Part 1: PATHOLOGY OF VASCULITIS

INFLAMMATORY VASCULAR DISEASES
(VASCULITIDES)

Infectious Vasculitis (caused by vessel invasion by pathogenic microorganisms)
- Bacterial
- Rickettsial
- Viral
- Fungal

Noninfectious Vasculitis (not known to be caused by vessel invasion by pathogens)
- Large vessel vasculitis
- Medium-sized vessel vasculitis
- Small vessel vasculitis
- Phlebitis
- Atherosclerosis

CASE #1
A 30 year old white male developed headaches, myalgias and fever. He noticed the onset of an erythematous rash on his arms and hands that became predominantly petechial.

He got bad advice from a friend. He developed abdominal pain and vomiting. Physical examination revealed a fever of 40°C, a petechial rash on all extremities and the trunk, an enlarged liver and increased respiratory rate. He had hiked along the Blue Ridge Parkway during Spring break. Skin biopsy was diagnostic. He was treated but died two weeks after the initial onset of symptoms.

Diagnosis: Rocky Mountain Spotted Fever
Immunologic Pathogenic Mechanisms of “Noninfectious” Vasculitis

- Immune Complex Mediated Vasculitis
- Direct Antibody Attack Mediated Vasculitis
- Anti-neutrophil Cytoplasmic Autoantibody (ANCA) Mediated Vasculitis (Pauci-immune Vasculitis)
- Cell Mediated Vasculitis

Three general categories of systemic vasculitis are:

- Large vessel vasculitis (chronic granulomatous arteritis)
- Medium-sized vessel vasculitis (necrotizing arteritis)
- Small vessel vasculitis (necrotizing polyangiitis)

Names Proposed by the Chapel Hill Nomenclature System

- Large Vessel Vasculitis
  - Giant Cell Arteritis
  - Takayasu Arteritis

- Medium-Sized Vessel Vasculitis
  - Polyarteritis Nodosa
  - Kawasaki Disease

- Small Vessel Vasculitis
  - Microscopic Polyangiitis
  - Wegener’s Granulomatosis
  - Churg-Strauss Syndrome
  - Henoch-Schönlein Purpura
  - Cryoglobulinemic Vasculitis
  - Cutaneous small vessel vasculitis

Large Vessel Vasculitis (chronic granulomatous arteritis)

Intima

Media

Adventitia
Large Vessel Vasculitis
(quiescent chronic granulomatous arteritis)

Takayasu Arteritis

Granulomatous inflammation of the aorta and its major branches. Usually occurs in patients younger than 50.
Giant Cell (Temporal) Arteritis

Granulomatous arteritis of the aorta and its major branches, with a predilection for the extracranial branches of the carotid artery. Often involves the temporal artery. Usually occurs in patients older than 50 and often is associated with polymyalgia rheumatica.
CASE #2

A 62 year old white woman developed progressive weight loss, fatigue, “morning stiffness” in her hips and shoulders and low grade fever. She also developed headaches and soreness over the right side of her face and scalp. A rheumatologist confirmed the fever and noted tenderness and slight nodularity of the right temporal artery, a faint bruit in the right neck, and reduced radial pulses bilaterally. Laboratory results included a markedly elevated ESR, mild anemia and slightly elevated platelet count. A segment of the right temporal artery was removed for pathologic evaluation.

Diagnosis: Giant Cell Arteritis

Giant Cell Arteritis (aortitis)

Giant Cell Arteritis (aortitis)

Giant Cell Arteritis (aortitis)

Giant Cell Arteritis (aortitis)
Medium-sized Vessel Vasculitis (Necrotizing Arteritis)

Necrotizing arteritis begins as a segmental necrotizing inflammation of arteries with conspicuous infiltration of neutrophils and monocytes. This typically induces fibrinoid necrosis characterized by accumulation of plasma proteins in injured tissue, including coagulation factors that are converted to fibrin.

A “large artery” in a small biopsy is in fact small artery

Skeletal Muscle Biopsy
Nerve Biopsy

Necrotizing arteritis in a small biopsy specimen does not distinguish between a medium-sized vessel vasculitis (e.g. PAN) and a small vessel vasculitis (e.g. MPA)

Sclerotic Necrotizing Arteritis

All necrotizing vasculitis enters a final common pathway of chronic inflammation and scarring.

The transformation to sclerotic lesions with a predominance of infiltrating T-lymphocytes and macrophages occurs as quickly as one or two weeks after the initiation of injury.
If the necrotizing inflammation extends through the vessel wall into the perivascular tissue, inflammatory aneurysms (pseudoaneurysms) will develop.

**Nomenclature History of Necrotizing Arteritis**

- Kussmaul & Maier 1866
- Godman & Churg 1954
- Arkin 1930
- Davson 1948
- Zeek 1952
- Chapel Hill 1994
- Microscopic polyangiitis
- Polyarteritis nodosa
- Wegener’s granulomatosis
- Churg-Strauss syndrome

**Polyarteritis Nodosa**

Necrotizing inflammation of medium-sized or small arteries without glomerulonephritis or vasculitis in arterioles, capillaries or venules.
Kawasaki Disease

Arteritis involving large, medium-sized and small arteries, and associated with mucocutaneous lymph node syndrome. Coronary arteries are often involved. Aorta and veins may be involved. Usually occurs in children.
CASE #3
A 43 year old male noted worsening arthralgias, myalgias, abdominal pain, migrating “tingling” in both lower extremities, and marked weakness in his left leg and foot. Physical examination revealed livido reticularis on his left lower extremity. Nerve conduction studies indicated a peripheral neuropathy. Laboratory data included mildly elevated LFTs, mildly increased CPK, positive stool occult blood, negative ANCA, negative rheumatoid factor, negative serology for hepatitis B and C, negative cryoglobulins, normal serum complement, negative ANA, normal CBC and unremarkable urine analysis. A biopsy of sural nerve and adjacent skeletal muscle was performed.

