Fibrillary Renal Deposits and Nephritis

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Fibrillary renal deposits and nephritis. The authors have studied 8 patients whose glomeruli contain abundant fibrils in their mesangial matrix and basement membranes. Although the location of these fibrils is very similar to that of amyloid, they are about twice the size of amyloid fibrils, averaging 20 nm in width, and fail to react as amyloid does with special stains. Immunofluorescence-microscopic studies are usually positive with antiserums to IgG, often IgM, and in some cases IgA, and also kappa and lambda light chains, C3, and C4. The fibrils are associated with diffuse mesangial widening and increased mesangial matrix strands. Although peripheral glomerular capillary walls appear to be spared initially, their eventual involvement leads to glomerular capillary collapse and glomerular obsolescence. Crescent formation occurred in 5 cases, focally in 3 and diffusely in 2. Tubular basement membrane involvement was seen in 1 case. These patients exhibit hematuria, and proteinuria, and often hypertension and renal insufficiency. Proteinuria was in the nephrotic range in 3 patients in whom involvement of glomerular capillary basement membranes was extensive. Unless electron microscopy is applied to renal biopsies, these cases may be considered to represent mesangiocapillary or rapidly progressive glomerulonephritis, or amyloidosis. The nature of these fibrils is as yet not determined. It is likely that they have been called "atypical amyloidosis" in the past. (Am J Pathol 1983, 113:279–290)

THE APPLICATION of electron microscopy to the study of normal and pathologic anatomy has led to the description of various extracellular microfibrils. A recent study of rapidly proliferating connective tissue in the chick embryo[1] has delineated three natural types: small, about 10 nm in diameter, such as may be seen in elastic as well as other tissues; large, 18–25 nm in diameter, which are believed to include collagen precursors; and unit collagenous fibrils 30 nm in diameter and greater. Also noted were thin filaments measuring 3–5 nm in diameter, which may be related to basement membranes.

Rodent glomerular basement membranes have been found to be composed of a meshwork of thin filaments (3–4 nm) embedded in a finely granular matrix.[2–4] Small tubular microfibrils (approximately 11 nm in diameter) have also been described in association with rodent basement membranes.[3] Similar fibrils have been found in the mesangial matrix.[5,6] A study of human fetal kidneys[7] reported 4-nm fibrils in the lamina densa of the glomerular basement membranes and larger 10 nm fibrils resembling those previously described in rodents. We have been unable to find a specific study of various fibrils in normal adult human kidneys, although such fibrils are mentioned without details.[8]

A recent study in which 72 human renal biopsies were evaluated for the presence of small microfibrils in various renal diseases (focal segmental glomerulosclerosis, transplant glomerulopathy, malignant hypertension, hemolytic uremic syndrome)[8] found approximately 12-nm (11–13 nm) fibrils in most cases studied, mainly in a subendothelial location. Most of the disease entities are rarely associated with complement or immunoglobulin deposition. In another

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Table 1—Summary of Clinical Features

	Patient								
	1	2	3	4	5	6	7	8_1	8_2
Age/Sex	36/M	49/M	52/F	39/M	42/F	26/M	64/F	47/M	50/M
Blood pressure	190/80	160/105	170/80	110/80	160/110	130/80	150/90	160/106	260/90
Hematuria	Gross	Micro	Micro	Micro	Micro	Gross	Micro	Micro	Micro
Urine protein (g/24 hrs)	0.8	1.8	1.9	2.1	3.3	4.7	5.2	1.1	8.5
Serum Urea Nitrogen (mg/dl)	30	46	110	21	2	18	20	25	110
Serum creatinine (mg/dl)	2.5	2.1	9.6	1.9	N	1.2	1.1	1.2	11.0
Creatinine clearance (ml/min)	66	64	8	77	NA	90	54	94	NA
Serum protein electrophoresis	N	*	N	N	*	N	N	N	N
Bence Jones protein	N	N	N	N	N	NA	N	N	N
Antinuclear antibody	N	1:20	N	N	1:320	N	N	1:10	N
Antistreptolysin 0 titer	N*	NA	*	N	NA	N	NA	N	NA
Serum cryoglobulins	NA	N	N	NA	NA	N	N	NA	NA
Reason for biopsy	H, P	H, P, BP	ARF	NS	H, P, BP	NS	P	H, P, BP	ARF

Patient 1 had a history of rheumatic fever 5 years before with a blood pressure of 160/100 and an ASLO titer of 1:500; the heart was not involved. He was diabetic, and his disease was controlled by diet.

Patient 2 had polyclonal gammopathy determined by serum electrophoresis.

Patient 3 had had hypertension for 15 years. Streptozyme test results, 200 units.

Patient 4 had had protein and blood found in the urine 10 years before. His father and paternal grandfather were diabetic. He had minimal high-frequency hearing loss and a history of muscle cramps.

Patient 5 had anaplastic carcinoma of the lung and polyarthritis 10 years before. Polyclonal gammopathy was found by serum electrophoresis; systemic lupus erythematosus was suspected.

Patient 6 had gross hematuria and proteinuria 10 years before.

Patient 7 had cellulitis of the leg.

Patient 8 had a history of "peptic ulcer."

* See comments; N, normal or negative; NA, not available; H, hematuria; P, proteinuria; BP, hypertension; NS, nephrotic syndrome; ARF, acute renal failure.

recent study the presence of large microfibrils was evaluated in 360 renal biopsies in children, involving a variety of diseases.[9] Seventeen- to 35-nm fibrils were found in 47% of the cases studied. They were not associated with any particular clinical or morphologic findings, nor did they appear to be related to immunofluorescent deposits of immunoglobulin or complement.

In both of these studies it appears that the authors dealt with normally occurring extracellular fibrils or modifications thereof.

A second group of fibrils that may be found in glomeruli appears to originate from proteins circulating in the blood. Among these fibrils are amyloid and its precursors, cryoglobulins, and similar substances.

Thus, there have been numerous reports of pathologic renal involvement by extracellular fibrils in the 10–12-nm range. Many of these represent renal amyloidosis. However, somewhat similar fibrils have been reported in other conditions. Fibrils measuring 11–14 nm have been described in mesangial monoclonal kappa light chain deposits.[10,11] Twenty-seven-nanometer tubular fibrils have been described in a patient with kappa glomerulopathy and negative serum cryoglobulin determinations.[12] Studies of renal disease associated with large extracellular fibrils have otherwise generally been limited to cases of cryoglobulinemia in which tubular fibrils measuring from about 20–30 nm and larger have been described.[13–16]

We have encountered 8 cases of renal fibrillosis which give negative results with Congo red and thioflavin-T stains and which by electron microscopy show numerous glomerular fibrils which, although they resemble amyloid fibrils in their location and distribution, are wider than amyloid but not as wide as cryoglobulins, and have a distinctive structure. These fibrils are generally loosely arranged, not infrequently curved or bent, and measure 20 nm in width on average, and up to 1500 nm in length in cut sections. These patients do not suffer from any condition usually associated with amyloidosis, but often exhibit hematuria, proteinuria, hypertension, and renal insufficiency.

