Kidney biopsy –
technique, interpretation, evaluation

Eberhard Ritz
Heidelberg (Germany)

History :

Brun C. et al

Kidney biopsy in acute glomerulonephritis

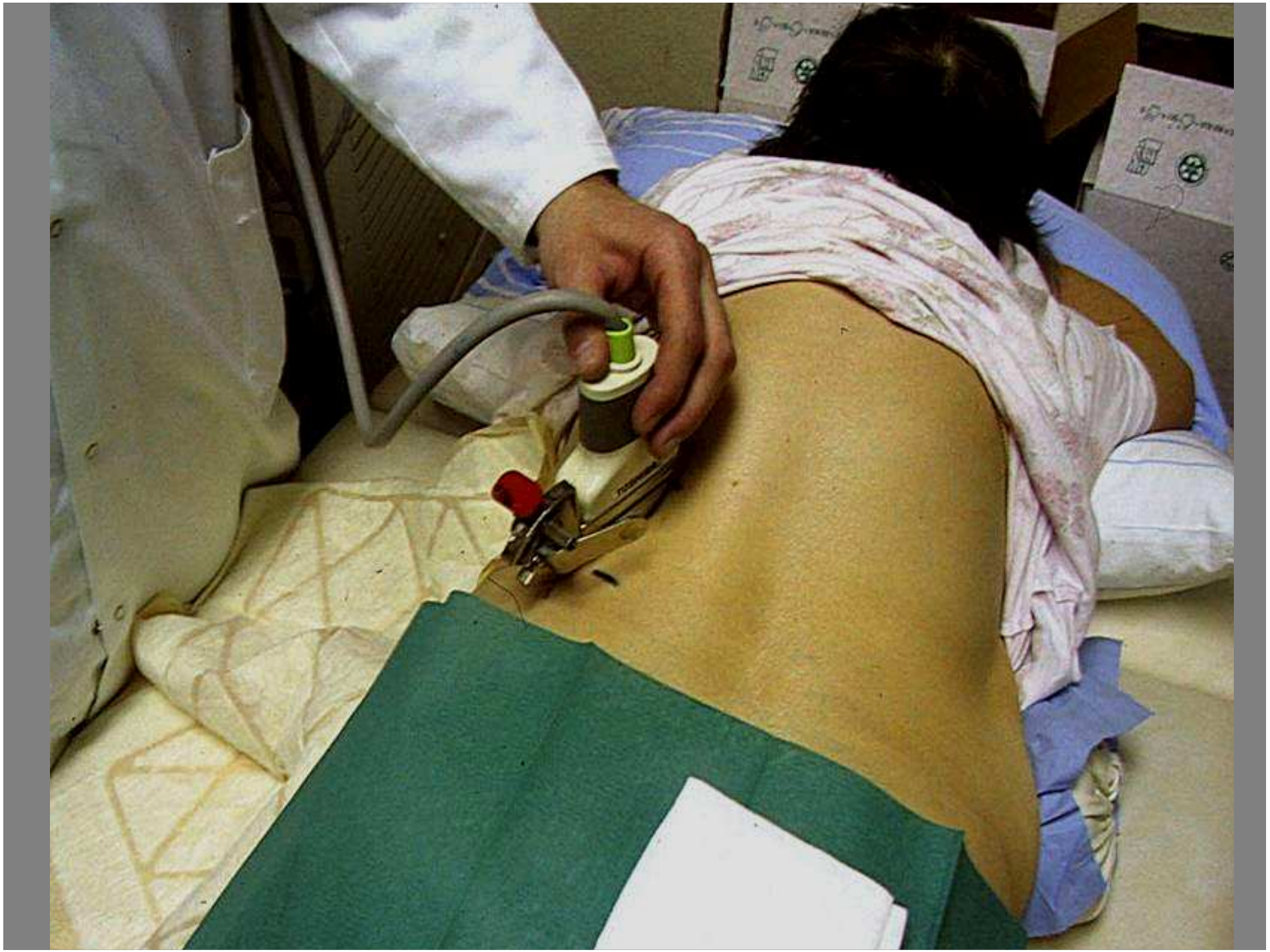
Acta Med. Scand.(1958) 166: 155

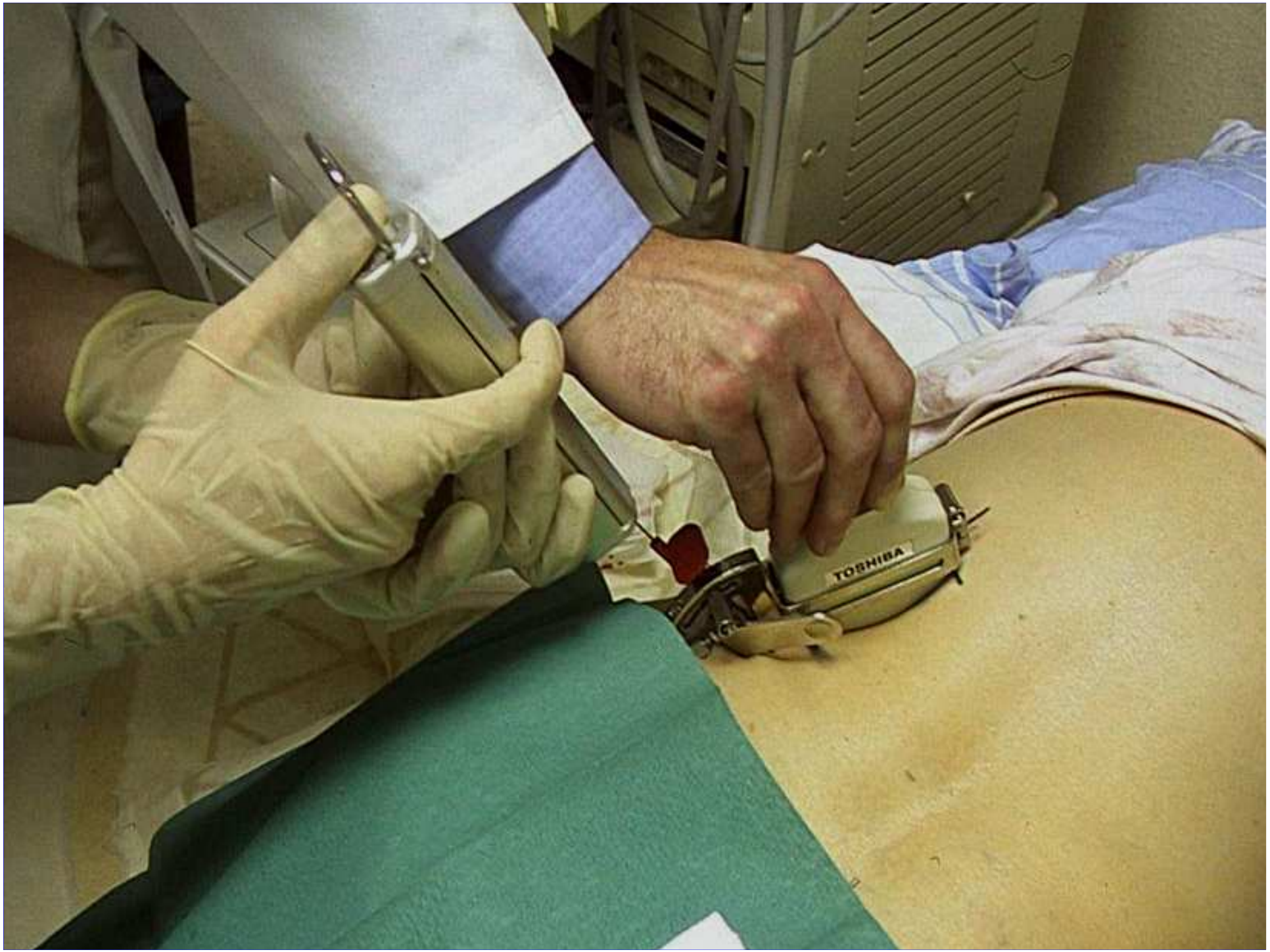
percutaneous renal biopsy

Biopty^{R} gun (*Radioplast AB*)

