

An Algorithmic Approach to Renal Biopsy Interpretation of Glomerular Diseases

J. Charles Jennette, MD

Brinkhous Distinguished Professor and Chair of Pathology and Laboratory Medicine

University of North Carolina at Chapel Hill, Chapel Hill, NC, USA

The diagnosis of glomerular disease in renal biopsy specimens often has at least 5 steps that may occur in different sequences: 1) preliminary review of available clinical data prior to specimen examination, 2) light microscopic examination, 3) immunohistologic examination, 4) electron microscopic examination, and 5) integration of all pathologic and clinical data into a final interpretation and diagnosis.

Step 1) Preliminary review of available clinical data: A pathologic diagnosis should ultimately be based on the pathologic findings; however, if you know what you are looking for you are more likely to find it. An important caveat is to not let what is expected based on clinical findings to cause you to see what is not present in the specimen. I have had occasional specimens referred to me for a second opinion that had a detailed description of the disease that was stated in the diagnostic line, only to find that the injury actually in the specimen was very different (i.e. the description in the report matched the diagnosis perfectly, but the findings in the specimens did not match the description).

Table 1 lists most of the categories of glomerular disease and their relative frequency among native kidney renal biopsy specimens evaluated in the UNC Nephropathology Laboratory over a number of years. Bear in mind that these are the frequencies of these diseases in patients who are selected for renal biopsy and thus is different from the frequency of these diseases among all patients with a particular clinical presentation. The clinical presentation and frequency of the diseases on his list will influence the hierarchy of the differential diagnosis prior to examining the biopsy specimen.

Table 1. Relative frequency of various diagnoses among 7257 native kidney biopsy specimens evaluated in the University of North Carolina Nephropathology Laboratory. (Modified from Jennette JC, Falk RJ: Glomerular Clinicopathologic Syndromes, in Greenberg A, Cheung AK, Coffman TM, Falk RJ, Jennette JC, eds., Primer on Kidney Diseases, 5th Ed., National Kidney Foundation, 2009.)

Diseases that typically cause the nephrotic syndrome (42% of all native renal biopsy specimens, n=3067)

- Focal segmental glomerulosclerosis (all variants) (920)
- Idiopathic membranous glomerulopathy (847)
- Minimal change glomerulopathy (398)
- Diabetic glomerulosclerosis (246)
- Type I membranoproliferative glomerulonephritis (190)
- Idiopathic mesangioproliferative glomerulonephritis (145)
- Amyloidosis (108)
- C1q nephropathy (99)
- Fibrillary glomerulonephritis (59)
- Monoclonal immunoglobulin deposition disease (26)
- Dense deposit disease (Type II membranoproliferative glomerulonephritis) (14)
- Preeclampsia/eclampsia (6)
- Immunotactoid glomerulopathy (6)
- Collagenofibrotic glomerulopathy (3)

Table 1 continued.

Diseases that typically cause hematuria and nephritis (29% of biopsies, n=2109)

- Lupus nephritis (all classes) (636)
- IgA nephropathy (538)
- Idiopathic immune complex proliferative glomerulonephritis (375)
- Pauci-immune/antineutrophil cytoplasm antibody glomerulonephritis (301)
- Postinfectious acute diffuse proliferative glomerulonephritis (86)
- Thin basement membrane lesion (82)
- Anti-glomerular basement membrane antibody glomerulonephritis (56)
- Alport syndrome (35)

Diseases other than glomerulonephritis that typically cause acute renal failure (5% of biopsies, n=371)

- Thrombotic microangiopathy (all types) (126)
- Acute tubulointerstitial nephritis (101)
- Acute tubular necrosis (69)
- Atheroembolization (34)
- Light chain cast nephropathy (31)
- Cortical necrosis (10)

Diseases other than glomerulonephritis that typically manifest as chronic renal failure (8%, n=583)

- Arterionephrosclerosis (229)
- Chronic sclerosing glomerulonephritis (166)
- End-stage renal disease not otherwise specified (114)
- Chronic tubulointerstitial nephritis (74)

Miscellaneous other diseases (3%, n=199)

Adequate tissue with nonspecific abnormalities (5%, n=370)

No pathologic lesion identified (2%, n=141)

Inadequate tissue for definitive diagnosis (6%, n=417)

Step 2) Light microscopic examination: Thin (~3 micrometer) paraffin sections should be examined with appropriate stains (e.g. H&E, PAS, Jones, Masson) at multiple levels of section. The different tissue compartments should be evaluated systematically, i.e. glomeruli, tubules, interstitium, and arteries and arterioles. Terms commonly used to describe alterations in glomeruli are defined in Table 2.