Diagnosis: necrotizing arteritis, differential diagnosis includes polyarteritis nodosa, and microscopic polyangiitis and other vasculitides that cause necrotizing arteritis (recommend ANCA serology)

CASE #4
A 3 year old child became irritable and febrile. The parents noticed that the baby had a red tongue and throat, and swollen “glands” in the neck. They were worried about “strep throat”. After the baby did not improve over several days, they took the baby to see a pediatrician. The baby became extremely agitated on the way to the doctor and went into cardiopulmonary arrest soon after arriving at the office. The baby could not be resuscitated. A forensic autopsy was ordered by the medical examiner.

Diagnosis: Kawasaki Disease

Small vessel vasculitis (necrotizing polyangiitis) often has clinical and pathologic evidence for involvement of capillaries (e.g., glomerulonephritis and pulmonary capillaritis) and venules (e.g., dermal leukocytoclastic venulitis), although arteries may be affected (e.g. epineural arteries causing peripheral neuropathy and intraparenchymal arteries causing abdominal pain).
Small Vessel Vasculitis

Immune Complex Small Vessel Vasculitis
Henoch-Schönlein Purpura
Cryoglobulinemic Vasculitis
Serum Sickness Vasculitis
Goodpasture’s Syndrome
Lupus Vasculitis
Rheumatoid Vasculitis
Hypocomplementemic Urticarial Vasculitis
Drug-Induced Immune Complex Vasculitis
Infection-Induced Immune Complex Vasculitis
Pauci-immune (ANCA) Small Vessel Vasculitis
Microscopic Polyangiitis
Wegener’s Granulomatosis
Churg-Strauss Syndrome
Organ-limited Disease (e.g. Pauci-immune Crescentic GN)

Immune Complex Vasculitis

One of many clinical presentations of small vessel vasculitis is palpable purpura.

Henoch-Schönlein Purpura

Vasculitis with IgA-dominant immune deposits affecting small vessels, i.e. capillaries, venules, or arterioles. Typically involves skin, gut & glomeruli, and is associated with arthralgias or arthritis.

Purpura
Abdominal pain
Arthralgias (Nephritis)

Edward Henoch
Johann Lukas Schönlein

Cryoglobulinemic Vasculitis

Vasculitis with cryoglobulin immune deposits affecting small vessels, i.e. capillaries, venules, or arterioles, and associated with cryoglobulins in serum. Skin and glomeruli are often involved.
Cryoglobulinemic Glomerulonephritis

Immune Complex Small Vessel Vasculitis

- Henoch-Schönlein Purpura
- Cryoglobulinemic Vasculitis
- Serum Sickness Vasculitis
- Goodpasture’s Syndrome
- Lupus Vasculitis
- Rheumatoid Vasculitis
- Sjögren’s Syndrome Vasculitis
- Hypocomplementemic Urticarial Vasculitis
- Behçet’s Disease
- Drug-Induced Immune Complex Vasculitis
- Infection-Induced Immune Complex Vasculitis

Anti-GBM Disease

Small Vessel Vasculitis

- Immune Complex Small Vessel Vasculitis
  - Henoch-Schönlein Purpura
  - Cryoglobulinemic Vasculitis
  - Serum Sickness Vasculitis
  - Goodpasture’s Syndrome
  - Lupus Vasculitis
  - Rheumatoid Vasculitis
  - Sjögren’s Syndrome Vasculitis
  - Hypocomplementemic Urticarial Vasculitis
  - Drug-Induced Immune Complex Vasculitis
  - Infection-Induced Immune Complex Vasculitis
- Pauci-immune (ANCA) Small Vessel Vasculitis
  - Microscopic Polyangiitis
  - Wegener’s Granulomatosis
  - Churg-Strauss Syndrome
- Organ-limited Disease (e.g. Pauci-immune Crescentic GN)

ANTIBODY MEDIATED GLOMERULONEPHRITIS AND VASCULITIS

- Anti-GBM disease
- Circulating anti-GBM antibodies with linear capillary IF staining
- Immune complex disease
- Pauci-immune (ANCA) disease
- Vascular immune
- Little or no vascular IF staining
- Granulomatous inflammation, asthma, and eosinophilia
- Granulomatous inflammation without asthma
- Granulomatous inflammation without asthma and eosinophilia
- Hypocomplementemic Urticarial Vasculitis
- Drug-Induced Immune Complex Vasculitis
- Infection-Induced Immune Complex Vasculitis
- Small vessel vasculitis
FREQUENCY OF DIFFERENT IMMUNOPATHOLOGIC TYPES OF CRESCENTIC GLOMERULONEPHRITIS RELATIVE TO AGE

<table>
<thead>
<tr>
<th>Age</th>
<th>Immune Complex</th>
<th>Anti-GBM</th>
<th>Pauci-Immune</th>
<th>Other</th>
</tr>
</thead>
<tbody>
<tr>
<td>All ages</td>
<td>24%</td>
<td>15%</td>
<td>60%</td>
<td>1%</td>
</tr>
<tr>
<td>Age 1-20</td>
<td>45%</td>
<td>12%</td>
<td>42%</td>
<td>0%</td>
</tr>
<tr>
<td>Age 21-60</td>
<td>35%</td>
<td>15%</td>
<td>48%</td>
<td>3%</td>
</tr>
<tr>
<td>Age 61-100</td>
<td>6%</td>
<td>15%</td>
<td>79%</td>
<td>0%</td>
</tr>
</tbody>
</table>

* Of patients with pauci-immune crescentic glomerulonephritis, ~80% have ANCA and ~75% have systemic vasculitis.

