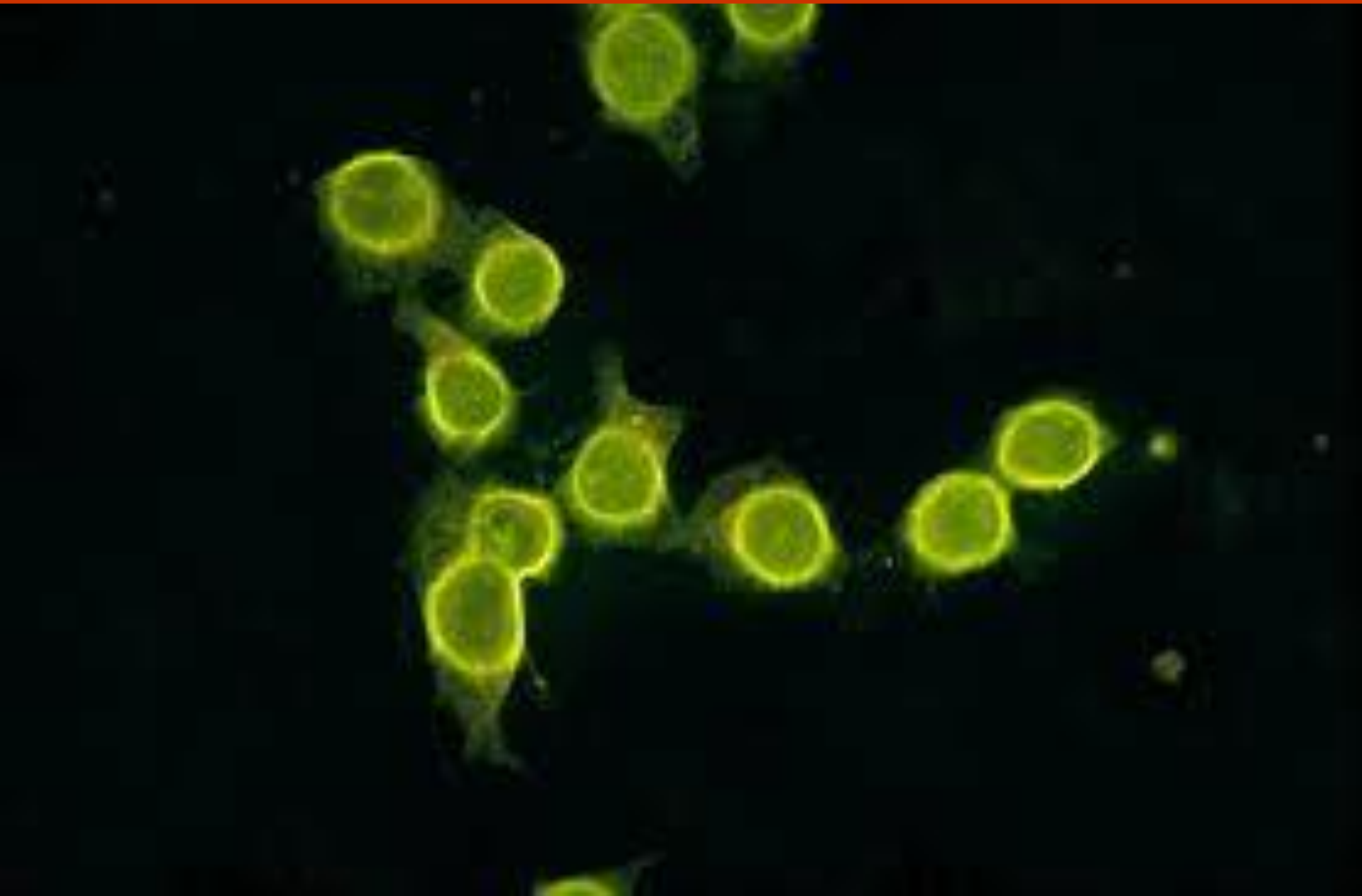
SITE	PATTERN	DISEASE ASSOCIATION
NUCLEOLAR		
	Homogenous	Scleroderma, Myositis, Raynaud's phenomenon, SLE Rheumatoid arthritis
	Clumpy	Systemic sclerosis & Scleroderma
CYTOPLASMIC		
	Mitochondrial	Primary Biliary Cirrhosis, Scleroderma Overlap syndrome
	Ribosomal	SLE(10-20%)