Table 2. Glossary of terms used to describe histologic lesions in glomeruli (Modified from from Jennette JC, Olson JL, Schwartz MM, Silva FG: Primer on the Pathologic Diagnosis of Renal Disease in Heptinstall's Pathology of the Kidney, 6th Edition, Jennette JC, Olson JL, Schwartz MM, Silva FG (eds), LWW, 2007, Chapter 3.

Focal	Involving <50% of glomeruli
Diffuse	Involving 50% or more of glomeruli
Segmental	Involving part of a glomerular tuft
Global	Involving all of a glomerular tuft
Mesangial hypercellularity	4 or more nuclei in a peripheral mesangial segment
Endocapillary hypercellularity	Increased cellularity internal to the GBM composed of leukocytes, endothelial cells and/or mesangial cells
Extracapillary hypercellularity	Increased cellularity in Bowman's space, i.e. > one layer of parietal or visceral epithelial cells, or monocytes/macrophages

Table 2 continued.

Crescent	Extracapillary hypercellularity other than the epithelial hyperplasia of collapsing variant of FSGS
Fibrinoid necrosis	Lytic destruction of cells and matrix with deposition of acidophilic fibrin-rich material
Sclerosis	Increased collagenous extracellular matrix that is expanding the mesangium, obliterating capillary lumens or forming adhesions to Bowman's capsule
Hyaline	<u>Glassy</u> acidophilic extracellular material
Membranoproliferative	Combined capillary wall thickening and mesangial or endocapillary hypercellularity
Lobular (hypersegmented)	Consolidated expansion of segments that are demarcated by intervening urinary space
Mesangiolytic	Detachment of the paramesangial GBM from the mesangial matrix or lysis of mesangial matrix

Table 3, lists the major patterns of glomerular injury that can be observed by light microscopy. Note that a specific category of disease (e.g. lupus nephritis, IgA nephropathy, post-infectious glomerulonephritis) can cause more than one pattern of injury.

Table 3: Patterns of glomerular injury observed by light microscopy and some but not all of the diseases that can cause each patterns of injury.

No abnormality by light microscopy:

1. No glomerular disease
2. Glomerular disease with no light microscopic changes (e.g. minimal change glomerulopathy, thin basement membrane nephropathy)
3. Mild or early glomerular disease (e.g. ISN/RPS Class I lupus nephritis, IgA nephropathy, C1q nephropathy, membranous glomerulopathy, amyloidosis, Alport syndrome, etc.)

Thick capillary walls without hypercellularity or mesangial expansion:

1. Membranous glomerulopathy (primary or secondary) (>Stage I)
2. Thrombotic microangiopathy with expanded subendothelial zone
3. Preeclampsia/eclampsia with endothelial swelling
3. Fibrillary glomerulonephritis with predominance of capillary wall deposits

Thick walls with mesangial expansion but little or no hypercellularity:

1. Diabetic glomerulosclerosis with diffuse rather than nodular sclerosis
2. Secondary membranous glomerulopathy with mesangial immune deposits
3. Amyloidosis
4. Monoclonal immunoglobulin deposition disease
5. Fibrillary glomerulonephritis
6. Dense deposit disease (type II membranoproliferative glomerulonephritis)

Focal segmental glomerular sclerosis without hypercellularity:

1. Focal segmental glomerulosclerosis (primary or secondary)
2. Chronic sclerotic phase of a focal glomerulonephritis
3. Hereditary nephritis (Alport syndrome)

Table 3 continued.

Mesangial or endocapillary hypercellularity:

1. Focal or diffuse mesangioproliferative glomerulonephritis*
2. Focal or diffuse (endocapillary) proliferative glomerulonephritis*
3. Acute (exudative) diffuse proliferative postinfectious glomerulonephritis
4. Membranoproliferative glomerulonephritis (type I, II or III)

Extracapillary hypercellularity:

1. ANCA crescentic glomerulonephritis (paucity of immunoglobulin by IFM)
2. Immune complex crescentic glomerulonephritis ((granular immunoglobulin by IFM)
3. Anti-GBM crescentic glomerulonephritis (linear immunoglobulin by IFM)
4. Collapsing variant of focal segmental glomerulosclerosis (including HIV nephropathy)

Membranoproliferative, lobular or nodular pattern:

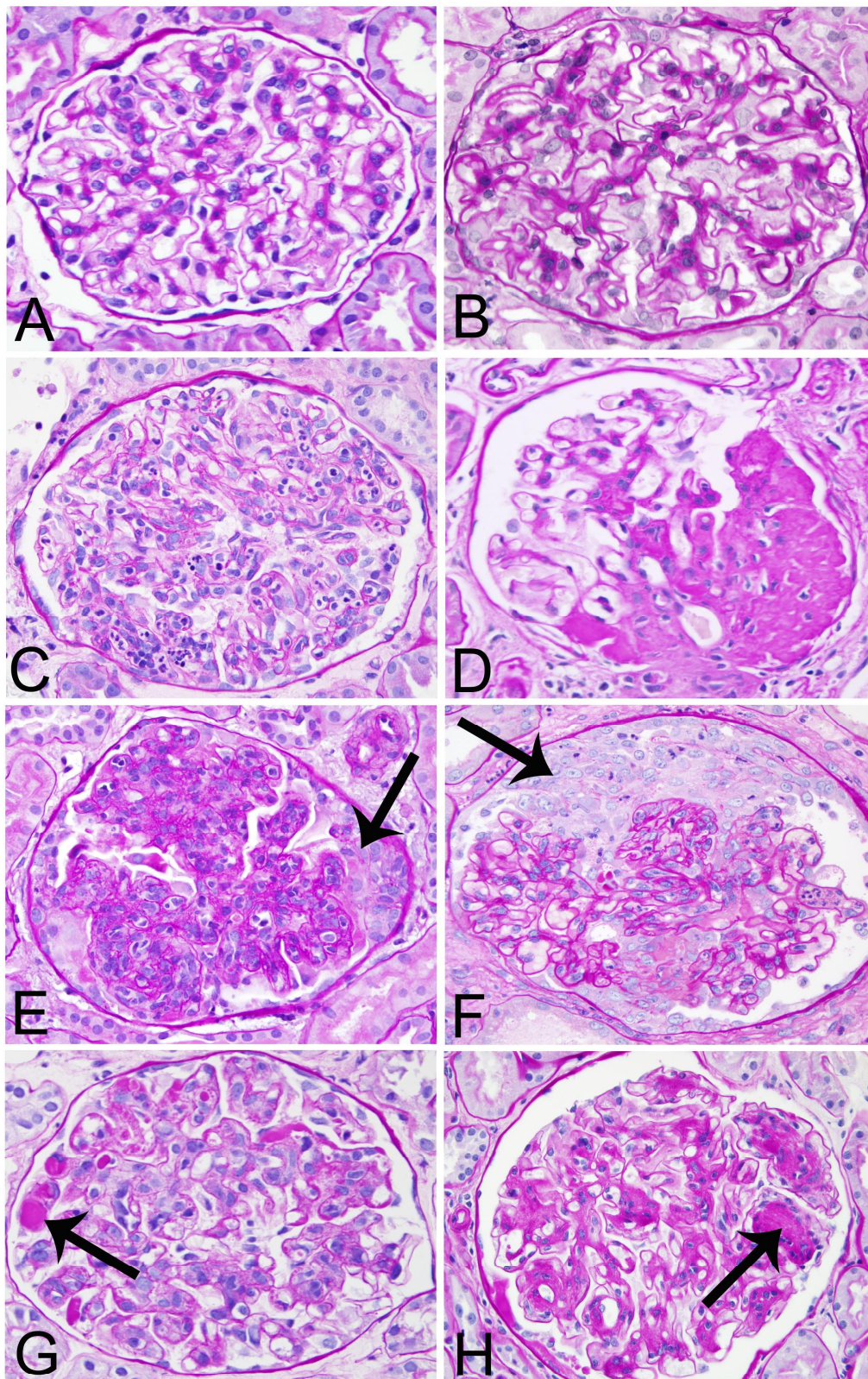
1. Membranoproliferative glomerulonephritis (type I, II/DDD, or IIIB/IIIS)
2. Diabetic glomerulosclerosis with nodular mesangial expansion (KW nodules)
3. Monoclonal immunoglobulin deposition disease with nodular sclerosis
4. Idiopathic (smoking associated) nodular glomerulosclerosis
5. Thrombotic microangiopathy
6. Fibrillary glomerulonephritis
7. Immunotactoid glomerulopathy