adaptor (UAGV 009A, Toshiba Co.)

- *primary biopsy : overnight in hospital ?*
- *transplant protocol biopsy : 12h observation and then dismissal*

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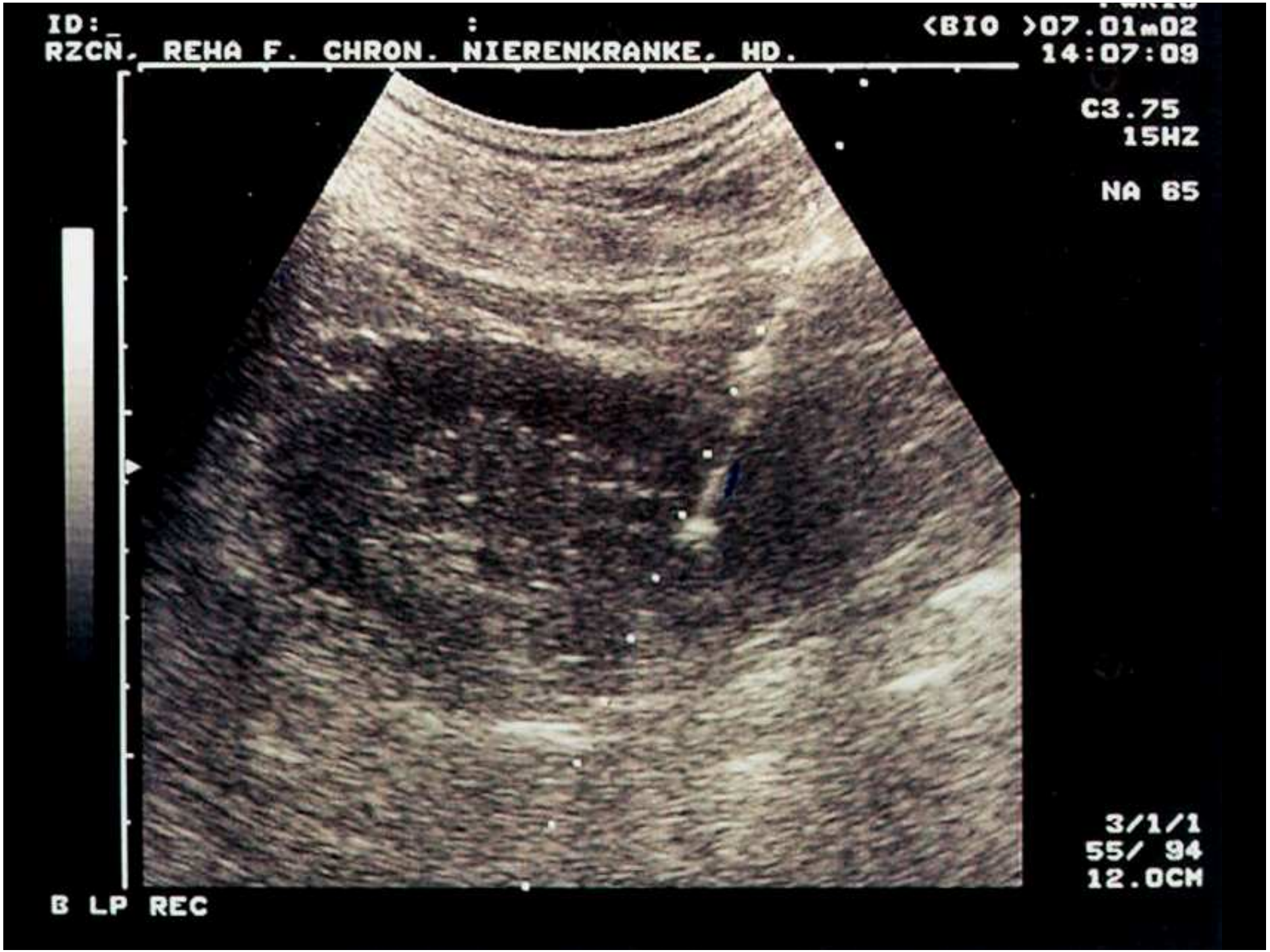
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Complications

