Solutions to Test Your Knowledge: Hepatitis C and the Kidney

1. C
2. B
3. A
4. D
5. B

1. **C: Hepatitis C virus–associated cryoglobulinemic glomerulonephritis secondary to type II cryoglobulinemia**. The characteristic findings are prominent glomerular hypercellularity with glomerular intracapillary PAS-positive "hyaline thrombi", which represents precipitated cryoglobulin. Immunofluorescence typically shows IgG and IgM staining in the glomerular capillary hyaline thrombi and in smaller deposits away from the hyaline thrombi. Ultrastructural examination frequently reveals a microtubular substructure within the glomerular deposits. The diameter of the microtubules is usually between 20 and 30 nm, but can be larger. In this biopsy (panel C), the diameter of the microtubules was 26 nm. The glomerular intracapillary hypercellularity is primarily secondary to a large influx of monocytes. Large intracapillary hyaline thrombi can be seen in proliferative lupus nephritis; however, in proliferative lupus nephritis the deposits are localized primarily along the glomerular capillary loops ("wire-loop" lesions) and immunofluorescence shows a "full-house" pattern (positive staining for IgG, IgA, IgM, C1q, C3). Microtubular substructure within the deposits does not occur in lupus nephritis. In Waldenström macroglobulinemia, the findings can be quite similar to cryoglobulinemic glomerulonephritis; however, the intracapillary hyaline thrombi in Waldenström macroglobulinemia are monoclonal and contain IgM only. Substructure in the glomerular deposits is frequently absent in Waldenström macroglobulinemia, and the glomerular hypercellularity is usually less prominent than in cryoglobulinemic glomerulonephritis. The glomerular intracapillary hyaline thrombi superficially resemble fibrin thrombi; however, fibrin thrombi are less homogeneous and less PAS positive. Also, immunofluorescence and electron microscopy make the distinction between hyaline thrombi composed of cryoglobulins and fibrin thrombi easy.

2. **B: Proliferative glomerulonephritis with a membranoproliferative pattern secondary to hepatitis C virus infection.** In some cases of hepatitis C associated kidney diseases, the kidney biopsy shows the morphology of type I membranoproliferative glomerulonephritis with IgG and complement-containing immune complex deposits and absence of substructure in the glomerular deposits by electron microscopy. However, the findings are frequently atypical and the diagnosis can be difficult. In this biopsy with hypercellular glomeruli, only a few scattered glomerular deposits are noted, as shown on the immunofluorescence image. Ultrastructural examination confirms the paucity of deposits; only segmental scattered subendothelial and a few paramesangial deposits are seen without substructure. It is likely that glomerular cryoglobulin deposits (just like circulating cryoglobulins) quickly come and go; therefore, if the biopsy is not performed at the time when glomerular cryoglobulin deposits are present, only a few or no deposits are seen. Also, the degree of cryoglobulin deposition may vary between glomeruli; therefore, in a small biopsy specimen, the large glomerular capillary "hyaline thrombi" may be missed because of sampling error. In our experience, it is not unusual to see prominent intracapillary proliferative glomerulonephritis with very few deposits in patients with hepatitis C infection/type II cryoglobulinemia. In spite of the paucity of the immune complex deposits, the findings are not consistent with pauci-immune crescentic and necrotizing glomerulonephritis because the glomerular hypercellularity

in this biopsy is intracapillary. No extracapillary hypercellularity (crescents) or glomerular necrosis was seen in this biopsy. Crescents and glomerular necrosis are rare in hepatitis C-associated glomerulonephritis. Although interferon treatment can induce proteinuria, proliferative glomerulonephritis secondary to interferon treatment is rare and the interferon treatment in this patient was stopped two years before the biopsy. Therefore, the glomerular findings and the proteinuria are not related to interferon treatment in this patient. C3 glomerulopathy can have a membranoproliferative pattern of injury. In fact, many cases diagnosed as membranoproliferative glomerulonephritis in the past would be diagnosed as C3 glomerulopathy now. In C3 glomerulopathy, there is a disorder in the alternate pathway complement activation with abundant glomerular deposits, which are positive for C3 only. In this biopsy, very few deposits were seen, which stained for IgG, IgM, and C3. Also, the normal serum C3 levels are not consistent with C3 glomerulopathy.