Advanced diffuse global glomerular sclerosis

1. End stage glomerular disease
2. End stage vascular disease
3. End stage tubulointerstitial disease

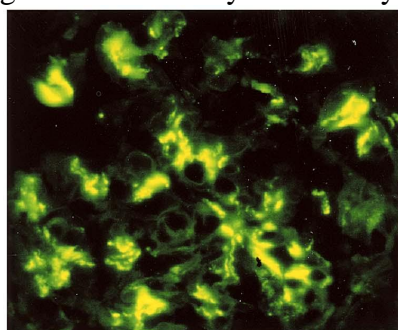
Figure 1 on the following page illustrates some of the patterns of glomerular injury observed by light microscopy (all stained with PAS). Panel A shows membranous glomerulopathy with thick capillary walls without hypercellularity or mesangial expansion. Panel B shows very mild segmental mesangial hypercellularity (e.g. ~5 nuclei in mesangial matrix at ~8:00). Panel C shows global endocapillary hypercellularity with numerous neutrophils in a patient with acute diffuse proliferative GN caused by streptococcal infection. Panel D shows perihilar segmental sclerosis and hyalinosis in a patient with FSGS. Panel E shows global endocapillary hypercellularity and two small foci of extracapillary hypercellularity (e.g. arrow) in a patient with Class IV-G lupus GN. Panel F shows a cellular crescent (arrow) and a focus of segmental fibrinoid necrosis (~6:00) in a patient with ANCA disease. Panel G shows capillary wall thickening, mild endocapillary hypercellularity and hyaline thrombi in a patient with cryoglobulinemic glomerulonephritis. Panel H shows nodular glomerulosclerosis in a patient with diabetic glomerulosclerosis (arrow points to a small KW nodule).

Figure 1. Patterns of glomerular injury observed by light microscopy (all stained with PAS). See text for explanation. (Reproduced from Heptinstall's Pathology of the Kidney, 6th Edition, Jennette JC, Olson JL, Schwartz MM, Silva FG (eds), LWW, 2007, Chapter 3, 100-126)

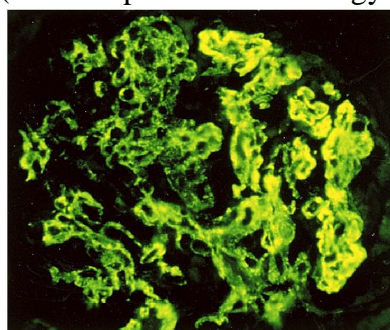


Step 2) Immunohistologic microscopic examination: Immunofluorescence or immunohistochemistry are required for diagnosis of glomerular disease.

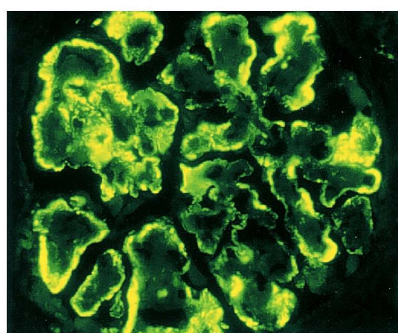
Figure 2: Some of the staining patterns that must be discerned when evaluating glomerular diseases by direct immunofluorescence microscopy. Antibody specificity is shown in parentheses. The patterns of staining are mesangial in IgA nephropathy, mesangial and capillary wall in proliferative lupus GN, peripheral capillary wall granular in type I MPGN, band-like capillary wall and coarsely granular mesangial in DDD (type II MPGN), coarsely granular capillary wall in acute postinfectious GN, finely granular capillary wall in membranous glomerulopathy, linear GBM staining in anti-GBM glomerulonephritis, and coarsely granular to chunky in fibrillary GN. (From Heptinstall's Pathology of the Kidney).



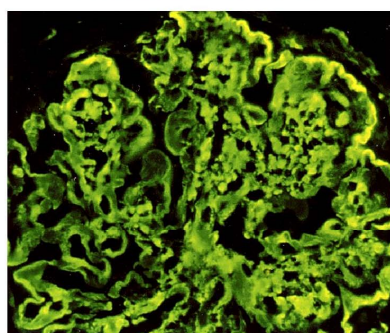
Mesangioproliferative GN (IgA)



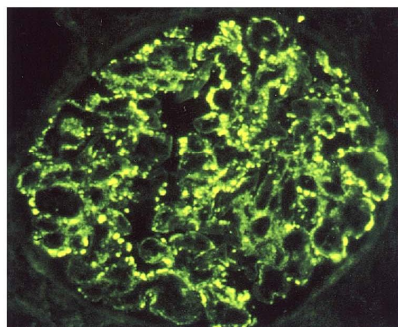
Proliferative GN (IgG)