- 1/1090 cases requiring interventional radiology (coil)
- 3/1090 cases blood transfusion
- no persisting hemodynamically relevant av fistules
- small hematomas (> 2x2 cm without decrease of Hb)

  in 2,2%

Hergesell O, NDT (1998) 13: 975

Renal biopsy only when :

- *strict normotension (< 140/90 mmHg)*
- *normal coagulation parameters (PT, PTT, f VIII, thrombocyte counts, bleeding time)*
- *no Aspirin or other non-steroidal antiinflammatory agents for at least 5 days*
- *no (active) urinary tract infection*

Main indications for renal biopsy :

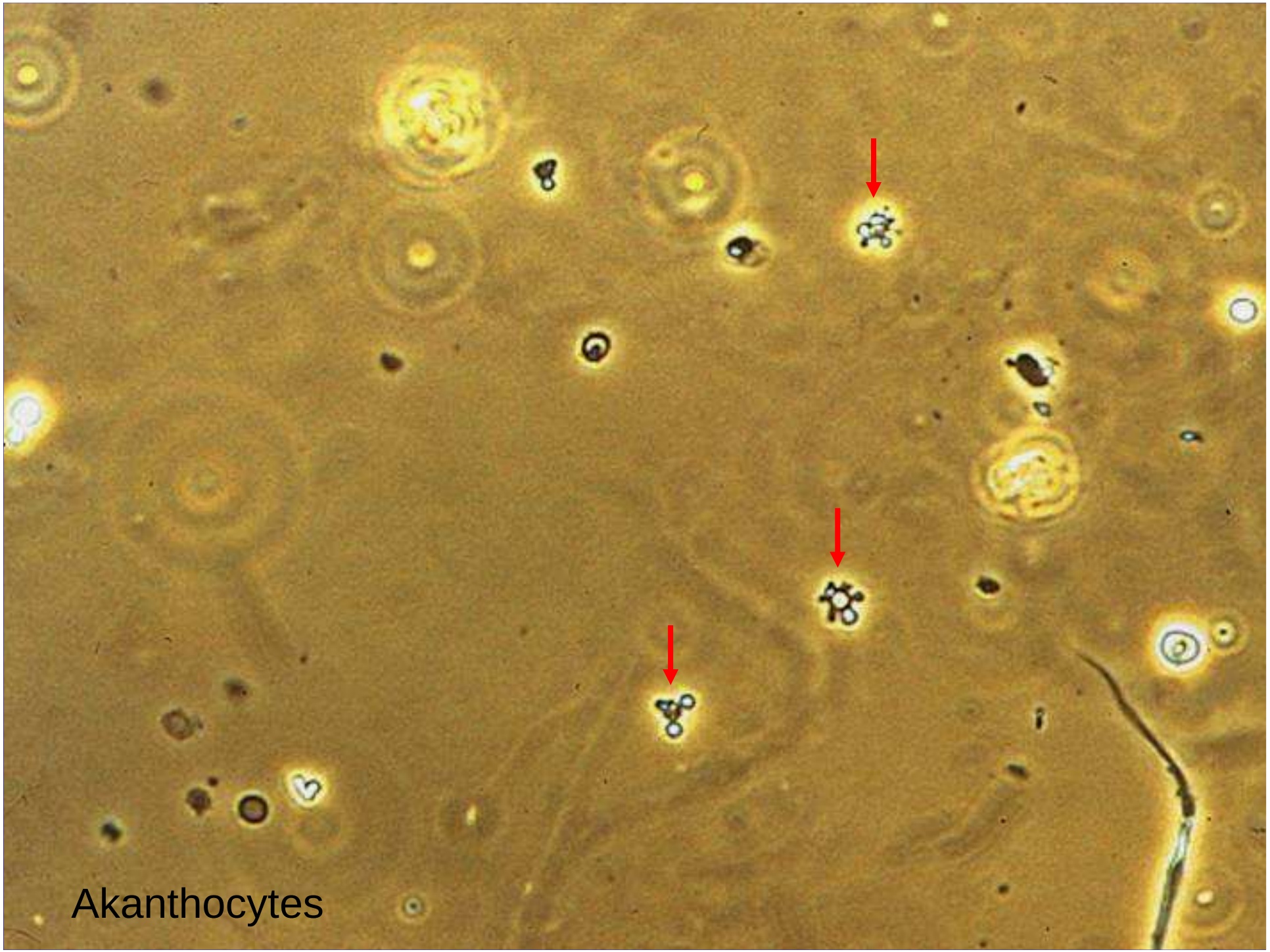
 # abnormal proteinuria and
 # pathological urinary sediment (phase contrast)