Materials and Methods

Patients were chosen for this retrospective study because electron-microscopic examination of their renal biopsies demonstrated numerous 20-nm-wide extracellular glomerular fibrils. Four of these cases were from one series of 536 renal biopsies in which glomeruli were available for study. All of the other 532 biopsies were carefully examined by electron microscopy for the presence of these fibrils, and none

Table 2—Immunofluorescence Microscopic Findings*

	Patient							
	1	2	3	4	5	6	8_1	8_2
IgG	3+ Mes 3+ GCW	3+ Mes	4+ Mes 4+ GCW 2+ TbW 2+ Art	4+ Mes 4+ GCW 3+ TbW	3+ Mes 3+ GCW	4+ Mes 4+ GCW	2+ Mes 2+ GCW	3+ Mes 3+ GCW
IgA	NA	Neg	2+ Mes 2+ GCW	Neg	Tr Mes Tr GCW	1+ Mes 1+ GCW	Neg	Neg
IgM	2+ Mes 2+ GCW	2+ Mes	Neg	Neg	2+ Mes 2+ GCW	2+ Mes 2+ GCW	1+ GCW	Neg
Kappa	NA	4+ Mes	3+ Mes 3+ GCW	3+ Mes 3+ GCW 3+ TbW	2+ Mes 2+ GCW	NA	NA	3+ Mes 3+ GCW
Lambda	NA	4+ Mes	3+ Mes 3+ GCW	2+ Mes 2+ GCW	2+ Mes 2+ GCW	NA	NA	2+ Mes 2+ GCW
C3	3+ Mes 2+ GCW	3+ Mes	4+ Mes 4+ GCW 3+ TbW 3+ Art	2+ mes 2+ GCW	3+ Mes 3+ GCW 2+ TbW 2+ Art	2+ Mes 2+ GCW	1+ GCW	3+ Mes 3+ GCW
C4	NA	NA	NA	3+ Mes 3+ GCW	NA	2+ Mes 2+ GCW	NA	3+ Mes 3+ GCW
Clq	NA	2+ Mes	NA	1+ Mes 1+ GCW	NA	NA	NA	3+ Mes 3+ GCW
Source of reagents	1	2	3	4	3	1	5	4

* Staining of mesangia and glomerular capillary walls is diffuse and granular in all cases except Cases 2 and 8, where it is segmental and granular. Staining of tubular walls and small vessels is focal and granular.

Art, arterioles and small arteries; GCW, glomerular capillary walls; Mes, mesangium; Neg, negative; NA, not available; TbW, tubular walls.

1, Hyland Laboratories, Costa Mesa, Calif; 2, Cal Biochem-Behring, LaJolla, Calif; 3, anti-kappa and -lambda, Cappel Laboratories, Cochranville, Pa, all other reagents from Burrows Wellcome, Research Triangle Park, Va; 4, Meloy Laboratories, Springfield, Va; 5, Behring Diagnostics, Summerville, NJ.

were found. The other 4 cases were from three different hospitals whose material has not been extensively searched for other cases. Patient historic information, including the reason for biopsy, is given in Table 1. It should be noted that the patients exhibited a nephritic or mixed picture rather than a purely nephrotic picture and did not suffer from conditions usually associated with amyloidosis. There is no evidence of involvement of other organs or tissues. However, studies have of necessity been limited to renal tissue obtained by percutaneous biopsy.

Tissues for light-microscopic study were stained with hematoxylin and eosin, periodic acid–Schiff (PAS), periodic acid–silver methanamine (PASM) (all examined with visible light), Congo red (examined with polarized light), and thioflavin-T (examined with ultraviolet light). Thioflavin-T studies are not available for Case 1; otherwise, all biopsies were studied with all stains.

For immunofluorescence-microscopic study, quick-frozen specimens, sectioned by cryostat, were stained with fluoresceinated antiserums obtained from established commercial laboratories (see Table 2) and examined under ultraviolet light.

For electron-microscopic study, material was either fixed primarily in 1.0% osmium tetroxide or fixed primarily in aldehydes such as glutaraldehyde, a mixture of formaldehyde and glutaraldehyde, or paraformaldehyde, and postfixed in 1.0% osmium tetroxide. For the first biopsy of Case 8, a small block of formalin-fixed paraffin-embedded tissue was deparaffinized and then postfixed in 1.0% osmium tetroxide. All of these specimens were then dehydrated in graded alcohols and embedded in epoxy resin.

For the purposes of this study, the epoxy-embedded tissue from all cases was brought to one laboratory for examination. Thus, the processing—sectioning, staining, electronmicrography, developing, enlarging, printing—for all cases was done under identical conditions. The only variables, therefore, are the biopsy procedure, fixation, and embedding. (The effect of these variables is probably small: the two biopsies from Case 8 showed essentially the same ultrastructural findings despite quite different processing.) Thin sections were stained with uranyl acetate and lead citrate, phosphotungstic acid (PTA), and PASM and studied with a Hitachi H-12 electron microscope operating at 75 kv. For high resolution studies and measurements, very thin sections (much less than 60 nm in thickness) were stained with uranyl acetate and lead hydroxide and examined in the same microscope, operating at 100 and 125 kv. The microscope was cali-

brated using a 28,800 line/inch grating replica obtained from Ernest F. Fullam, Inc., Schenectady, New York, according to the manufacturer's instructions. The micrographs used for high-resolution studies and measurements were enlarged 2.5 times from the original negatives taken at magnifications of ×48,000 and examined with a ×7 magnifying glass with a built-in millimeter scale. Only distinct symmetric cross-sections were measured.