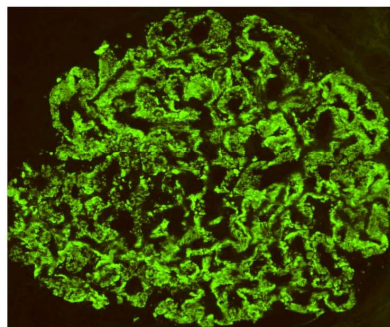
Type I MPGN (C3)



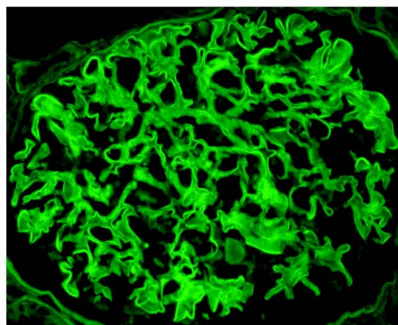
Type II MPGN (C3)



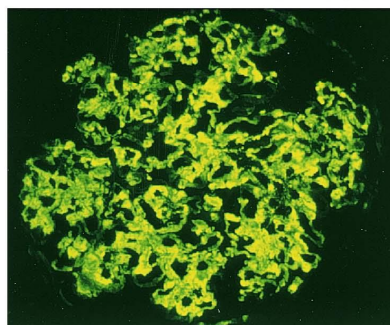
Post-infectious GN (C3)



Membranous Glomerulopathy (IgG)



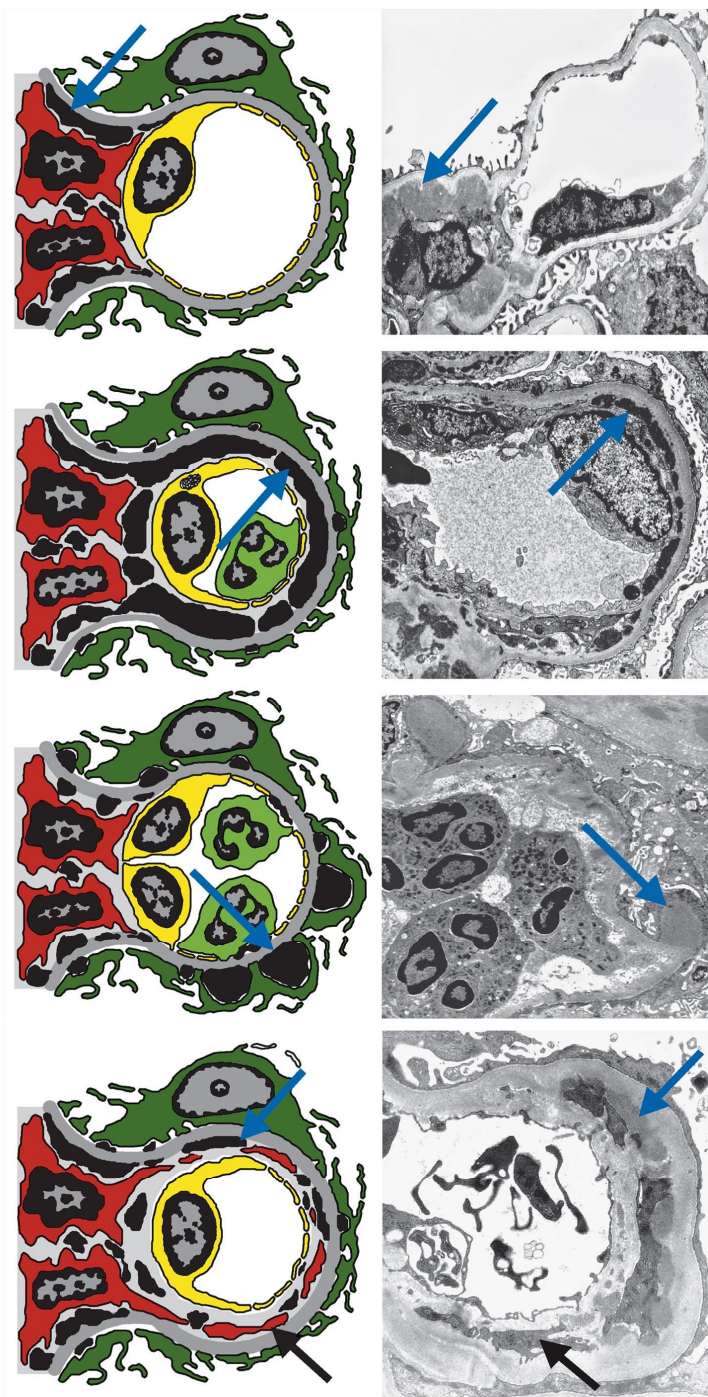
Anti-GBM GN(IgG)