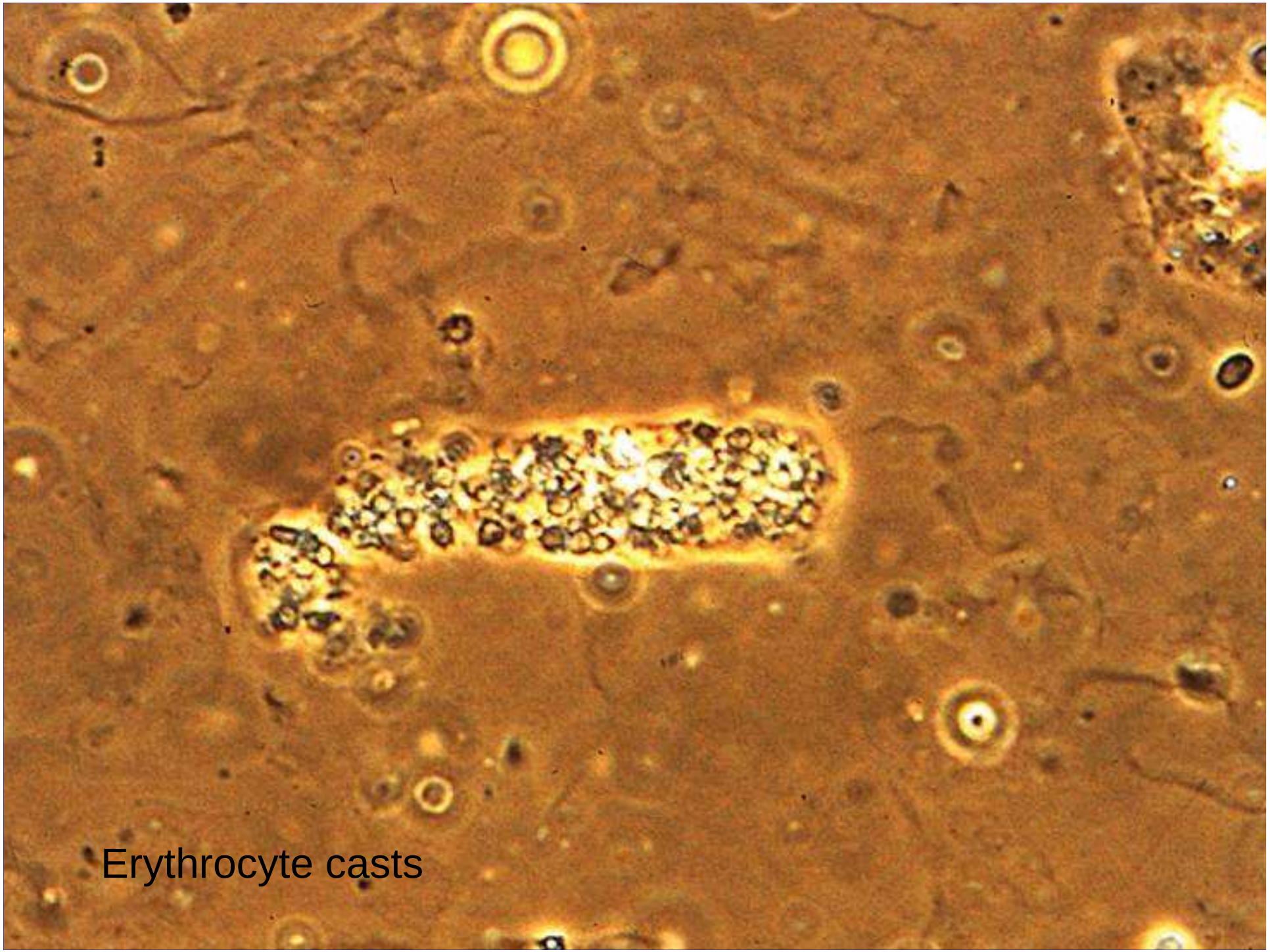
Main purpose of biopsy information :

***# # what is prognosis of known renal disease* :**
activity of the process
(e.g. rebiopsy vs watchful waiting)
 # is there a renal problem
(e.g. vs orthostatic proteinuria)
 # renal vs postrenal problem
(biopsy vs urological investigation)

Urinary sediment –

the Cinderella of nephrological investigations


Akanthocytes


Erythrocyte casts


Cellular casts

Pathophysiology/Complications

ORIGINAL ARTICLE

Acanthocytes in the Urine

Useful tool to differentiate diabetic nephropathy from glomerulonephritis?