3. **A: Proliferative glomerulonephritis, most likely secondary to infection, probably staphylococcus infection**. There is an emergence of cases of staphylococcus-associated proliferative glomerulonephritis, including hepatitis C virus positive patients (particularly if they are IV drug abusers or diabetic). This type of glomerulonephritis is typically associated with glomerular hypercellularity and IgA and C3-containing glomerular deposits. The presentation is usually acute, unlike idiopathic IgA nephropathy. This patient turned out to have *Staphylococcus aureus* pneumonia after steroid treatment. Immunosuppression, diabetes mellitus, and morbid obesity are risk factors for the development of such glomerulonephritis. Although staphylococcus infection-associated glomerulonephritis is not directly related to hepatitis C infection, it should be considered in the differential diagnosis, particularly if immunofluorescence shows IgA deposits in the mesangium. The morphology is not consistent with cryoglobulinemic glomerulonephritis secondary to type II cryoglobulinemia because of the absence of relevant glomerular IgG and IgM staining. Electron microscopy did not show any substructure in the deposits, and several deposits were subepithelial (humps) (see panel C). Chronic liver disease with liver cirrhosis can result in mild mesangial IgA deposition, but such cirrhosis-related mesangial IgA deposits are usually not associated with acute clinical symptoms or relevant hematuria or proteinuria. Also, C3 staining is unusual in cirrhosis-associated mesangial IgA deposits. In acute post-streptococcal glomerulonephritis, IgA deposits do not occur in the glomeruli; the glomerular deposits either stain for C3 only or for C3 and IgG. Another important difference is that in post-streptococcal glomerulonephritis the infection resolves by the time the acute glomerulonephritis develops, whereas in many cases of staphylococcus-associated proliferative glomerulonephritis, the infectious process is still ongoing at the time the acute kidney injury secondary to the glomerulonephritis manifests.

4. **D: Membranous glomerulonephritis, probably secondary to chronic hepatitis C virus infection**. Light microscopy revealed normocellular glomeruli with diffusely thickened glomerular capillary loops and complex spikes on methenamine silver stain (not shown). The granular IgG staining by immunofluorescence and the abundant subepithelial deposits seen by electron microscopy are diagnostic of membranous glomerulonephritis. Membranous glomerulonephritis can be associated with both hepatitis B and C virus infections. By morphologic criteria, it is difficult to tell if a membranous glomerulonephritis is hepatitis C–associated or primary; however, in our experience, in hepatitis C virus-associated membranous glomerulonephritis, the glomerular immune complex deposits are usually IgG1 predominant. In contrast, most cases of primary membranous glomerulonephritis exhibit glomerular deposits

comprised of IgG4 dominant. Considering this patient's clinical history, the high hepatitis C viral load, and the IgG1 dominance in the glomerular capillary deposits, it is likely that this membranous glomerulonephritis is secondary to hepatitis C virus infection. In HIV-associated nephropathy, the heavy nephrotic syndrome is related to glomerular capillary collapse with prominent glomerular epithelial cells and acute tubulointerstitial injury. In HIV-associated collapsing glomerulopathy, usually no immune complex deposits are present; however, if there is co-infection with hepatitis C infection, some deposits may occur. In cryoglobulinemic glomerulonephritis (see Question 1), the deposits are localized within the glomerular capillary loops, the mesangium, and the subendothelial space; subepithelial deposits are not characteristic of cryoglobulinemic glomerulonephritis.

5. **B: Fibrillary glomerulonephritis**. The glomeruli were enlarged with moderate mesangial expansion and segmental capillary loop thickening (panel A). Methenamine silver stain was relatively weak in the expanded mesangium (not shown). Immunofluorescence showed diffuse smudgy mesangial and glomerular capillary IgG deposits, which were predominantly positive for IgG4 (panel B) and less intensely for IgG1. Electron microscopy showed randomly arranged fibrillary material deposited in the mesangium (panel C) and along the glomerular capillary loops. The diameter of the fibrils was 16 nm. There is some association between fibrillary glomerulonephritis and hepatitis C infection. In contrast, there appears to be no association between the absence or presence of cryoglobulinemia and fibrillary glomerulonephritis. The association between hepatitis C viral load and fibrillary glomerulonephritis is also unclear. In fibrillary glomerulonephritis, the fibrillary deposits are present all over in the glomerular extracellular matrix, including the mesangial matrix and the glomerular basement membrane. The fibril diameter in fibrillary glomerulonephritis is most frequently between 12 and 20 nm. In cryoglobulinemic glomerulonephritis, the microtubules are thicker than 20 nm, localized to intracapillary hyaline thrombi or to well-circumscribed subendothelial or mesangial deposits, and not seen diffusely throughout the glomerular extracellular matrix. The immunofluorescence profile is also different; in fibrillary glomerulonephritis the deposits are IgG4 dominant with lesser amounts of IgG1 and are present throughout the glomerulus. In cryoglobulinemic glomerulonephritis, the deposits contain IgM and IgG with dominance of IgG1, some IgG2 and IgG3. IgG4 containing deposits are usually absent in cryoglobulinemic glomerulonephritis. Amyloidosis may resemble fibrillary glomerulonephritis; however, in fibrillary glomerulonephritis Congo Red stain is negative (such as it was in this biopsy), and the fibrils are thinner (usually 8-10 nm) than in fibrillary glomerulonephritis (usually between 12-20 nm). The most frequent type of amyloidosis in the United States is AL (light chain) amyloidosis, but rare cases of AH (heavy chain) amyloidosis may occur. In AL or AH amyloidosis, the amyloid fibrils are composed of monoclonal immunoglobulins; in fibrillary glomerulonephritis, the fibrils are composed of polyclonal IgG.