Fibrillary Glomerulonephritis (IgG)

Step 4: Electron microscopic examination. Electron microscopy is not essential for the diagnosis of most forms of glomerular disease, but EM is helpful in many diseases for further confirmation and nuanced observations, EM is required for some diagnoses (e.g. fibrillary GN, immunotactoid glomerulopathy, thin basement membrane nephropathy), and EM is helpful in recognize unsuspected categories of disease that were not recognized by LM or IM (e.g. hereditary nephritis, collagenofibrotic glomerulopathy, Fabry disease).

Figure 3. Diagrams and electron micrographs of some of the glomerular capillary changes that must be discerned when evaluating glomerulonephritis by electron microscopy. A) mesangial electron dense deposits, B) subendothelial electron dense deposits, C) subepithelial hump-like electron dense deposits (postinfectious GN), D) subendothelial dense deposits with mesangial interposition (type I MPGN).



Electron microscopy occasionally reveals deposits in glomeruli that have distinctive organized substructure that point to specific diagnostic categories (Figure 4).

Figure 4. Algorithm for evaluating patterned (organized) deposits in glomeruli. Modified from D'Agati V, Jennette JC, Silva FG: Non-Neoplastic Renal Disease, American Registry of Pathology, Washington, D.C., 2005.

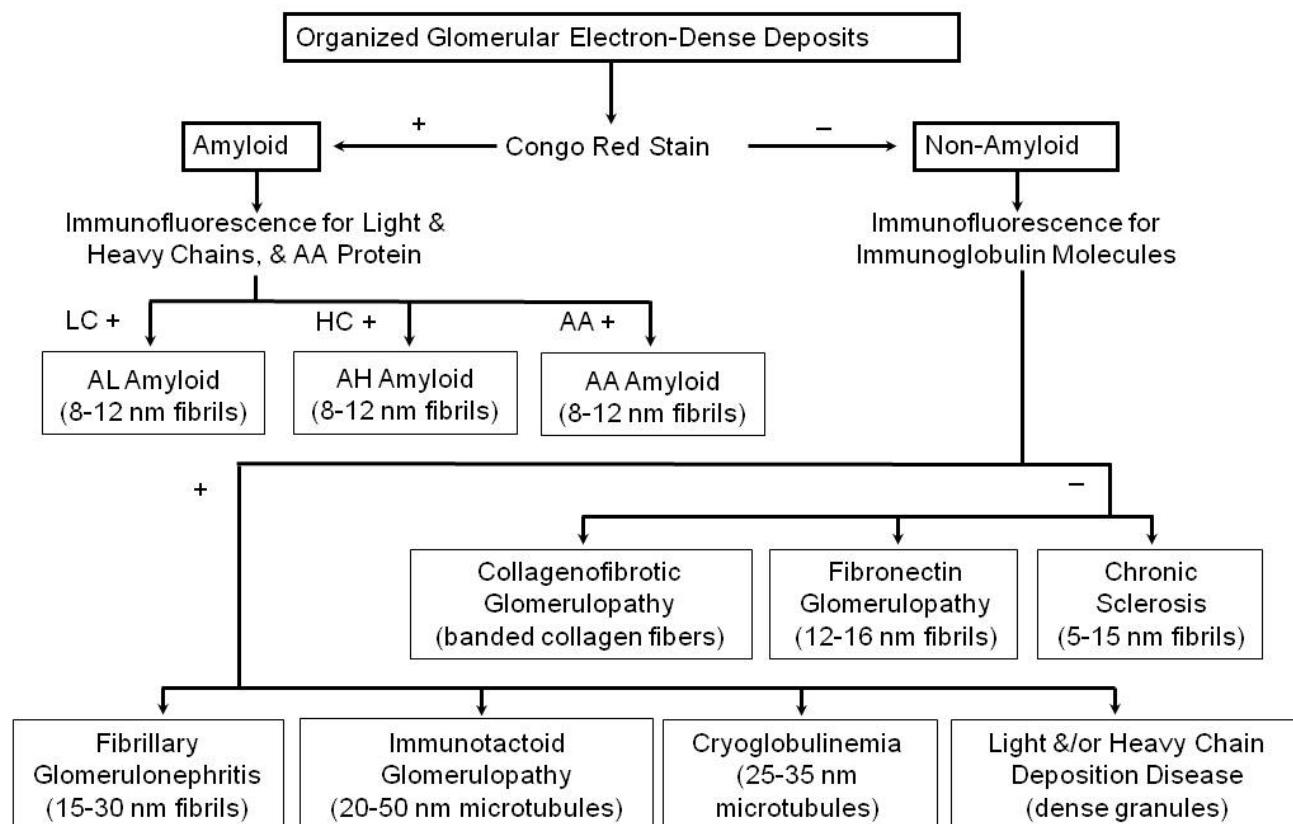
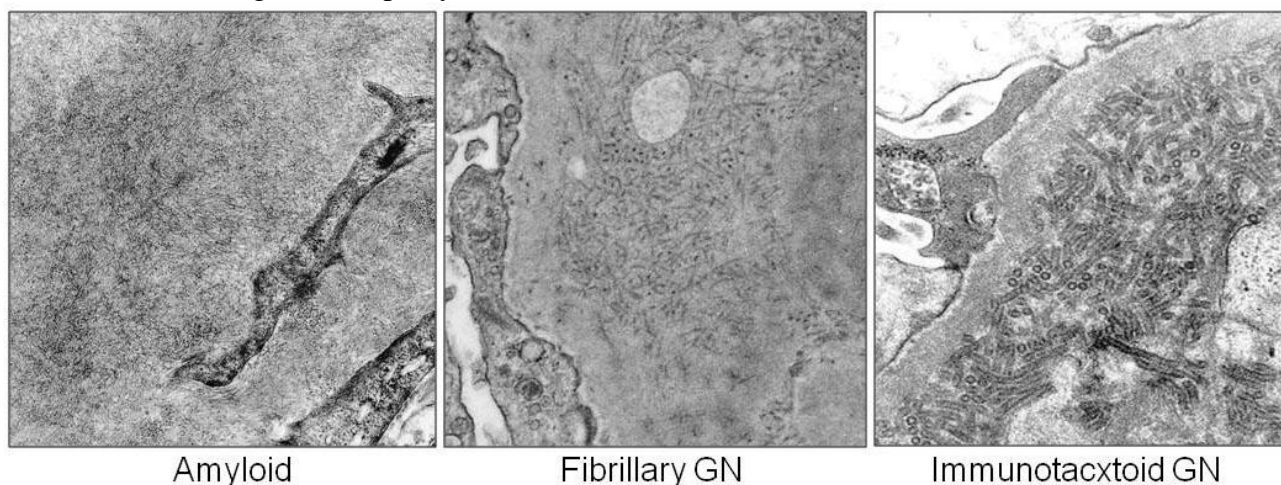