**Pauci-Immune (ANCA) Small Vessel Vasculitis**

- Necrotizing Venulitis
- Necrotizing Arteritis
- Glomerular Capillaritis
- Pulmonary Capillaritis

**Acute ANCA Renal Vasculitis**

- Necrotizing Glomerulonephritis
- Necrotizing Medullary Angitis
- Necrotizing Arteritis
- Segmental Fibrinoid Necrosis

**Anaphylactic Warfarin Allergy**

**Anti-neutrophil Cytoplasmic Autoantibodies (ANCA)**

- Staining of alcohol-fixed neutrophils
- Staining of formalin-fixed neutrophils
- Immunoassay antigen specificity

**C-ANCA**
- Cytoplasmic
- Cytoplasmic

**P-ANCA**
- Cytoplasmic
- Cytoplasmic

Proteinase 3 (PR3-ANCA), BPI, others

Myeloperoxidase (MPO-ANCA), elastase, others

**ANCA-Associated Pulmonary Lesions**

- Hemorrhagic Capillaritis
- Necrotizing Granulomatus Inflammation
- Eosinophilic Inflammation
Microscopic Polyangiitis

Necrotizing vasculitis with few or no immune deposits affecting small vessels, i.e. capillaries, venules, or arterioles. Necrotizing arteritis involving small and medium-sized arteries may be present. Necrotizing glomerulonephritis is very common. Pulmonary capillaritis often occurs.

Wegener's Granulomatosis

Granulomatous inflammation involving the respiratory tract, and necrotizing vasculitis affecting small to medium-sized vessels, e.g. capillaries, venules, arterioles, and arteries. Necrotizing glomerulonephritis is common.

ANCA Pulmonary Granulomatous Inflammation

ANCA Pulmonary Venulitis and Microabscess

Churg-Strauss Syndrome

Eosinophil-rich and granulomatous inflammation involving the respiratory tract and necrotizing vasculitis affecting small to medium-sized vessels, and associated with asthma and blood eosinophilia.
Churg-Strauss Syndrome

**CASE #5-1**

A 54 year old white male developed a “flu-like” illness with fever, arthralgias and myalgias that cleared after about a week, but was followed by the onset of sinusitis and runny red eyes. Two weeks later, he developed a rash on his ankles and lower legs bilaterally. He visited a physician who identified palpable purpura on both lower extremities. The patient was referred to a dermatologist who performed a skin biopsy. A diagnosis of severe leukocytoclastic angitis consistent with hypersensitivity vasculitis was made.
CASE #5-2
He developed massive hemoptysis. Physical examination revealed new and old crops of purpuric skin lesions on the lower extremities, nasal crusting and ulceration, and grossly bloody sputum. Radiographs showed severe sinusitis and extensive pulmonary infiltrates. Laboratory results included serum creatinine 2.1 mg/dl, 3+ proteinuria, 2+ hematuria with dysmorphic erythrocytes, and negative serology for anti-GBM, ANCA, cryoglobulins, and ANA. A nasal biopsy showed “nonspecific” acute and chronic inflammation with no granulomatous inflammation.

CASE #5-3
An open lung biopsy showed acute inflammation and hemorrhage but no granulomatous inflammation. A kidney biopsy showed focal necrotizing glomerulonephritis with 40% crescents but no granulomatous inflammation.

CASE #5-4
A diagnosis of “presumptive” Wegener’s granulomatosis was made by the clinicians and the patient was begun on treatment with high dose corticosteroids and cyclophosphamide.

Pathologic Diagnoses: Cutaneous leukocytoclastic angiitis, acute and chronic sinusitis, pulmonary capillaritis and focal necrotizing glomerulonephritis, most consistent with microscopic polyangiitis but can not rule out Wegener’s granulomatosis

Case #5
Important!

A negative ANCA result does not rule out any of the ANCA-associated vasculitides.

Causes of Pulmonary-Renal Syndrome

<table>
<thead>
<tr>
<th>Cause</th>
<th>Percentage</th>
<th>(Cases/Total Cases)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ANCA Vasculitis</td>
<td>54.5%</td>
<td>(48/88)</td>
</tr>
<tr>
<td>ANCA &amp; Anti-GBM Vasculitis</td>
<td>8.0%</td>
<td>(7/88)</td>
</tr>
<tr>
<td>Anti-GBM Vasculitis</td>
<td>6.8%</td>
<td>(6/88)</td>
</tr>
<tr>
<td>Other</td>
<td>30.7%</td>
<td>(27/88)</td>
</tr>
</tbody>
</table>

* 8 Wegener’s granulomatosis
40 microscopic polyangiitis

Conclusion: ANCA vasculitis, especially microscopic polyangiitis, is the most common cause for pulmonary-renal vasculitic syndrome

Niles et al, Arch Int Med 156:440-5, 1996
CASE #6

A 23 year old male developed lower extremity palpable purpura, arthralgias and abdominal pain. He visited his family physician who made a diagnosis of HS purpura. Urine analysis revealed 2+ proteinuria and 1+ hematuria; consistent with HS purpura nephritis. The physician scheduled him for a return visit in two weeks. At that time, the patient reported the added feature of nausea and a "funny feeling" in his hands and feet. Urine analysis revealed 4+ proteinuria and 3+ hematuria. He had a serum creatinine of 3.5 mg/dl. The patient was referred to a nephrologist who obtained additional laboratory data and performed a renal biopsy.

Summary
What is the diagnosis?

Skeletal Muscle Biopsy  Nerve Biopsy

What is the diagnosis?

Skeletal Muscle Biopsy  Nerve Biopsy

What major category of vasculitis can be ruled out?

What is the diagnosis?

What major categories of vasculitis can be ruled out now?