Peripheral (rim)		anti-DNA (not seen on HEp-2)	SLE
Homogeneous (diffuse)		anti-DNA anti-histone anti-DNP (nucleosomes)	RA & SLE Misc. Disorders (anti-ssDNA)
Speckled		anti-Sm & RNP anti-Ro & La anti-Jo-1 & Mi-2 anti-Scl-70	SLE & SS PM/DM PSS (Systemic)
Centromere		anti-centromere	PSS (CREST)
Nucleolar		anti-nucleolar	SLE & PSS



Homogeneous immuno-fluorescence staining pattern of double stranded DNA antibodies on HEp-2 cells. Interphase cells show homogeneous nuclear staining while mitotic cells show staining of the condensed chromosome regions

Positive antinuclear antibody (ANA) test with a rim pattern, with Hep2 cell substrate, using FITC immunofluorescence



Nucleolar staining pattern of ANAs

ANA testing by Enzyme-linked mmunosorbent assay (ELISA)

- Enzyme-linked immunosorbent assay (ELISA) uses antigen-coated microtitre plates for the detection of ANAs.
- Each well of a microtitre plate is coated with either a single antigen or multiple antigens to detect specific antibodies or to screen for ANAs, respectively.
- There are significant differences in the detection of ANA by immunofluorescence and different ELISA kits.

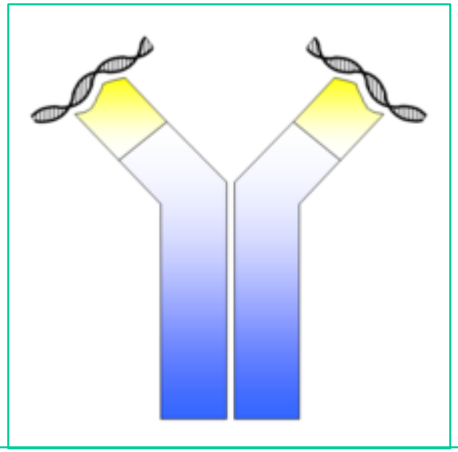
Extractable nuclear antigens (ENA)

- Extractable nuclear antigens (ENA) are a group of autoantigens that were originally identified as antibody targets in people with autoimmune disorders.
- They are termed ENA because they can be extracted from the cell nucleus with saline.
- The ENAs consist of ribonucleoproteins and non-histone proteins, named by either the name of the donor who provided the prototype serum (Sm, Ro, La, Jo), or the name of the disease setting in which the antibodies were found (SS-A, SS-B, Scl-70)

Anti-Extractable Nuclear Antigen Testing

Anti-double stranded DNA (Anti-dsDNA)

- Highly specific for SLE
- Present in 70% of cases
- The anti-dsDNA antibody titers also tend to reflect disease activity, although not in all cases
- High results are characteristic of active SLE
- The variable regions (yellow) are complementary to the dsDNA strands



dsDNA antibody

- **Anti-Sm antibody – usually seen only in those with SLE.**
- **Anti-SSA and Anti-SSB – may also be positive. SS-A and SS-B confer a specific risk for heart conduction block in neonatal lupus.**
- **Anti-RNP – may be positive.**
- **Anti-chromatin antibodies – may be present in people with SLE who are positive for ANA but negative for anti-dsDNA.**
- **Histone antibodies – for drug-induced lupus.**
- **Antiphospholipid antibodies – such as lupus anticoagulant, anticardiolipin, anti-β2 glycoprotein I**

Table 5: Autoantibodies in Systemic Lupus Erythematous (SLE)

Antibody	Prevalence, %	Antigen Recognized	Clinical Utility
Antinuclear antibodies	98	Multiple nuclear	Best screening test; repeated negative tests make SLE unlikely
Anti-dsDNA	70	DNA (double-stranded)	High titers are SLE-specific and in some patients correlate with disease activity, nephritis, vasculitis
Anti-Sm	25	Protein complexed to 6 species of nuclear U1 RNA	Specific for SLE; no definite clinical correlations; most patients also have anti-RNP; more common in blacks and Asians than whites
Anti-RNP	40	Protein complexed to U1 RNA	Not specific for SLE; high titers associated with syndromes that have overlap features of several rheumatic syndromes including SLE; more common in blacks than whites
Anti-Ro (SS-A)	30	Protein complexed to hY RNA, primarily 60 kDa and 52 kDa	Not specific for SLE; associated with sicca syndrome, predisposes to subacutecutaneous lupus, and to neonatal lupus with congenital heart block; associated with decreased risk for nephritis
[illegible]	[illegible]	47-kDa protein complexed to hY	Usually associated with anti-Ro;