Results

Light Microscopy

Light-microscopic findings (Figures 1–5) are summarized in Table 3. The most striking finding, and the major finding in mild cases, is diffuse mesangial matrix increase and mesangial widening. This is in distinct contrast to the rather amorphous nodular enlargement of mesangial areas usually seen in amyloidosis. This matrix increase does not appear to encroach appreciably upon mesangial cells. Rather, they appear to be increased in number in some cases, and a complex mixture of mesangial matrix and cytoplasm results. In contrast to amyloidosis, the involved mesangial matrix stains rather strongly with PAS or PASM stains, resulting in a rather distinctive mottled appearance at high magnifications (Figures 2 and 4). This mesangial widening appears to be proportional to the severity of the disease process. Involvement of glomerular capillary basement membranes occurs to a variable degree in each case. It is rather inconspicuous in mild cases (Figure 1) and may not be readily apparent by light microscopy. However, practically every peripheral capillary wall is involved in more severe cases (Figure 3). The extent of this involvement appears to be proportional to the degree of proteinuria. The resultant basement membrane thickening is also variable and frequently very irregular. Rarely, spikelike projections reminiscent of membranous nephropathy[17] are seen. Periglomerular fibrosis is seen in most cases and appears to occur early. Focal crescent formation is seen in 5 of 8 cases and is a prominent finding in 2 cases (Figure 5). Glomerular capillary collapse is noticeable in all cases, even those with apparently mild or early involvement. This appears to lead to eventual total glomerular collapse and sclerosis with resultant focal tubular atrophy, interstitial fibrosis, and round cell infiltration. Widespread tubular and interstitial changes do not occur until late in the course of the disease (Figure 5). However, the extent of this finding, as well as the degree of glomerular sclerosis, appears to correlate roughly with the severity of hypertension and renal insufficiency.

Special Light Microscopy

All cases examined failed to stain with thioflavin-T or to exhibit green birefringence in Congo red stained sections with polarized light.

Immunofluorescence Microscopy

Immunofluorescence findings are summarized in Table 2. The usual finding is diffuse coarsely granular staining of mesangia and glomerular capillary walls (Figure 6) with antiserum to IgG, kappa and lambda light chains, C3, and C4. Five cases stained with anti-IgM and 3 with anti-IgA. Some cases also show focal

Table 3—Light-Microscopic Findings

	Patient								
	1	2	3	4	5	6	7	8_1	8_2
Mesangial matrix increase	+ +	+ +	+ +	+ +	+ +	+ + +	+ +	+ +	+ + + +
Mesangial widening	+ +	+	+ +	+ +	+ +	+ + +	+ +	+	+ + + +
Mesangial PAS staining	+ +	+ +	+ +	+ + +	+ +	+ +	+ +	+ +	+ +
Mesangial PASM staining	+ +	+ +	+ +	+ +	+ + +	+ + +	+ +	+ +	+ +
Glomerular capillary wall involvement	+ +	+	+ + +	+ +	+ +	+ + + +	+ + +	+	+ + + +
Crescent formation*	0/7	2/8	13/16	0/10	0/5	1/8	4/11	2/23	3/6
Periglomerular fibrosis*	2/7	2/8	8/16	0/10	3/5	2/8	2/11	2/23	6/6
Glomerular sclerosis*	1/7	3/8	4/16	0/10	2/5	3/8	0/11	6/23	3/6
Tubular atrophy	+ + F	+ + F	+ + +	N	+ + F	+ + F	+ F	+ F	+ + + +
Interstitial change	+ + F	+ + + + F	+ + + +	N	+ + + F	+ + F	+ F	+ F	+ + + +
Overall tubulointerstitial change	+	+	+ + +	N	+	+	+	+	+ + + +

* Glomerular findings: glomeruli showing finding/total glomeruli observed.
F, focal involvement; N, negative.

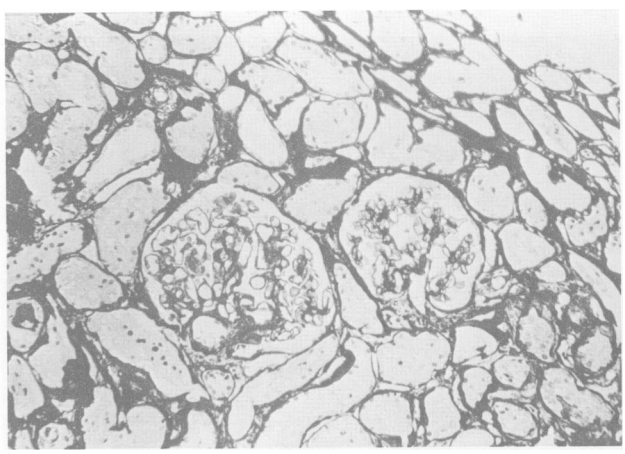
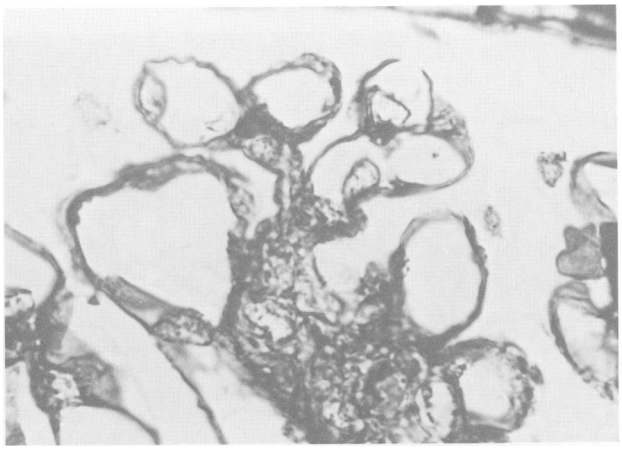
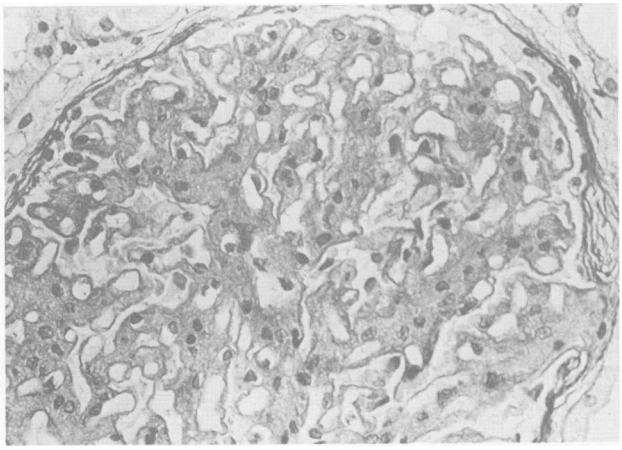
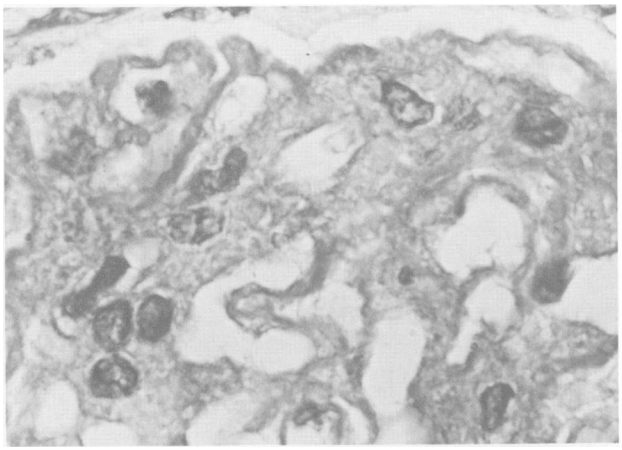
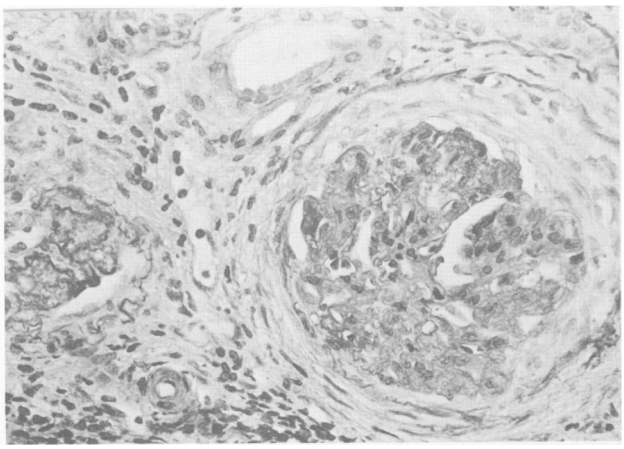
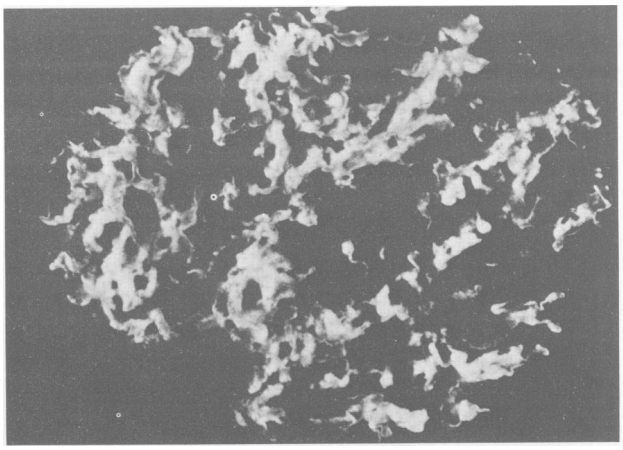