What is the diagnosis?

What major categories of vasculitis can be ruled out now?

Diagnosis: pauci-immune small vessel vasculitis, most consistent with microscopic polyangiitis but can not rule out Wegener’s granulomatosis or Churg-Strauss syndrome without additional data.
Though we name the things we know, we do not necessarily know them because we name them.

Homer W. Smith
Nephrologist
Part II: Vasculopathies

Monoclonal Immunoglobulin Deposition Disease
Amyloidosis
Antiphospholipid Syndrome
Thrombotic Microangiopathies

Case # 7

70-year-old male
Renal transplantation 6 years ago
Underlying native kidney disease “unknown”
No previous graft biopsies performed
Patient has been “healthy”
Previous “monoclonal gammopathy” (undeterm. signif)

Case # 7 (continued)

Baseline immunosuppression:
  previously: tacrolimus, azathioprine, steroids
  currently: rapamycin, cellcept, steroids
Serum creatinine: 3 mg/dl
Proteinuria: 0.4 grams/24 hrs

Case # 7 (continued)

Diagnostic procedure:
Renal allograft biopsy
( Five images were provided )

Case # 7 (#1, PAS x16)

Case # 7 (#2, H&E x100)
Differential Diagnosis (by LM):

A) Diabetes mellitus
   - versus -
   Monoclonal immunoglobulin deposition disease

B) Calcineurin inhibitor induced toxicity

Diagnosis?
Case 7 (#7, IF: lambda light chains)

Case 7 (#8, EM: glomerulus)

Case 7 (#9 and 10, EM: glomerulus and tubulus)

Diagnosis:

A) Monoclonal immunoglobulin deposition disease (kappa dominant)

B) Calcineurin inhibitor induced arteriolopathy (mild)

Monoclonal Immunoglobulin Deposition Disease (#1)

Definition:

Synthesis and accumulation of structurally abnormal monoclonal light chains (usually kappa) and/or heavy chains (usually gamma) along basement membranes including arterial walls

Organ involvement:

Most frequently kidneys, also lungs, liver, spleen, heart, eyes, bone marrow and others

Monoclonal Immunoglobulin Deposition Disease (#2)

Symptoms:

- Variable, most often proteinuria (> 90% of cases; may result in renal failure)
- Serum and/or urine monoclonal paraproteins in 70%-80% of patients
- Approx. 60% of patients have plasmacytoma; some patients have unclassified lymphoproliferative disorders
Monoclonal Immunoglobulin Deposition Disease (1-3)

**Diagnostic tests:**
Renal biopsy
(immunofluorescence microscopy and electron microscopy mandatory)

Monoclonal Immunoglobulin Deposition Disease (4)

**Diagnostic features (kidneys):**
1) Light microscopy:
   - Nodular glomerulosclerosis (approx. 60% of cases)
   - Crescent formation (approx. 20% of cases)
   - Thickened tubular basement membranes
2) Deposits by IF:
   - tubules (approx. 99% of cases)
   - glomeruli (approx. 80%-95% of cases)
   - arteries (approx. 55% of cases)
   - interstitium (approx. 25% of cases)
3) Deposits by EM:
   - granular, commonly non-fibrillary deposits mainly along basement membranes (100% of cases) (Virchows Archiv (1994), 425: 271-280)

Vascular Deposits and Injury

“Amyloid-Like” (Congo Red negative)
Monoclonal Immunoglobulin Deposition Disease

Differential diagnosis of “nodular” glomerular lesions (by LM)

A) Diabetes mellitus  
B) Amyloidosis 
C) Membranoproliferative GN  
D) Idiopathic nodular glomerulosclerosis

Amyloidosis

Definition:
- Accumulation of "fibrillary" proteins derived from soluble, proteolytically altered circulating precursor proteins. In case of AL-amyloid, precursors are "structurally normal" variable portions of lambda or kappa light chains (compare to monoclonal immunoglobulin deposition diseases).

- The nomenclature of amyloidosis is based on the precursor protein.

- The terms "primary" and "secondary" amyloidosis are "old fashioned".

<table>
<thead>
<tr>
<th>Amyloid</th>
<th>Precursor in serum</th>
<th>Clinical Disease</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>apo SAA</td>
<td>Secondary amyloidosis, Mediterranean fever</td>
</tr>
<tr>
<td>AL</td>
<td>lambda light chains, kappa light chains (3:1)</td>
<td>Primary amyloidosis (e.g. multiple myeloma)</td>
</tr>
<tr>
<td>AH</td>
<td>IgG1</td>
<td>Heavy chain amyloid</td>
</tr>
<tr>
<td>ATTR</td>
<td>transthyretin</td>
<td>Hereditary amyloid polyneuropathy or cardiomyopathy</td>
</tr>
<tr>
<td>AGei amyloidosis</td>
<td>gelsoin</td>
<td>Finnish familial</td>
</tr>
<tr>
<td>A(ApoA1)</td>
<td>apoA1</td>
<td>Familial amyloid polyneuropathy</td>
</tr>
<tr>
<td>A(beta2)M</td>
<td>beta2-microglobulin</td>
<td>Amyloid found with dialysis</td>
</tr>
</tbody>
</table>
Amyloidosis

A) Systemic:
- Often kidneys, GI tract, liver, spleen, heart, nerves
  1) AL amyloidosis (precursors: normal light chains)
  2) AA amyloidosis (precursor: normal serum amyloid A, SAA)
  3) ATTR amyloidosis (precursor: typically mutated transthyretin)
  4) Aβ2M amyloidosis (precursor: normal β2 microglobulin)
  Often nerves and heart, less frequent kidneys

B) Localized: e.g. brain, heart (senile amyloidosis), pancreas
- Aβ amyloid (β protein; brain)
- A amylin (pancreas islets)