Table 5: Autoantibodies in Systemic Lupus Erythematous (SLE)

Antibody	Prevalence, %	Antigen Recognized	Clinical Utility
Antihistone	70	Histones associated with DNA (in nucleosome, chromatin)	More frequent in drug-induced lupus than in SLE
Antiphospholipid	50	Phospholipids,	Three tests available—ELISAs for cardiolipin and
		$_2$ glycoprotein 1 cofactor, prothrombin	$_2$G1, sensitive prothrombin time (DRVVT); predisposes to clotting, fetal loss, thrombocytopenia
Antierythrocyte	60	Erythrocyte membrane	Measured as direct Coombs' test; a small proportion develops overt hemolysis
Antiplatelet	30	Surface and altered cytoplasmic antigens on platelets	Associated with thrombocytopenia but sensitivity and specificity are not good; this is not a useful clinical test
Antineuronal (includes anti-glutamate receptor)	60	Neuronal and lymphocyte surface antigens	In some series a positive test in CSF correlates with active CNS lupus.
Antiribosomal P	20	Protein in ribosomes	In some series a positive test in serum correlates with depression or psychosis due to CNS lupus

Antiphospholipid antibodies

- Antiphospholipid syndrome can be primary or secondary. Primary antiphospholipid syndrome occurs in the absence of any other related disease. Secondary antiphospholipid syndrome occurs with other autoimmune diseases, such as SLE.
- The antiphospholipid syndrome responsible for most of the miscarriages in later trimesters seen in concomitant systemic lupus erythematosus and pregnancy.
- Antiphospholipid antibodies (lupus anticoagulant [LA], IgG and IgM Anticardiolipin [aCL] antibodies; and IgG and IgM anti-beta2-glycoprotein [GP] I should be tested.

Prognostic markers and the role of autoantibodies

- Serum anti-dsDNA titres have been correlated with LN, progression to end stage renal disease and increased disease severity, damage or poor survival.

- Antiphospholipid antibodies are strongly associated with features of the Antiphospholipid syndrome (APS), CNS involvement, severe LN, damage accural and death.

- **Anti-Ro(SS-A) and Anti-La (SS-B) antibodies have been associated with neonatal lupus and congenital heart block in children of seropositive mothers.**

- **Antibodies to other extractable nuclear antigens (Anti-Ro/La/Sm/RNP) have been associated with mucocutaneous involvement and less severe nephropathy in most studies.**

Measurement of Complement

Measurement of Complement

- C3 decreased ,
- C4 decreased ,
- CH50 decreased

Other tests

- **Urinalysis – may show blood, urinary casts, or protein in the urine, which can indicate kidney involvement**
- **Complete blood count (CBC) – may reveal anemia and decreased numbers of white blood cells and platelets, which can occur with lupus**
- **Kidney and Liver function test indicates current status of the kidneys and liver as well as electrolyte and acid/base balance and levels of blood glucose and blood proteins**
- **Rheumatoid factor (RF) – may be positive or negative**

- **Serum protein electrophoresis (SPEP) – increased gamma globulin proteins**
- **Erythrocyte sedimentation rate (ESR) – increased with inflammation, such as with lupus as well as other inflammatory conditions**
- **C-reactive protein (CRP) – another marker of inflammation that may be elevated with lupus**
- **Cryoglobulin – frequently positive; cryoglobulins are abnormal proteins in the blood that will precipitate when the body temperature drops below normal, causing blockage of the blood vessels.**

Treatment

- There is no cure for SLE, and complete sustained remissions are rare. Therefore, the physician should plan to induce improvement of acute flares and then maintain improvements with strategies that suppress symptoms to an acceptable level and prevent organ damage. Usually patients will endure some adverse effects of medications.

- Therapeutic choices depend on (1) whether disease manifestations are life-threatening or likely to cause organ damage, justifying aggressive therapies; (2) whether manifestations are potentially reversible; and (3) the best approaches to preventing complications of disease and its treatments.