Figure 1 — Two glomeruli that appear relatively normal at low magnification. First biopsy of Case 8. (PASM, × 100) **Figure 2** — Higher magnification of the larger glomerulus in Figure 1, showing distinctive mesangial change as well as irregular thickening of some capillary walls. (PASM, × 1000) **Figure 3** — Advanced glomerular changes consisting of diffuse mesangial widening and capillary wall thickening. Case 6. (PAS, × 250) **Figure 4** — Detail from Figure 3, showing mottled mesangium and irregular thickening of practically all capillary walls. (PAS, × 1000) **Figure 5** — Advanced stage, showing crescent formation and severe tubulointerstitial change. Second biopsy of Case 8. (PAS, × 250) **Figure 6** — Example of immunofluorescence results with staining of both mesangium and glomerular capillary walls. Case 6. (Anti-IgG, × 250)

staining in scattered tubules and small arteries. Case 4 and the second biopsy specimen in Case 8 are also examined for the presence of IgD and IgE, Case 6 for IgD, all with negative results. No immunofluorescence studies are available for Case 7.

Electron Microscopy

Electron-microscopic findings are summarized in Table 4. Low magnification studies reveal numerous, frequently rather loosely packed, straight, curved,

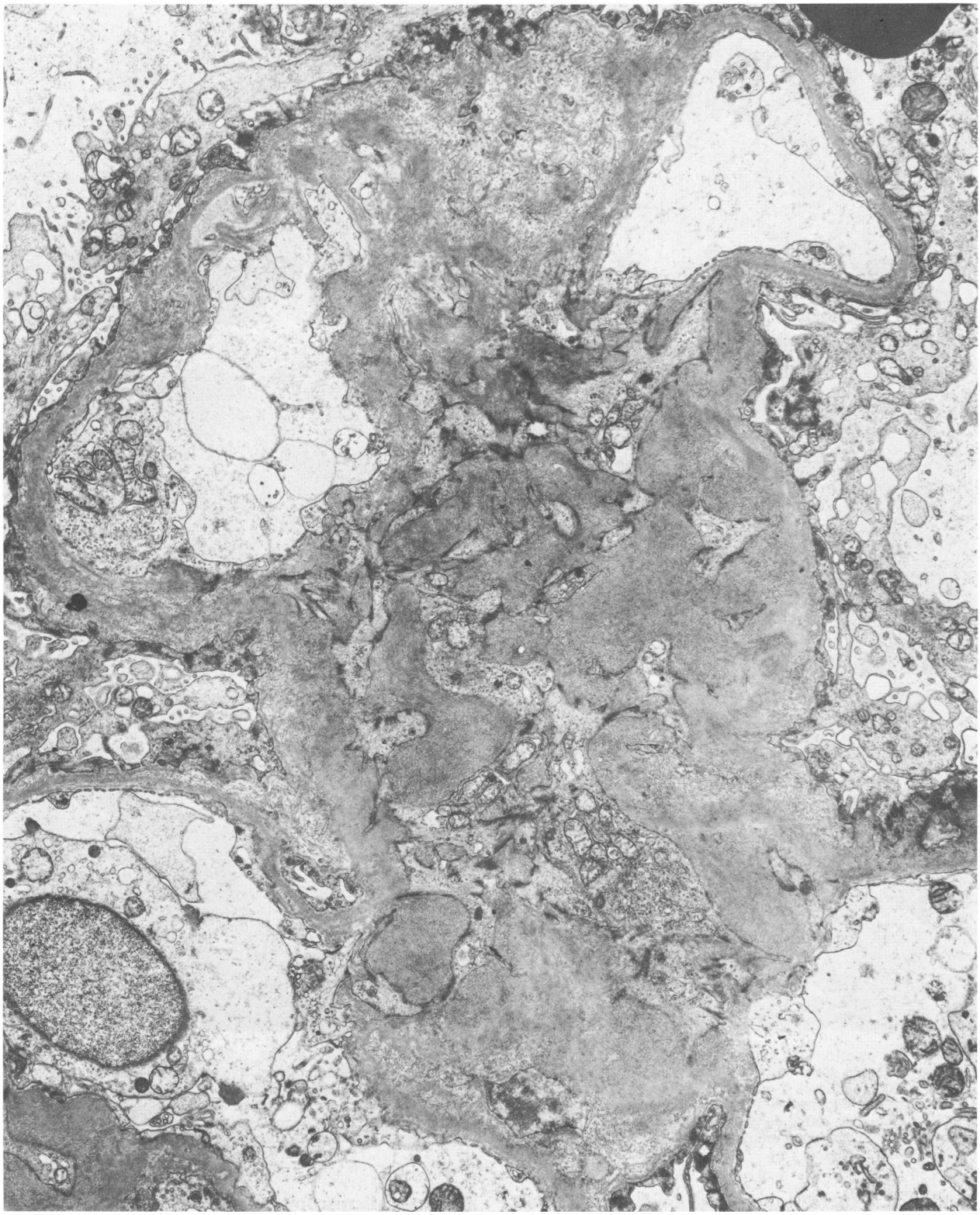

Figure 7 — Portion of glomerulus with fibrils in mesangial matrix and glomerular capillary basement membrane. The mesangial area consists of a complex mixture of mesangial cell cytoplasm and matrix strands containing the fibrils. The capillary basement membrane in the upper left is greatly disorganized and appears prone to rupture. Case 4. (Uranyl and lead, ×7500; primary fixation, 1.0% OsO4). (With a photographic reduction of 4%)

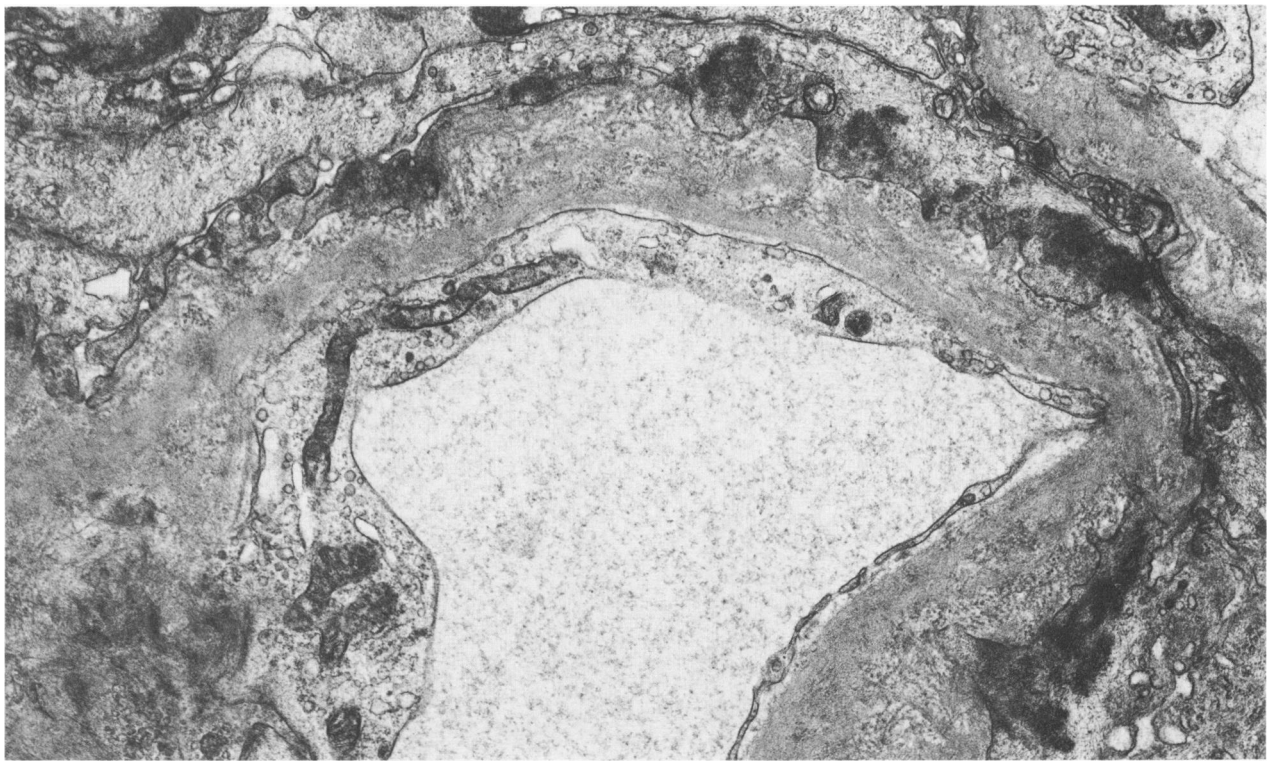