Amyloidosis: Histologic tests

A) General detection (all forms of amyloid)
  1) Congo Red stain: high sensitivity, high specificity
  2) Serum amyloid P component (SAP): high sensitivity, low specificity
  3) Thioflavine T: high sensitivity, low specificity
  4) Electron microscopy: high sensitivity, high specificity
      (8-12 nm non-branching fibrils)

B) Subtyping
  1) Immunohistochemistry
  2) Congo Red incubation with KMNO4 treatment
     AL amyloid: KMNO4 resistant
     other types of amyloid: KMNO4 sensitive

Amyloidosis: Vascular amyloid deposits:

Arterioles > larger arteries > capillaries > veins

Deposits initially surround smooth muscle cells, medial cell necrosis, progressive transmural amyloid deposits
vascular wall thickening, wall rigidity, ischemic injury

Ischemic type tissue atrophy (occasional hemorrhage)

Uncommon: pseudoaneurysms, thrombosis
Amyloid vasculopathy in the brain

IHC: β amyloid

Right "racoon eye sign" due to amyloidosis (often AL type)


**AL Amyloid**

A) Congo Red LM
B) Congo Red (polarized light)
C) Congo Red KMnO4 Pre-treatment

Immunohistochemistry on formalin fixed tissue
**Amyloidosis**

**Most common systemic forms:**
1) AL type:
   - caused by plasma cell dyscrasia / multiple myeloma
   - lambda (V) / kappa (I) light chain deposits, 3:1

2) AA type:
   - caused by "chronic inflammatory conditions"

**AA Amyloidosis**

<table>
<thead>
<tr>
<th>Disease</th>
<th>Lifetime incidence of AA amyloidosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rheumatoid arthritis</td>
<td>3 – 10%</td>
</tr>
<tr>
<td>Psoriatic arthritis</td>
<td>3 – 13%</td>
</tr>
<tr>
<td>Chronic juvenile arthritis</td>
<td>0.14 – 17%</td>
</tr>
<tr>
<td>Ankylosing spondylitis in children</td>
<td>4 – 5%</td>
</tr>
<tr>
<td>Inflammatory bowel disease</td>
<td>0.4% – 2%</td>
</tr>
<tr>
<td>Chronic inflammation</td>
<td></td>
</tr>
<tr>
<td>(TB, bronchiectasis, osteomyelitis</td>
<td>up to 10%</td>
</tr>
<tr>
<td>pyelonephritis, leprosy)</td>
<td></td>
</tr>
</tbody>
</table>

**Diagnostic work-up for (systemic) amyloidosis**

- **Biopsy** (containing arterioles / small arteries)
  - kidney
  - abdominal subcutaneous tissue
  - deep rectal / GI
  - liver
  - synovium in case of dialysis related AL, M amyloidosis

**Electron microscopy**

A) Amyloid is destructive
B) Non-branching fibrils (8-12 nanometers)
Amyloidosis: Outcome

- Dependent on type of amyloid
- Dependent on amyloid load
  - Dependent on organ involvement
  - Dependent on underlying disorder

Median survival:
- AL amyloidosis 1 year
- AA amyloidosis 2 - 4 years
- ATTR amyloidosis 10-15 years

Amyloidosis

…..always suspect amyloid…..

Differential diagnosis of vascular amyloid deposits (LM):

- Severe subendothelial hyalinosis / arteriolosclerosis (PAS+)
- Thrombotic microangiopathy with severe hyalinosis of the media (PAS+)

Case # 8

45-year-old male
Past medical history:
At age 15 digital ischemia with amputation of one toe
At age 20 claudication of the right leg
Small bowel perforation
Between ages 30 – 35 several transient CNS attacks

Case # 8 (continued)

At age 40 myocardial infarction
On current admission: bowel infarction
  - sepsis
  - acute renal failure

Two images were provided from the bowel resection specimen (mesenteric vessels)
Diagnosis:

Organizing thrombosis
(suggestive of antiphospholipid syndrome, APS)

Differential-Diagnosis

1) All thrombotic events

2) If small arteries are affected: thrombotic microangiopathies
Antiphospholipid Syndrome (#1)

**Definition:**
Hypercoagulable state with arterial and/or venous thrombus formation due to high titers of circulating antibodies against phospholipid-protein complexes - often the β2 glycoprotein 1 / phospholipid complex - on platelets and/or endothelial cells.

- Disruption of phospholipid dependent anticoagulant mechanisms?
- Induction of expression of procoagulant endothelial molecules?
- Endothelial cell injury/activation prerequisite/1st hit?

Primary APS: No underlying connective tissue disorder

Secondary APS: Associated with other autoimmune diseases, i.e. lupus erythematosus

Antiphospholipid Syndrome (#2)

**Organ involvement:**
Highly variable (thrombi may be limited to veins or arteries)

- Venous thrombosis: legs, kidney, liver, retina
- Arterial thrombosis: brain, heart, eye, bowel, legs, kidney

Kidney: sometimes additional thrombotic microangiopathies (TMA)

Heart: Libman-Sacks endocarditis (4%)

Rarely (1%) simultaneous multiple organ involvement (= catastrophic APS)

Antiphospholipid Syndrome (#3)

**Symptoms:**
- Variable, caused by ischemia
- Pulmonary hypertension (secondary to embolization) in 20%
- Renal hypertension
- Livedo reticularis
- Miscarriages
- Thrombocytopenia, hemolysis and shistocytes (often in cases of a concurrent TMA)

Antiphospholipid Syndrome (#4)

**Diagnostic tests:**
- Detection of HIGH titers of IgG (or IgM) autoantibodies against cardiolipin/protein complexes (e.g. β2 glycoprotein 1 / phospholipid complex)
- Lupus anticoagulant antibodies (detected on 2 occasions)