GUNNAR H. HEINE, MD
URBAN SESTER, MD
MATTHIAS GIRNDT, MD
HANS KÖHLER, MD

Diabetes Care(2004) 27:190

of patients who are most likely to have a nondiabetic glomerulopathy and to selectively perform renal biopsy in this subgroup of patients.

Urinary erythrocyte morphology ex-

Hematuria > 8 red blood cells/µl
-84% of patients with glomerulonephritis
-62% of patients with diabetic nephropathy
-20% of healthy individuals

Acanthocytes < 5%
-46% of patients with glomerulonephritis
-4% of patients with diabetic nephropathy

be aware:

IgA-glomerulonephritis frequently associated with diabetic nephropathy

Gans, Am.J.Kidn.Dis.(1992) 20:255
van der Niepen, Nephrol.Dial.Transplant.(1995) 10:1254
Cavarape, Nephrol.Dial.Transplant.(2005) 20:1514

Advances in urinary sediment

- *Szeto, Clin.Nephrol.(2005) 64:337*

  Telomer shortening in urinary sediment of patients with IgA glomerulonephritis

- *Szeto, Am.J.Kidn.Dis.(2006) 47:578*

  mRNA expression of target genes as a noninvasive indicator of CKD

- *Wang, J.Rheumatol (2007) 34:2358*

  Messenger RNA expression of podocyte-associated molecules in urinary sediment of patients with lupus nephritis

Investigations prior to renal biopsy

- kidney **ultrasonography**
  - *single kidney?*
  - *orthotopic?*
  - *normal width of parenchyma?*
  - *aberrant vessels (Duplex-sonography)*
- blood pressure **< 140/90 mmHg**
- normal PT, PTT, f VIII, thrombocytes, **bleeding time**
- sterile urine culture

Yield of kidney biopsy

-sufficient tissue for histopathology
1077/1090 = 98,8%

- Glomeruli per cylinder
n=9 (1-37)

Hergesell O, NDT (1998) 13: 975

Dream of pathologists : entire kidney (*diagnostic autopsy*)
Consensus conference (Wien, 2001) :
8 glomeruli, optimal 10-15

http://www.kidney-euract.org/Rbpathyology consensus.htm

“If the percentage of glomerular involvement in a biopsy is used to determine the severity of a focal lesion, a small biopsy sample size will lead to considerable misclassification of disease severity and will make exclusion of focal disease difficult“

Corwin, Am.J.Nephrol.(1988) 8:85

- *2 biopsy cylinders*
- *minimal length 1 cm*
- *diameter 1.2 mm*

isotonic saline – fast local transport

- *cryopreservation of one piece for immunefluorescence*
- *fixation with paraformaldehyde or buffered (4%) formaldehyde for paraffin embedding*
- *fixation with 3% glutaraldehyde for electron microscopy*

or

direct fixation with paraformaldehyde or formaldehyde and shipping (indirect immunehistology by APAAP (alkaline phosphatase) or others

Nephrol Dial Transplant (2006) 21: 1157–1161
doi:10.1093/ndt/gfk037
Advance Access publication 9 January 2006

What you should know about the work-up of a renal biopsy

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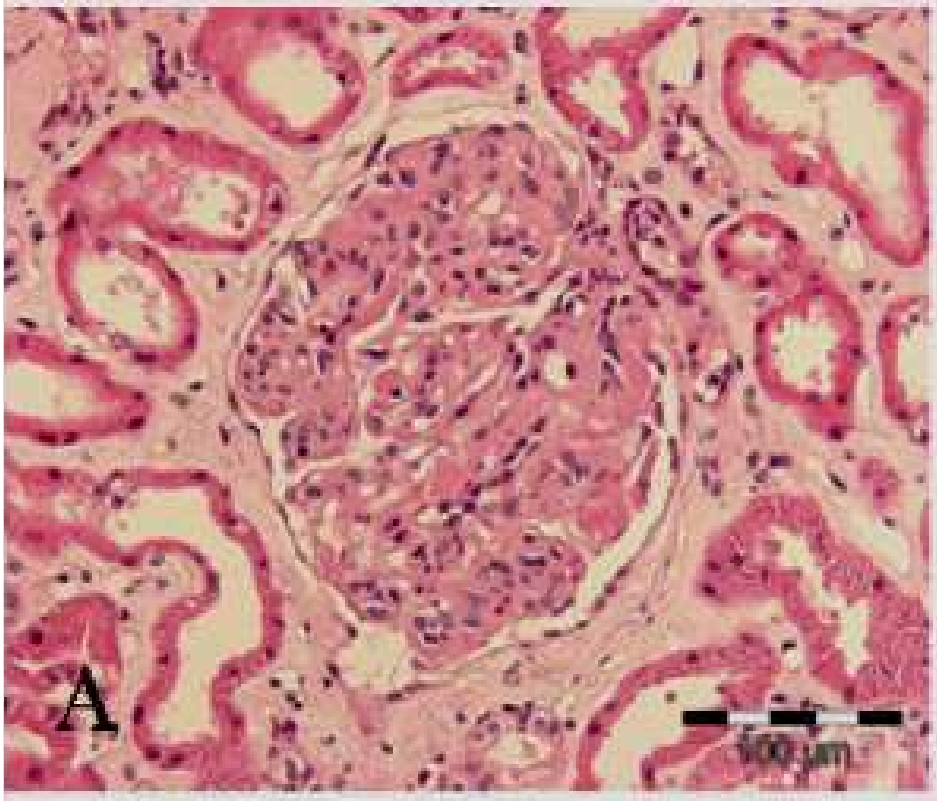
Departments of [1]Pathology and [2]Internal Medicine IV, University of Erlangen-Nürnberg, Germany