Figure 8—Glomerular capillary wall with fibrillar deposits located primarily in the outer aspect. The resulting picture resembles membranous nephropathy. Case 3. (Uranyl and lead, ×15,000; primary fixation, 1.0% OsO4)

and bent fibrils primarily in mesangial matrix but also to a variable degree in glomerular capillary basement membranes (figures 7 and 8) and in one case (Case 4) in tubular basement membranes. Because of their size and abundance, these fibrils are quite noticeable at final magnifications as low as ×3000. These accumulations appear to be haphazardly arranged and are generally more concentrated in mesangial areas. They are of moderate electron density, comparable to that of basement membrane or mesangial matrix. Two cases (Case 1 and 2) also show very small (trace and 2+, respectively) paramesangial aggregates of finely granular electron-dense deposits. At magnifications up to ×52,500 (Figure 9) the fibrils appear to be rather uniform and to measure 20 nm in width, the longest observed was 1500 nm in length. However, these studies are of sections rather than of isolated fibrils, so that most fibrils are considerably shorter than that and greatest length is uncertain. Affected mesangial areas show a complex increase in mesangial matrix strands and mesangial cell cytoplasm (Figure 7) rather than the massive homogeneous nodular mesangial expansion usually seen with amyloidosis. Glomerular capillary basement membrane

Table 4—Electron-Microscopic Findings

	Patient								
	1	2	3	4	5	6	7	8_1	8_2
Foot process obliteration	+	Tr	+ + + +	+ + + +	+ + + +	+ + + +	+ + +	NA	+ + + +
Capillary wall involvement	+ ±	Tr	+ + +	+ + +	+ +	+ + + +	+ + +	Tr	+ + + +
Average basement membrane thickening (nm)	1000–2100	N	700–2200	500–1400	900–1600	1200–3000	900–1200	Rare focus up to 700	1000–4000
Mesangial involvement	+ + + +	+ +	+ + +	+ +	+ + +	+ + + +	+ + +	+	+ + + +
Paramesangial dense deposits	Tr	+ +	N	N	N	N	N	N	N

N, normal or negative; NA, not available; Tr, trace.