Table 6: Medications for the Management of SLE

Medication	Dose Range	Drug Interactions	Serious or Common Adverse Effects
NSAIDs, salicylates (Ecotrin[a] and St. Joseph's aspirin[a] approved by FDA for use in SLE)	Doses toward upper limit of recommended range usually required	A2R/ACE inhibitors, glucocorticoids, fluconazole, methotrexate, thiazides	NSAIDs: Higher incidence of aseptic meningitis, transaminitis, decreased renal function, vasculitis of skin; entire class, especially COX-2-specific inhibitors, may increase risk for myocardial infarction Salicylates: ototoxicity, tinnitus Both: GI events and symptoms, allergic reactions, dermatitis, dizziness, acute renal failure, edema, hypertension
Topical glucocorticoids	Mid-potency for face; mid to high potency other areas	None known	Atrophy of skin, contact dermatitis, folliculitis, hypopigmentation, infection

Table 6: Medications for the Management of SLE

Medication	Dose Range	Drug Interactions	Serious or Common Adverse Effects
Topical sunscreens	SPF 15 at least; 30+ preferred	None known	Contact dermatitis
Hydroxychloroquine[a] (quinacrine can be added or substituted)	200–400 mg qd (100 mg qd)	None known	Retinal damage, agranulocytosis, aplastic anemia, ataxia, cardiomyopathy, dizziness, myopathy, ototoxicity, peripheral neuropathy, pigmentation of skin, seizures, thrombocytopenia. Use in pregnancy may be acceptable. Pregnancy category DQuinacrine usually causes diffuse yellow skin coloration
DHEA (dehydroepiandrosterone)	200 mg qd	Unclear	Acne, menstrual irregularities, high serum levels of testosterone

Table 6: Medications for the Management of SLE

Medication	Dose Range	Drug Interactions	Serious or Common Adverse Effects
Methotrexate (for dermatitis, arthritis)	10–25 mg once a week, PO or SC, with folic acid; decrease dose if CrCl <60 mL/min	Acitretin, leflunomide, NSAIDs and salicylates, penicillins, probenecid, sulfonamides, trimethoprim	Anemia, bone marrow suppression, leukopenia, thrombocytopenia, hepatotoxicity, nephrotoxicity, infections, neurotoxicity, pulmonary fibrosis, pneumonitis, severe dermatitis, seizures. Teratogenic. Pregnancy category X
Glucocorticoids, oral[a] (several specific brands are approved by FDA for use in SLE)	Prednisone, prednisolone: 0.5–1 mg/kg per day for severe SLE0.07–0.3 mg/kg per day or qod for milder disease	A2R/ACE antagonists, antiarrhythmics class III, $_2$, cyclosporine, NSAIDs and salicylates, phenothiazines, phenytoins, quinolones, rifampin, risperidone, thiazides, sulfonylureas, warfarin	Infection, VZV infection, hypertension, hyperglycemia, hypokalemia, acne, allergic reactions, anxiety, aseptic necrosis of bone, cushingoid changes, CHF, fragile skin, insomnia, menstrual irregularities, mood swings, osteoporosis, psychosis

Table 6: Medications for the Management of SLE

Medication	Dose Range	Drug Interactions	Serious or Common Adverse Effects
Methylprednisolone sodium succinate, IV[a] (FDA approved for lupus nephritis)	For severe disease, 1 g IV qd x 3 days	As for oral glucocorticoids	As for oral glucocorticoids (if used repeatedly); anaphylaxis
Cyclophosphamide[b,c]?IV	7–25 mg/kg q month x 6; consider mesna administration with dose	Allopurinol, bone marrow suppressants, colony-stimulating factors, doxorubicin, rituximab, succinylcholine, zidovudine	Infection, VZV infection, bone marrow suppression, leukopenia, anemia, thrombocytopenia, hemorrhagic cystitis (less with IV), carcinoma of the bladder, alopecia, nausea, diarrhea, malaise, malignancy, ovarian and testicular failure. Teratogenic. Pregnancy category D
Mycophenolate mofetil[b] or mycophenolic acid	MMF: 2–3 g/d PO; max 1 g bid if CrCl <25 mL/min.MPA: 360–1080 mg bid. Caution if CrCl <25 mL/min	Acyclovir, antacids, azathioprine, bile acid–binding resins, ganciclovir, iron, salts, probenecid, oral contraceptives	Infection, leukopenia, anemia, thrombocytopenia, lymphoma, lymphoproliferative disorders, malignancy, alopecia, cough, diarrhea, fever, GI symptoms, headache, hypertension, hypercholesterolemia, hypokalemia, insomnia, peripheral edema, transaminitis, tremor, rash. Teratogenic. Pregnancy category D