Antiphospholipid Syndrome (#5)

**Histologic features:**
1) Fibrin thrombi of various ages (often with recanalization) in arteries and/or veins
2) Reactive fibrous intimal hyperplasia (often in smaller caliber arteries) with marked proliferation of myofibroblasts and fibrin deposits
3) Occasionally thrombotic microangiopathy (in the kidney; 30% of patients with renal involvement)
4) Signs of ischemic atrophy and fibrosis

Livedo reticularis
(from: PH McKee, Pathology of the Skin, 2nd edition, Mosby-Wolfe publishers)
Antiphospholipid Syndrome

**Histologic differential diagnosis**

A) “Other” causes of thrombosis / intimal remodeling
B) Other causes of a thrombotic microangiopathy
C) Lupus erythematosus

---

Case # 9

54-year-old Caucasian female
Metastatic neuroendocrine tumor for 10 months
Previous unsuccessful radiation and fluorouracil treatment
Additional therapy with ‘DOTATOC’ (dota-D-phe-tyroctreotide)
Recent deterioration of renal function
Proteinuria: 2 grams/24 hrs
Clinical evidence of hemolysis; low platelets

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Case # 9 (continued)

*Diagnostic procedure:*
Renal biopsy
( Four images were provided )

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Histopathologic renal lesions in primary APS (16 patients)

- Arteriosclerosis (12) 75%
- Fibrous intimal hyperplasia (12) 75%
- Tubular thyroidization (12) 75%
- Arteriolar occlusions (11) 68%
- Focal cortical atrophy (10) 62%
- Organizing thrombosis (6) 37%
- Thrombotic microangiopathy (5) 31%

Data kindly provided by Dominique Nochy and Gary Hill (Paris)
Diagnosis:
Thrombotic microangiopathy
(HUS-like - radiation induced/DOTATOC)

Differential-Diagnosis
Other potential causes of a TMA
**Thrombotic Microangiopathies (TMA)**

**Definition:**
Systemic or localized vascular "occlusive" disease involving very small arteries, arterioles and capillaries due to severe endothelial and smooth muscle cell injury

**Organ involvement:**
Most frequently kidneys (HUS) and brain (TTP), also GI tract (colon), liver, heart, pancreas

**Causes of TMA (Table 6)**

- **Idiopathic**
- **Autoimmune Diseases**
  - Systemic Lupus Erythematosus (SLE / APS)
  - Scleroderma
- **Malignant Hypertension**
- **Enterohemorrhagic E. coli – HUS D+**
  - (typically E. coli 0157:H7)
- **Other infectious agents**
  - Streptococcus pneumoniae
  - HIV/AIDS
- **Abnormal vWF cleavage**
  - Typical TTP
- **Pregnancy/ Postpartum**
  - (HELLP syndrome)
- **Drug Therapy**
  - Chemotherapy (mitomycin, cisplatin, bleomycin etc)
  - Immune mediated drug toxicity (quinine, ticlopidine, clopidogrel)
  - Immunosuppressive drugs (FK 506/ cyclosporine)
  - Contraceptives
- **Radiation (Dotatoc)**
- **Bone marrow / peripheral stem cell transplantation**

**DOTATOC**

- Binds to the somatostatin receptor
- Is conjugated to the beta emitter yttrium 90
- Is eliminated by glomerular filtration
- Is re-absorbed by proximal tubules

  - "Internal" renal radiation
  - TMA is dose dependent (> 200 miCi/m²)

**Thrombotic Microangiopathies Clinical Symptoms/ Laboratory Data:**

- Renal dysfunction / failure (HUS)
- Sudden onset of neurologic disorders / seizures (TTP)
- Thrombozytopenia
- Hemolytic anemia / reticulocytosis / LDH
- Fragmented red blood cells / shistocytes
- Hypertension
- Occasionally (bloody) Diarrhea (D+ versus D - cases)
- Occasionally abdominal pain
- Occasionally fever

*Caveat:* Sometimes only very minor symptoms!

**PATHOGENIC EVENTS LEADING TO TMA**

```
SEVERE ENDOTHELIAL CELL INJURY / NECROSIS

- Endothelial swelling with increased vascular shear stress
- Leukocyte adhesion to damaged endothelium
- Complement activation
- Loss of endothelial thromboresistance
- Abnormal vWF multimer fragmentation
- Platelet activation and aggregation

THROMBOTIC MICROANGIOPATHY

ISCHEMIA / Hemorrhage
```

**Scleroderma**

Macrosopic changes secondary to TMA

**Malignant hypertension (kidney)**

**Malignant hypertension**
The etiology of TMAs can often not be unveiled based on morphologic examination.

Histologic features:
Characteristic changes by light microscopy in arterioles/ small arteries and/or glomeruli

(Immunofluorescence microscopy and electron microscopy in kidney biopsies adjunct tools)

Larger arteries (interlobar) spared.

Thrombosis Microangiopathies
Changes in small arteries / arterioles

Early: - Severe intimal swelling (often “mucoid”)
- Red blood cell extravasation into intima and media
- Fibrin insudation into intima and media and necrosis
- Nodular proteinaceous deposits in media (PAS positive, secondary to single medial cell necrosis)

Intermediate: - Cellular intimal remodeling (rich in myofibroblasts)

Late / sclerosing: - Intimal sclerosis (“onion skinning”)
- Pronounced transmural and intraluminal proteinaceous insudates
- Stenosis (secondary ischemic atrophy)
Thrombotic Microangiopathies

Changes in glomeruli

**Early**
- Endothelial cell swelling
- Fibrin / platelet thrombi
- Segmental minimal duplication of the GBM
- Mesangiolysis
- Focal (segmental) cellular crescent formation

**Late**
- Pronounced double duplication of GBM
- EM: “flocculent material”, no or small deposits only
- Mesangiolysis
- Focal fibrous crescent formation
- Secondary FSGS

The etiology of TMAs can often not be unveiled based on morphologic examination: 
*HUS versus TTP*
Hemolytic Uremic Syndrome (HUS) and Thrombotic Thrombocytopenic Purpura (TTP)

!! Vast Clinical Overlap !!