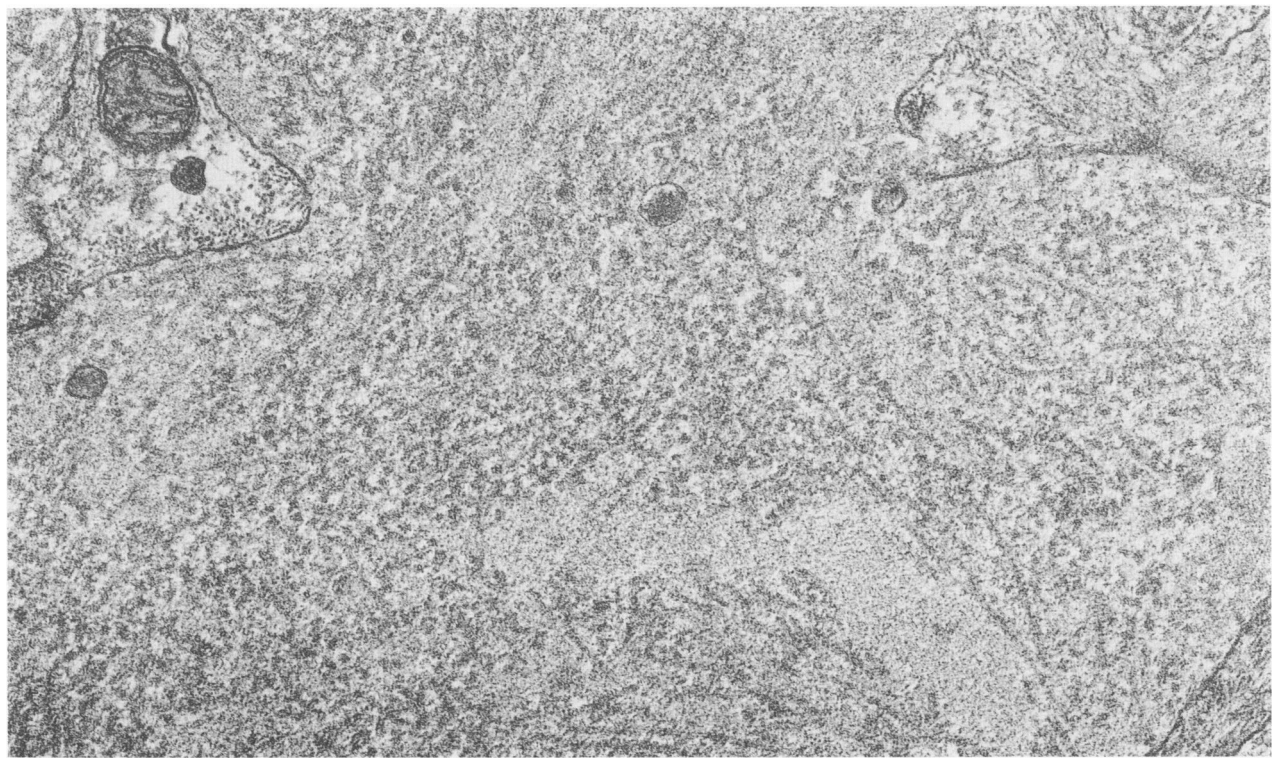

Figure 9—Higher magnification of fibrils cut at various angles. When seen in cross-section they appear to consist of indistinct tubular structures surrounded by an electron-lucent matrix. Case 2. (Uranyl and lead, ×52,500, primary fixation, 2.5% glutaraldehyde)

deposits are generally transmembranous, with a considerable variation in concentration from a sparse speckling of deposits to uniform dense involvement throughout. In some areas, deposits are seen to involve primarily the inner or, more commonly, the outer aspect of the basement membrane. The latter occurrence may lead to spikelike projections containing these fibrils in a pattern resembling membranous nephropathy[17] (Figure 8). Although it is by no means certain, glomerular capillary basement membrane involvement appears to occur after mesangial involvement. In other words, it appears as though mesangial deposits occur before contiguous peripheral capillary walls are involved. Although there is generally marked effacement of epithelial foot processes, the basement membrane deposits do not appear to disrupt significantly either epithelial or endothelial cells. Mild or early basement membrane involvement results in an irregular thickening, which may be barely noticeable or absent by light microscopy. Eventually, however, very obvious irregular thickening and apparent reduplication is seen. This appears in some instances to greatly weaken the capillary wall and lead to its disruption and collapse (Figure 7).

Detailed examination of the fibrils (Figures 10, B and 11) shows that they vary from 16 to 24 nm in diameter, 92% measuring between 18 and 22 nm, and a very sharp peak occurring at approximately 20 nm. They have a distinctive ultrastructure, tubular but with a double contour. Measurements of cross-sections reveal an inside diameter of about 8 nm, an outside diameter of about 20 nm, and a double wall consisting of inner and outer layers, each about 1.6 nm in thickness and separated by a lucent area of about 2.8 nm. In some sections there appears to be a small gap in the wall. A regular periodicity was not found.

In all cases, the 20-nm fibrils failed to stain with PTA but occasionally stained faintly with PASM. This staining is weaker than that of cryoglobulins and much weaker than that of collagen.

In addition to the 20-nm fibrils, at least four other populations of filaments and fibrils were found in many locations.

1) Numerous filaments in the 3–5-nm range are noted scattered throughout. These are considered to be analogous to filaments described by Yamada,[2] Farquhar et al,[3] and Farquhar[4] and are not evaluated further in this report.

2) Rather numerous microtubular fibrils are noted measuring between 11 and 13 nm. These appear to be identical to the fibrils studied by Hsu and Churg,[8] and also mentioned by previous authors, and are not studied further.