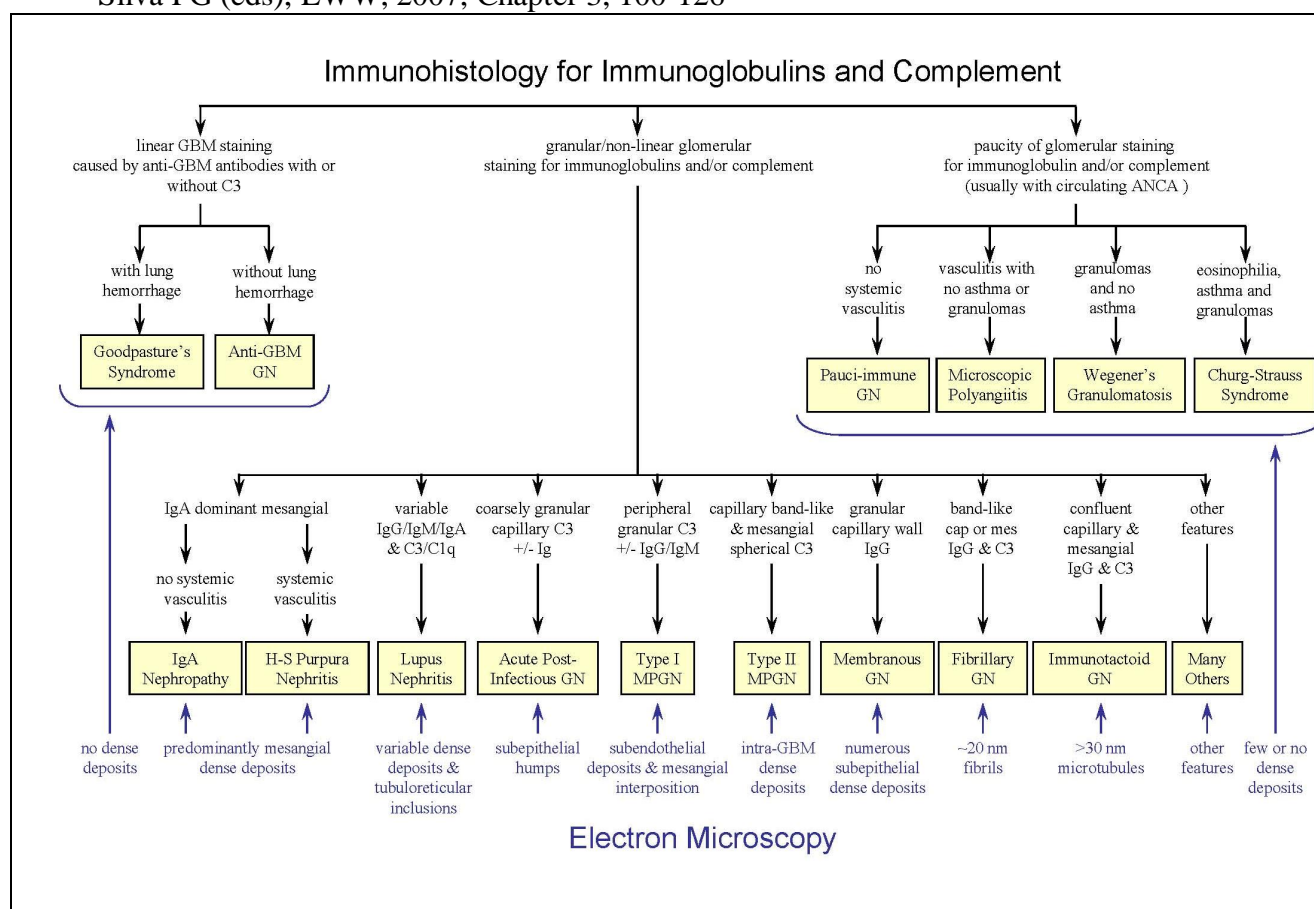
Figure 5. Electron micrographs showing the difference in appearance at the ultrastructural level at comparable magnifications (~20,000x) of amyloid fibrils (randomly arranged and only slightly wider than actin filaments), randomly arranged fibrillary GN fibrils (much larger than actin filaments), and more organized, much larger, partially parallel microtubules of immunotactoid glomerulopathy.