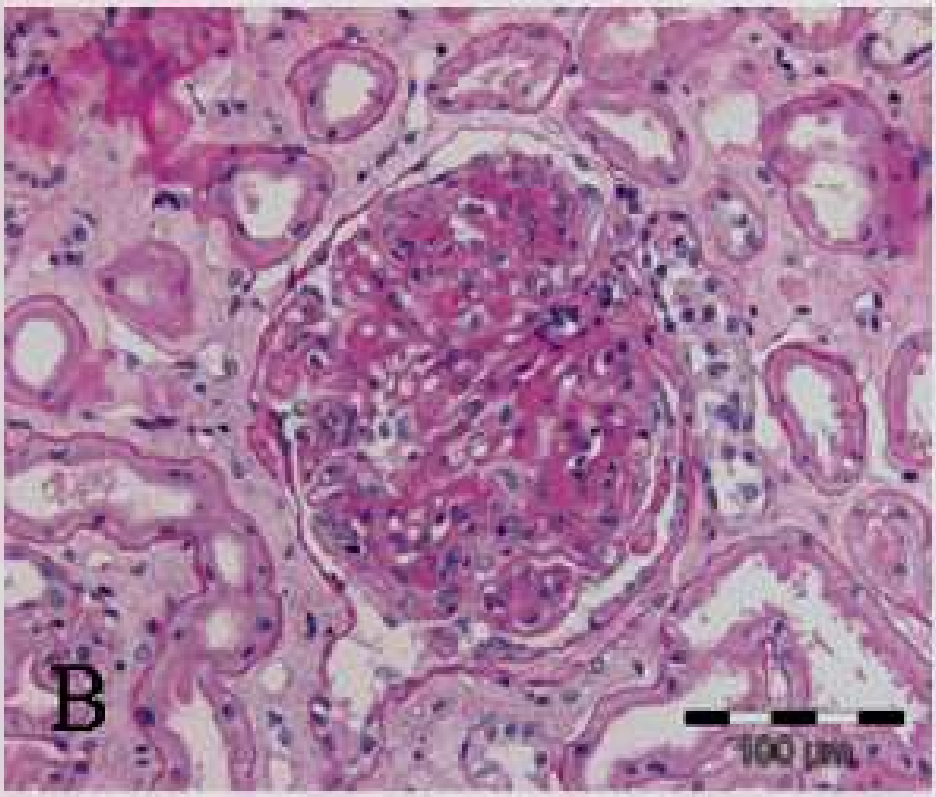
Routine stains

information complementary

HE-stain
(hematoxylin-eosin)

PAS-stain
(Periodic acid-Schiff's base)