3) We also encounter a smaller population of

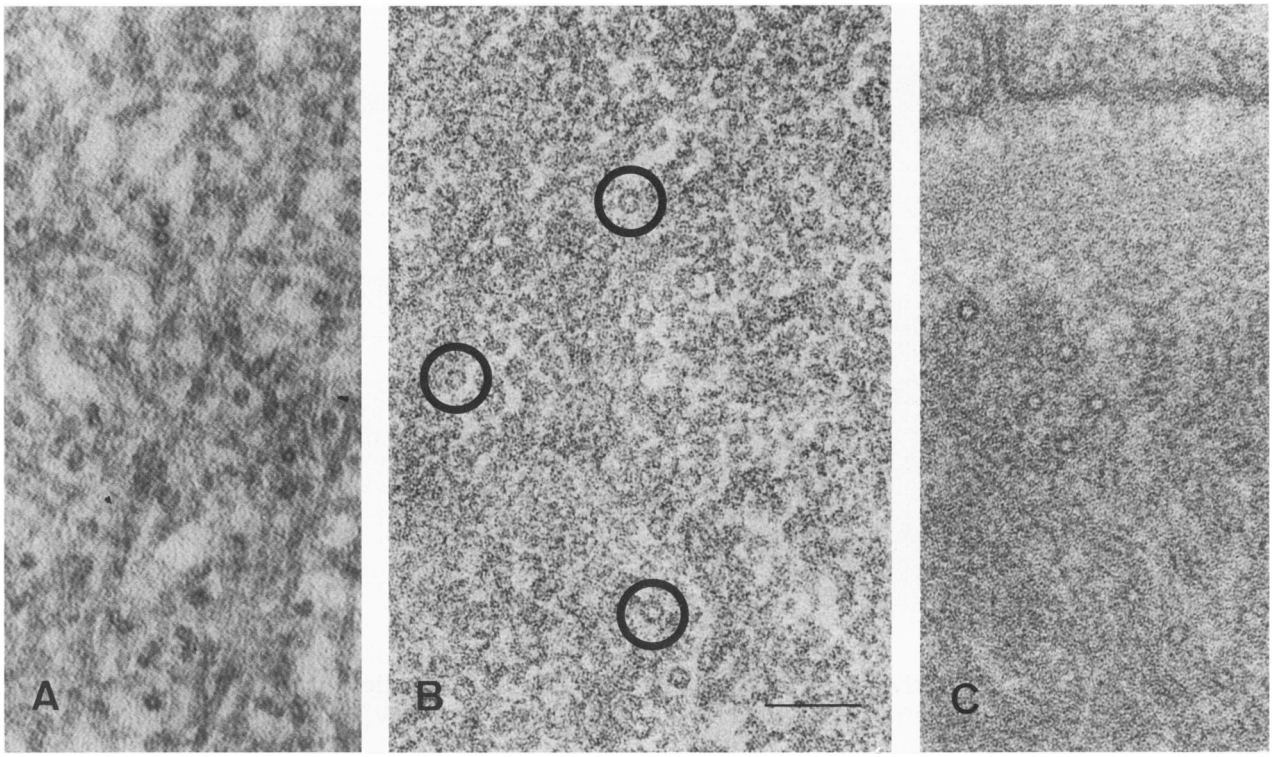

Figure 10—Comparison of amyloid, 20-nm fibrils being reported, and cryoglobulins. **A**—Secondary amyloidosis. Fibrils are all of the same type and measure about 12 nm in diameter. (Uranyl and lead, × 120,000; primary fixation, 1.0% OsO4) **B**—Twenty-manometer fibrils reported herein, some outlined by *circles*. The fibril wall exhibits a double contour and in some, an apparent gap in wall (*upper circle*). Note the admixture of various other fibrils. Second biopsy of Case 8. (Uranyl and lead, × 120,000; line equals 100 nm; primary fixation, 1.0% OsO4) **C**—Cryoglobulinemic nephropathy. Note the dense wall and distinct closed circles on cross-section. Only one type of fibril is present. (Uranyl and lead, × 120,000; primary fixation, 1.0% OsO4)

microtubular fibrils in the range of 7–10 nm. They seem to be distinct from the 11–13-nm group. We have not studied these fibrils in detail.

4) Another small but broad population of fibrils is found to measure 16–32 nm. Rather than being tubular, these fibrils appear to consist of bundles of smaller fibrils or filaments, the latter being similar to the 3–5-nm filaments described above. They are thought to represent the same type of fibrils described in childhood glomerulopathies by Yoshikawa et al.[9]

Discussion

The nature of the 20-nm fibrils remains speculative at this time. They do not seem to fit into the category of natural fibrils.

We found two reports of what might be examples of these fibrils. Both were considered to represent amyloidlike material, although they gave negative results with Congo red and thioflavin-T stains. Thus, Panner[18] reported 2 cases of rapidly progressive glomerulonephritis and possible amyloidosis. We think that his Case 2 is very similar to the present cases in that there is a history of hypertension with both hematuria and proteinuria. Panner describes fibrils in the mesangium and glomerular capillary walls. The designation of rapidly progressive glomerulonephritis is especially intriguing, because many of our patients exhibited crescent formation, and 2 suffered acute renal failure. Rosenmann and Eliakim[19] also report a case with similar fibrils. The patient, a 45-year-old woman, was nephrotic, as were 2 of our cases.

Griffel and Bernheim[20] reported the case of a 47-year-old nephrotic man with numerous subepithelial and intramembranous aggregates of tubular fibrils producing a pattern typical of membranous nephropathy. These tubular fibrils measured 20 nm in diameter and showed a single wall with round, dotlike structures. This, together with their electron density, arrangement in multiple compact aggregates, and scant mesangial involvement, makes it unlikely that they were the same as the fibrils described herein.