Step 5. Integration of all pathologic and clinical data into a final interpretation and diagnosis.

Once the light microscopy, immunohistology and electron microscopy data are collected, as well as relevant clinical data (e.g. age, sex, race, clinical features, serology, extrarenal disease distribution, etc.) an integrated interpretation must be formulated and a diagnosis rendered. For glomerular diseases in particular, this diagnosis often includes both terms that describe the histologic pattern of injury seen by light microscopy (e.g. focal proliferative and sclerosing glomerulonephritis with 10 % crescents) and terms that reflect findings by immunohistology (e.g. IgA nephropathy) or electron microscopy (e.g. fibrillary glomerulonephritis) (Figure 4). For example, the final diagnosis could be δ IgA nephropathy with focal proliferative and sclerosing glomerulonephritis with 10 % crescents δ or δ fibrillary glomerulonephritis with focal proliferative and sclerosing glomerulonephritis with 10 % crescents. δ The diagnosis could be further modified by knowledge of findings in the patient not observed in the renal biopsy specimen, e.g. δ Henoch-Schönlein purpura nephritis with focal proliferative and sclerosing glomerulonephritis with 10 % crescents. δ

Figure 4. Algorithm for integrating IM and EM findings when diagnosing some of the pathologic expressions of glomerular disease. Reproduced from Jennette JC, Olson JL, Schwartz MM, Silva FG: Primer on the Pathologic Diagnosis of Renal Disease in Heptinstall's Pathology of the Kidney, 6th Edition, Jennette JC, Olson JL, Schwartz MM, Silva FG (eds), LWW, 2007, Chapter 3, 100-126



Major references:

- 1) Jennette JC, Olson JL, Schwartz MM, Silva FG (eds): *Heptinstall's Pathology of the Kidney, Volumes 1 and 2, 6th Edition, Lippincott Williams & Wilkins, Philadelphia, 2007, Vol. 1 & 2*
- 2) D'Agati V, Jennette JC, Silva FG: *Non-Neoplastic Renal Disease, American Registry of Pathology, Washington, D.C., 2005*