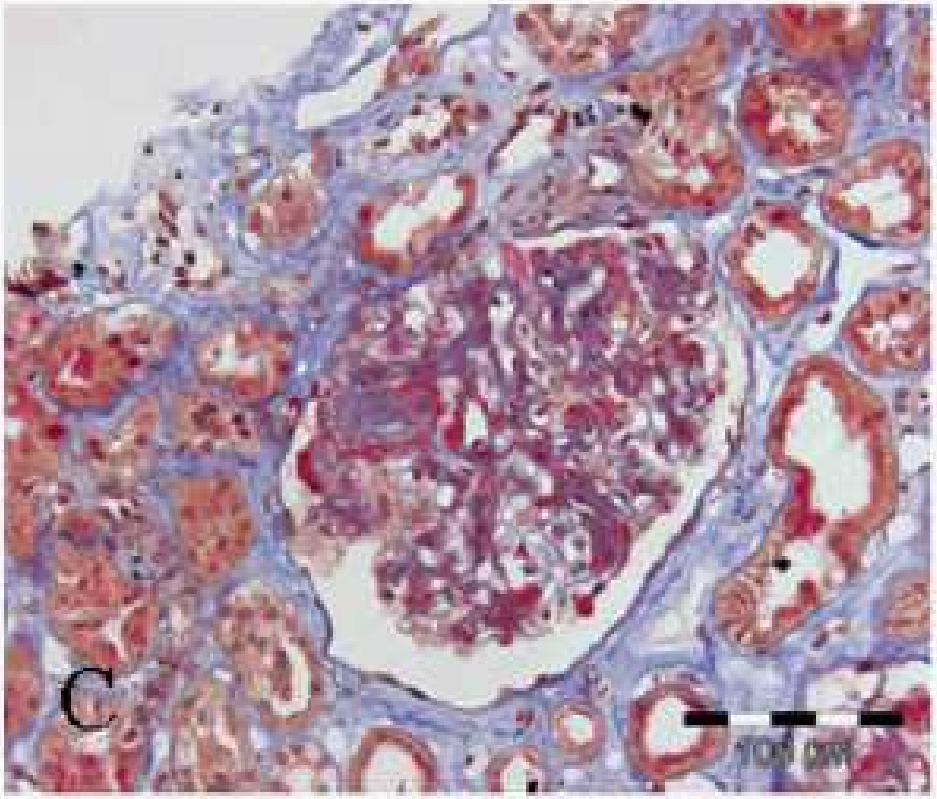


composition of tissue:
cortex vs medulla,
number of glomeruli
cellular infiltrates ..

analysis of glomerulus and vessels:
glomerular cells,
mesangial matrix
glomerular basement membrane
fibrinoid necrosis ..

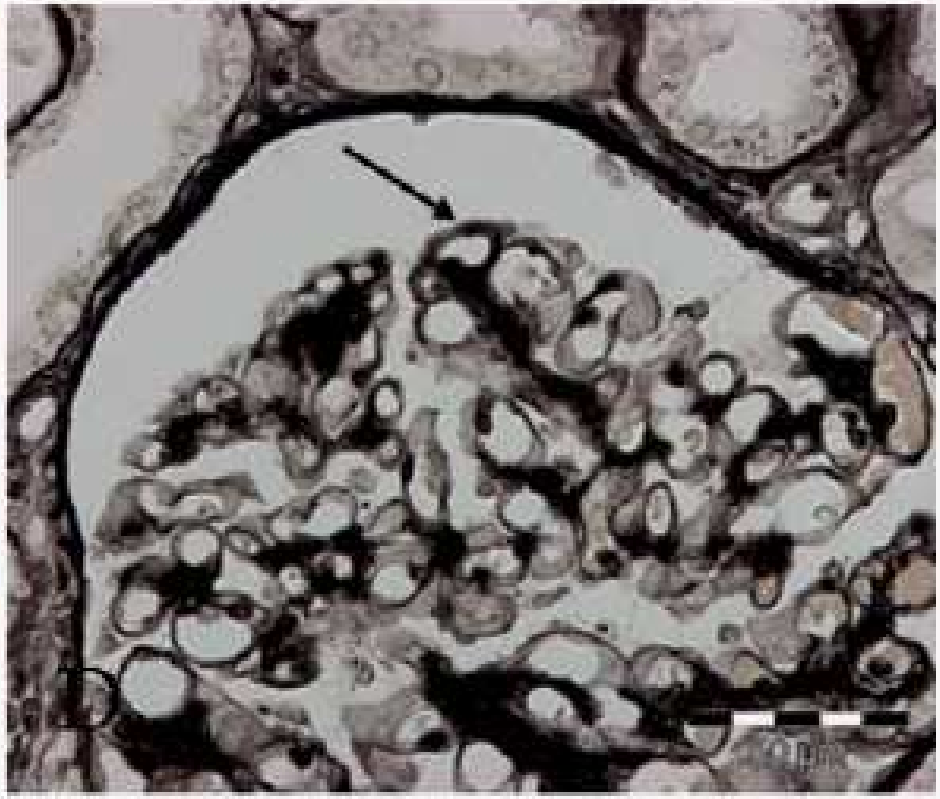
Routine stains

protein stain
(SFOG; *acid fuchsin-Orange G*)