The fibrils are not amyloid. Although the localization of the deposits appears to be more like amyloid than other types of fibrils, their staining reactions are

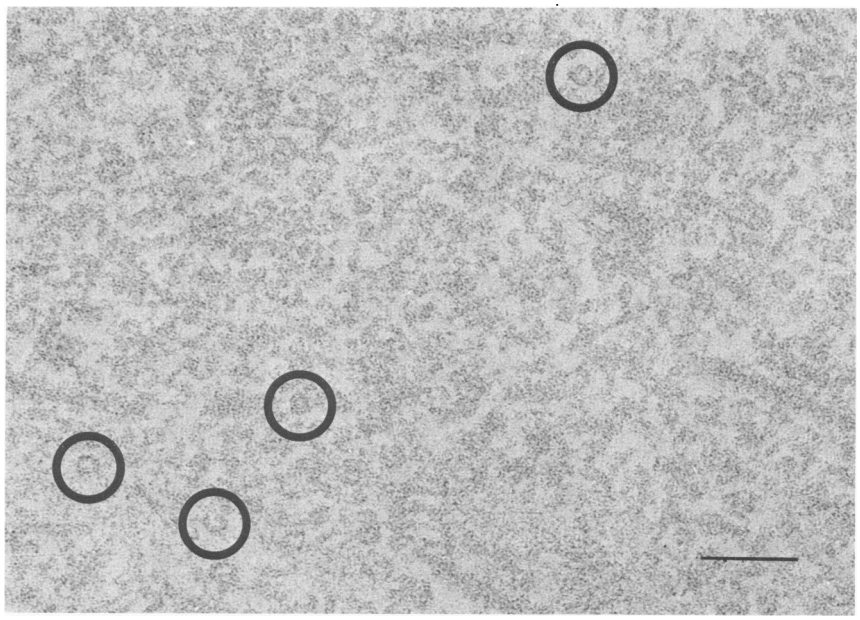

Figure 11 — Another example of 20-nm fibrils (*circles*) mixed with various other fibrils. Note the double contour of the wall and the occasional gap in the wall (*upper and lower circles*). Case 5. (Uranyl and lead, × 120,000; line equals 100 nm; primary fixation, 4.0% paraformaldehyde)

not those of amyloid, in that they do not stain with thioflavin-T nor give green birefringence with Congo red under polarized light.

Amyloid was reported originally as having a width of 7–14 nm in tissue sections.[21] However, subsequent studies describe 10-nm-wide fibrils in tissue sections, while isolated fibrils were found to measure 7.5–8 nm.[22] A later study finds sectioned fibrils to vary from 10 to 15 nm.[23] Our observations of typical amyloid fibrils are in general agreement with this latter figure. The fibrils seen in the present study are significantly larger than this, and their structure is quite different from amyloid. In addition, they tend to be less densely packed, as if surrounded by some type of electron-lucent matrix, and are not infrequently curved or bent. During the present study we compared known examples of both primary and secondary amyloidosis involving the kidneys with the cases in the present study. We found it rather easy to resolve the amyloid fibrils into the 10–15-nm tubular structure usually described, (Figure 10A) and found little resemblance to the 20-nm fibrils described herein (Figure 10B). It was also noted that amyloid deposition tends to be more homogeneous, ie, not so admixed with background fibrils of various sizes as are the 20-nm fibrils. We believe that it is this admixture of natural fibrils that causes the mesangial strands in the cases reported herein to exhibit significant staining with PAS and PASM by light microscopy, as opposed to the usual insignificant staining seen with amyloid.

Fibrils measuring 11–14 nm have been reported in cases of light-chain disease.[10,11] Thus, the size is different, and there is no evidence of light-chain disease in our patients. Our immunofluorescence studies were equally positive with both kappa and lambda light chains, rather than with just one, as would be expected in light-chain disease.[10,24]

Fibrils have also been reported in diabetics. An early report only briefly mentions 10-nm fibrils, which probably represent natural fibrils.[25] Another report describes 10-nm fibrils in the vessels of many organs.[26] The fibrils mentioned in another study of diabetics[27] appear much too large to be related to the fibrils in this report.

Serious consideration must be given to the possibility of cryoglobulins. Cryoglobulins can have a tubular structure but are described as being approximately 20–30 nm wide and even wider,[13–16] are frequently seen to have a very distinctive cylindrical structure, and are often paired in bundles.[13,28,29] There are also cases of glomerulonephritis associated with cryoglobulinemia in which the organized structure of the deposits is not typical, and in some instances they have no structure at all. Cryoglobulins also have thick walls but are denser and form closed circles (Figure 10C). Furthermore, they are closely packed and form electron-dense aggregates, mainly in a subendothelial location, sparing basement membranes[13,14,28,29]; thus, they are usually not admixed with other fibrils. Cryoglobulin determination was done in only one-half of our cases, but all 4 gave negative results.

Could these fibrils be the result of a reparative or scarring process, that is, the result of injury rather than the cause? This would appear to be a possible explanation of the more advanced cases. However, 2 cases (Case 2 and the first biopsy in Case 8) show

what is considered to be early involvement. In these cases there is practically no evidence of glomerular damage except the fibrils, and no diagnosable disease entity is apparent. The major morphologic finding in these cases is mild mesangial widening associated with numerous 20-nm fibrils in the mesangial matrix fibers. Furthermore, these 20-nm fibrils do not stain with PTA and only faintly, if at all, with PASM, nor do they exhibit a periodicity suggestive of collagen formation such as one might expect with scar formation.

If these are not natural fibrils, could they be derived from blood proteins such as immunoglobulins and/or complement? Both of these appear in large amounts in immunofluorescence preparations; yet electron-dense deposits like those usually associated with immune complexes are not present except in very small amounts in 1 case and in trace amounts in another. The localization of the immunofluorescent deposits is not that usually seen. Because of this, we feel that it is the 20-nm fibrils that are staining. It has been observed that both the ninth complement component[30,31] and the membrane attack complex of complement[32,33] can form very short tubular structures with diameters similar to the fibrils described in this study. Also, crystalization of cryoglobulins in the form of tubules is well known.[15] Perhaps immunoglobulins and/or complement components can crystalize in a similar manner in tissues under appropriate conditions. It would not be surprising if this were to occur in conditions where immunoglobulins might be found.

Of course, one should consider another possibility, that the 20-nm fibrils represent some other, as yet unidentified, blood protein to which immunoglobulins and complement attach or with which they coprecipitate.

Except for the 2 cases cited above, we feel that these fibrils are different from most previously reported and that they are associated with distinct clinical symptoms. One obvious and unanswered question is whether or not these 8 cases represent a specific disease entity. We are reporting them together because they have clinical features in common: hematuria, proteinuria, hypertension, and renal insufficiency, and because their glomerular mesangia and basement membranes suffer accumulations of fibrils which seem to be the same by all the parameters we can use. We thus consider them as much an entity as, for example, cryoglobulinemic nephropathy. We must conclude that we cannot identify their nature or origin at this time, nor whether their occurrence is limited to the kidney.

Whether these fibrils are related to complement, immunoglobulins, or immune complexes is uncertain, but should be considered. Unless renal biopsies are studied by electron microscopy, the fibrils are apt to be missed and renal disease associated with them misdiagnosed, because the light-microscopic picture and immunofluorescence findings may mimic mesangiocapillary as well as rapidly progressive glomerulonephritis.

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