**Hemolytic Uremic Syndrome:**
- Severe renal involvement
- Typically occurs in children
- Often associated with bloody diarrhea (D+)
- Prognosis often good

**Thrombotic Thrombocytopenic Purpura:**
- Predominant extra-renal involvement with prominent neurological symptoms.
- Most often occurs in adults
- Often multi organ involvement
- Often less favorable prognosis

Pathophysiology of typical diarrhea associated HUS

- (E. coli O157:H7)
- Shigella dysenteriae
- Shiga like toxins enter intestinal circulation → surface of platelets and monocytes → binding to Gb3 receptors on endothelial cells (high density in kidney) → endothelial cell injury/necrosis

Differences in the pathobiology:

**Diarrhea HUS:**
- Gb3 receptor mediated, toxin induced
- Endothelial necrosis

**TTP:**
- Clotting due to very low ADAMTS13 activity (< 5%)
- Platelet rich thrombi


(Recurrent) TTP: "clinical" TTP

... or could it rather be APS?
**Histopathologists role:**

- diagnose TMA
- stage TMA (early versus late)
- make clinician search for etiology

**Differential diagnosis of glomerular changes:**

Membranoproliferative glomerulonephritis type I

**Features distinguishing TMA from MPGN**

1) TMA much less cellular  
2) TMA with little or no cell interpositions  
3) TMA with prominent mesangiolysis  
4) TMA no specific deposits by IF  
5) TMA with 'flocculent' material by EM

**Thrombotic microangiopathy**

**Differential diagnosis of vascular changes**

**Table 7**

<table>
<thead>
<tr>
<th>Pathology</th>
<th>Predominant pathology</th>
<th>Major pathogenic event</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemolytic anemia</td>
<td>Glomerular capillary</td>
<td>Endothelial injury caused by many different etiologies, predominantly</td>
</tr>
<tr>
<td>Hemolytic anemia</td>
<td>subendothelial expansion,</td>
<td>in renal vessels, causing renal injury, thrombosis and renal ischemia</td>
</tr>
<tr>
<td>Hemolytic anemia</td>
<td>arteriolar fibrinoid necrosis, arteriolestenosis, intimal hypertrophy</td>
<td></td>
</tr>
<tr>
<td>Thrombotic thrombocytopenia purpura</td>
<td>Platelet-rich thrombi in many organs</td>
<td>Enhanced platelet aggregation, e.g. caused by abnormal vWF cleavage and increased von Willebrand factor</td>
</tr>
<tr>
<td>Disseminated intravascular coagulation</td>
<td>Fibro-fibrin-rich thrombi in many organs</td>
<td>Enhanced coagulation and fibrinolysis, e.g. caused by thrombocytopenia and decreased tPA activity</td>
</tr>
<tr>
<td>Anti-phospholipid antibody syndrome</td>
<td>Thrombosis in veins, arteries, arterioles and capillaries, and glomerular subendothelial expansion and GBM remodeling</td>
<td>Antibodies to phospholipid or phospholipid-binding protein that bind to endothelial cells or platelets to cause thrombosis</td>
</tr>
<tr>
<td>Prothrombosis / Eclampsia</td>
<td>Glomerular capillary endothelial swelling (endotheliosis)</td>
<td>Abnormal proliferation with generation of tumoral facades that injure maternal endothelial cells</td>
</tr>
</tbody>
</table>

**Thrombotic Microangiopathies**

Long term prognosis:

- Changes limited to glomeruli favorable outcome (i.e. shiga toxin induced D+ HUS of infancy)
- Changes involving small arteries less favorable (ischemic injury; renal hypertension)
- Early changes potentially treatable / reversible
- Sclerosing changes irreversible
Part III Transplant Vascular Disease
- Calcineurin inhibitor induced toxicity
- Vascular Rejection

Case # 10
59-year-old African American male
Renal transplantation 6 years ago
Underlying native kidney disease
“arterionephrosclerosis”
No previous graft biopsies performed
Patient has been “healthy”

Case # 10 (continued)
Immunosuppression:
tacrolimus (trough levels 10 ng / ml)
Cellcept
steroids
Serum creatinine:
  baseline: 1.8 mg/dl
  current peak: 2.6 mg/dl
Proteinuria: 1.2 grams/24 hrs
Hematuria: trace

Case # 10 (continued)
Diagnostic procedure:
Renal allograft biopsy
( Four images were provided )
Diagnosis:
Calcineurin inhibitor induced toxicity:
1) arteriolopathy (moderate)
2) glomerulopathy (mild) with secondary focal and segmental glomerulosclerosis
3) striped interstitial fibrosis (mild to moderate)

Calcineurin inhibitor induced toxicity
Induced by cyclosporine or tacrolimus
Affects native kidneys and renal allografts
Functional and morphological / structural changes
Linked to dose (wide response range)
Calcineurin inhibitor induced toxicity

Morphological changes

A) Typical (not pathognomonic):
- Tubulopathy (proximal tubules) reversible
- Arteriolopathy (afferent arterioles) largely irreversible
- Glomerulopathy largely irreversible

B) Secondary (non-specific):
- Nephron loss (striped fibrosis)
- Focal segmental glomerulosclerosis

Calcineurin inhibitor induced toxicity

- Arteriolar AND glomerular changes identical to thrombotic microangiopathies (TMA)
- TMA like changes range from mild (common) to severe (uncommon)
- Systemic signs of TMA typically absent
- Non-specific IF staining pattern (in arterioles and glomeruli: IgM, C3, C4d)
  !! No C4d along peritubular capillaries !!