Table 6: Medications for the Management of SLE

Medication	Dose Range	Drug Interactions	Serious or Common Adverse Effects
Azathioprine[b]	**2–3 mg/kg per day PO; decrease frequency of dose if CrCl <50 mL/min**	**ACE inhibitors, allopurinol, bone marrow suppressants, interferons, mycophenolate mofetil, rituximab, warfarin, zidovudine**	**Infection, VZV infection, bone marrow suppression, leukopenia, anemia, thrombocytopenia, pancreatitis, hepatotoxicity, malignancy, alopecia, fever, flulike illness, GI symptoms. Use in pregnancy may be acceptable. Pregnancy category D**
Belimumab	**10 mg/kg i.v.**	**wk 0,2,4 then monthly**	**Infusion reactions Allergy Infections probable**
Rituximab (for patients resistant to above therapies)	**375 mg/M2 q wk x 4 or 1 g q 2 wks x 2**	**IVIg**	**Infection (including PML), infusion reactions, headache, arrythmias, allergic responses. Pregnancy category C**
Belimumab	**10 mg/kg i.v.**	**IV Ig**	**Infec**

Patient Outcomes, Prognosis, and Survival

Patient Outcomes, Prognosis, and Survival

- **Poor prognosis (50% mortality in 10 years) in most series is associated with (at the time of diagnosis) high serum creatinine levels (>1.4 mg/dL), hypertension, nephrotic syndrome (24-h urine protein excretion >2.6 g), anemia [hemoglobin (<12.4 g/dL)], hypoalbuminemia, hypocomplementemia, aPL, male sex, and ethnicity (African American, Hispanic with mestizo heritage)**

- **Disability in patients with SLE is common due primarily to chronic fatigue, arthritis, and pain, as well as renal disease. As many as 25% of patients may experience remissions, sometimes for a few years, but these are rarely permanent.**

- **The leading causes of death in the first decade of disease are systemic disease activity, renal failure, and infections; subsequently, thromboembolic events become increasingly frequent causes of mortality.**

Laboratory tests available for Autoimmune disorders

- ANA
- ENA
- Anti-ds DNA
- Anti-ds DNA IFA
- Anti-ds DNA IFA in Dilution
- Anti-ds DNA EIA
- ANTI-ss DNA Antibody

Laboratory tests available for Autoimmune disorders

- LE Cell
- Anti-Cardiolipin Antibody
- Anti-Phospholipid Antibody
- Smooth Muscle Antibody (ASMA), IFA
- Smooth Muscle Antibody (ASMA), IFA in Dilution

Panel or Group Tests

- **Lupus Erythematous Panel; SLE Panel**

  ANA, Ds DNA, Sm, U1RNP, CRP, Histone Antibody

- **Autoimmune Liver Panel**

  AMA-M2, SP 100, LKM1, gp120, LC1, SLA

- **Autoimmune Vaculitis Panel**

- **Autoimmune Neuro Panel**

- **Collagen Disease Antibodies Panel**

  ANA, Anti-ds-DNA, Sm, SS-A/Ro, SS-B/La, Scl-70, Jo-1 , Centromere, U1RNP

Panel or Group Tests

- **Rheumatoid Arthritis Panel**

  ANA, Rheumatoid Factor, Anti Cyclic Citrullinated Peptide

- **Rheumatoid Autoimmune Panel**

  ANA, Anti-ds DNA, Sm, U1RNP, Rheumatoid Factor, C3, C4

- **Rheumatoid Autoimmune Comprehensive Panel**

  ANA, Anti-ds DNA, Sm, U1RNP, Rheumatoid Factor, C3, C4, SS-A (Ro), SS-B (La), Centromere Antibody

For Predisposing Factors

- **HLA-DR3**
- **HLA-DR3**
- **HLA-DQB1**
- **EB Virus**

Supporting Tests

- **CBC**
- **Urine RE**
- **Creatinine**
- **Urinary Protein**

Thank you

Dr Lal PathLabs

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