Early Arteriolopathy

Major: thrombotic microangiopathy (C4d -)
Minor: swelling ("ballooning") of medial smooth muscle cells due to dilated endoplasmatic reticulum
Calcineurin inhibitor induced toxicity

Late Arteriolopathy

Hyaline nodules replacing smooth muscle cells (due to single cell necrosis)

Differential Diagnosis:

A) Other causes of a thrombotic microangiopathy (e.g., recurrent disease)
B) Hypertension / diabetes induced arteriolosclerosis

Calcineurin Inhibitor induced Arteriolopathy

Differential Diagnosis:

A) Other causes of a thrombotic microangiopathy (e.g., recurrent disease)
B) Hypertension / diabetes induced arteriolosclerosis

Calcineurin inhibitor induced toxicity

Differential diagnosis

A) Arteriolopathy

1) Hyalinosis due to hypertension
   (generally no proteinaceous nodules in MEDIA)
2) Recurrent TMA (rare)
   (DD: underlying kidney disease)
3) Diabetes mellitus
   (DD: clinical history)

Calcineurin inhibitor induced toxicity

Glomerulopathy

a) Early major: Fibrin thrombi (TMA; C4d -)
b) Late major: GBM duplication with rare cell interposition, no proliferations
   no significant immune complexes
   C4d -

Calcineurin inhibitor induced glomerulopathies (early or late) are ALWAYS associated with calcineurin inhibitor induced arteriolopathies (early or late)
Calcineurin inhibitor induced toxicity

Differential diagnosis

Glomerulopathy

1) Recurrent TMA (rare)
   (DD: underlying kidney disease)
2) Membranoproliferative GN
   (DD: more cellular, immune complex deposits by IF and EM)
3) Transplant Glomerulopathy
   (DD often: C4d positivity in peritubular capillaries, tx glomerulitis, signs of vascular rejection)

Late “non-diagnostic” phenomena:

Marked arteriolopathy and glomerulopathy result in nephron loss → striped fibrosis → secondary FSGS

Striped interstitial fibrosis
Calcineurin inhibitor induced toxicity

**Therapy / Outcome**

1) Generally dose reduction or discontinuation of calcineurin inhibitors
2) Outcome favorable (except for severe and diffuse TMA or severe and diffuse late arteriopathy)
3) Mild and moderate changes are partially reversible

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**Case # 11**

- 42-year-old African American female
- Heart transplantation in 1/2002
- Underlying native heart disease: severe atherosclerosis remote infarcts
- Immunosuppression with tacrolimus, Cellcept and prednisone (? compliance)
- Multiple rejection episodes during first 2 years
- 1/2004: coronary angiogram with focal 20% stenosis

On admission 7/2004:
- chest pain
- occlusion right coronary artery
- ECG with signs of infarction
- No anti-rejection therapy

Patient expired 4 days after admission

( Six images were provided )
Diagnosis: Sclerosing transplant vasculopathy with intimal inflammation (no intramyocardial capillary C4d accumulation) ‘So-called chronic active vascular rejection’
**Sclerosing Transplant Vasculopathy**

**‘Chronic Vascular Rejection’**

**Definition:**
Sclerosis and fibrous thickening of the intima of arteries induced by vascular rejection episodes
- inactive (scarred)
- active (intimal inflammation, C4d positivity along parenchymal capillaries)

**Organ involvement:**
Most frequently kidney and heart allografts, also seen in pancreas, rarely liver and lung tx

**Histologic features:**
- Intimal sclerosis due to collagen I and III deposition
- Intimal hypercellularity (irregular cell distribution, “busy look”)
- Occasional foam cells
- Enlarged ‘activated’ endothelial cells
- Occasional ‘neo-media’ formation
- Only scant elastic fibers
  (!! No fibroelastosis !!)

**The lack of abundant elastic lamellae in the intima is THE best tool to distinguish sclerosing vascular rejection from hypertension induced arteriosclerosis**

**Note:** Differential diagnosis of intimal sclerosis without elastosis
1) sclerosing vascular allograft rejection
2) thrombotic microangiopathy
3) adaptive intimal fibrosis in end-stage kidneys

**Transplant endarteritis is commonly preceding sclerosing transplant vasculopathy**
Within 2 months after tx:
Transplant endarteritis

3 years after tx:
Sclerosing transplant vasculopathy
(inactive)

Allograft Rejection involving Arteries

Transplant Endarteritis
- cellular
  - C4d negative
- cellular and humoral
  - C4d positive

Sclerosing Transplant Vasculopathy
- so-called chronic vascular rejection

A New Monkey Model of Chronic Kidney Rejection

“Early” arterial changes

CD 3

Macrophages CD 68

Cell proliferation Ki 67

Myofibroblasts alpha SMA
“Late” arterial changes
Chronic active vascular rejection / Sclerosing transplant vasculopathy with inflammation

Sclerosing Transplant Vasculopathy ‘Chronic Vascular Rejection’

Long term prognosis:
Dependent on intimal sclerosis/stenosis/activity
- Often unfavorable with graft loss
- Therapeutic attempts may be beneficial in cases with activity

Differential-Diagnosis
Severe hypertension induced arteriosclerosis
(prominent elastic lamellae in intima, no or minimal lymphocytes, little hypercellularity)
<table>
<thead>
<tr>
<th>Sclerosing Tx vasculopathy</th>
<th>Arteriosclerosis</th>
</tr>
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<tbody>
<tr>
<td>![Image 1]</td>
<td>![Image 2]</td>
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