immune deposits
in this section: subendothelial and intramembranous

silver stain

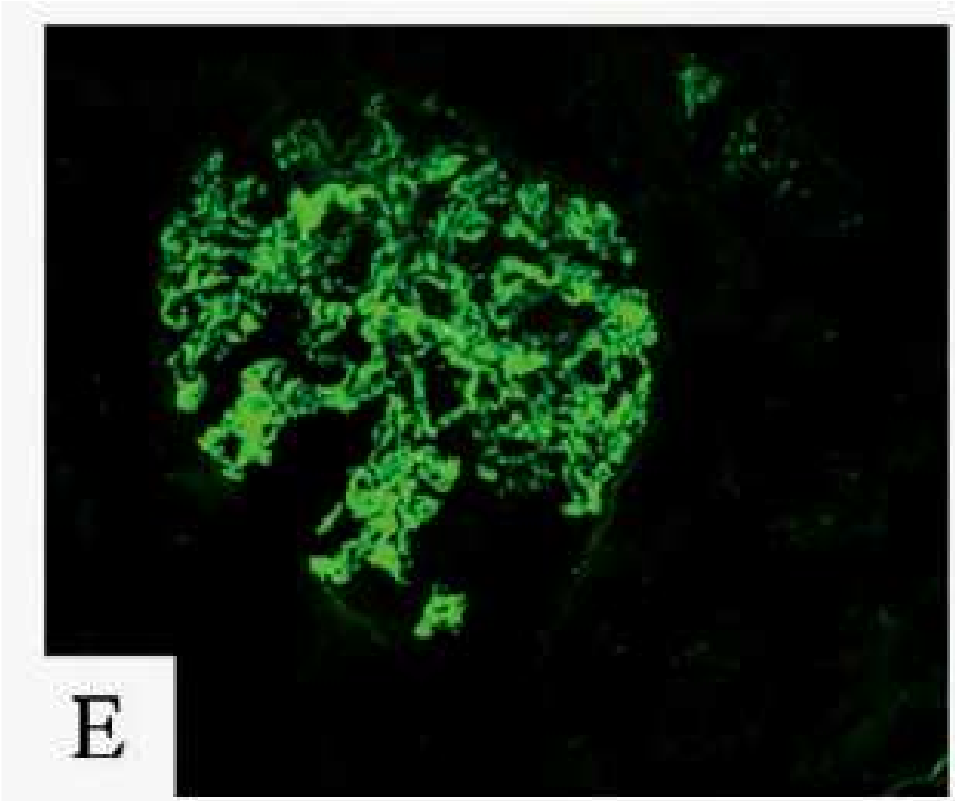


changes of basement membrane
e.g. thickening, reduplication, spikes

for specific indications Congo red or Kossa stain

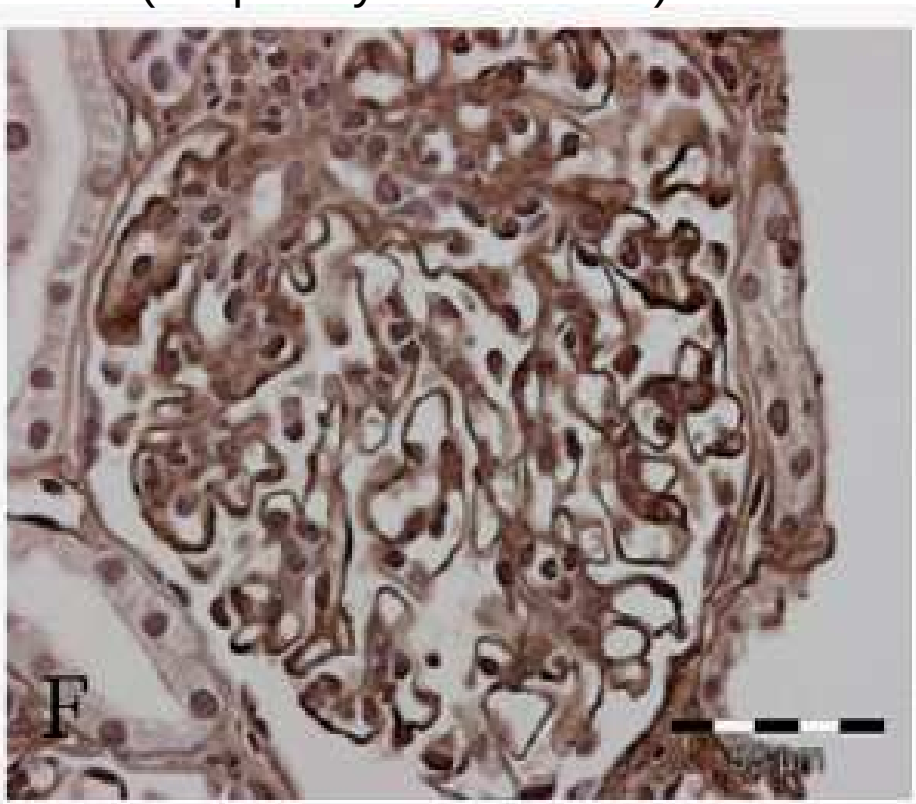
Routine stains

Immunefluorescence



immune deposits:
here IgG ab, IgG deposits

Immunehistochemistry
(antiperoxydase method)



immune deposits:
antiperoxydase method

Electron microscopy :

glutaraldehyde fixation or initially formaldehyde

- **Alport's disease**
- **thin basement membrane disease**
- **immunotactoid disease**
- **minimal change nephropathy** *(foot process fusion)*
- **(amyloidosis, L-chain deposit disease...)**
- **(lupus:** *fingerprints; tubuloreticular structures***..)**

Why renal biopsy ?
What are the goals ?

1. **diagnosis (*specific therapy?*)**
2. **prognosis**
3. **acute (reversible) vs chronic (irreversible) changes ⇨**

   (e.g.aggressive immune suppression yes/no)

4. **diagnosis of superimposed second kidney disease,** *e.g. IgA-GN after Wegener's granulomatosis, Wegener's granulomatosis + Goodpasture, etc.*

For which nephrologic syndromes should renal biopsy be considered :

- **asymptomatic hematuria**

  (dysmorphic erythrocytes,erythrocyte casts) ± proteinuria

- **nephrotic syndrome**

- **acute nephritic syndrome**

- **rapidly progressive glomerulonephritis !!!**

  (nephrological emergency)

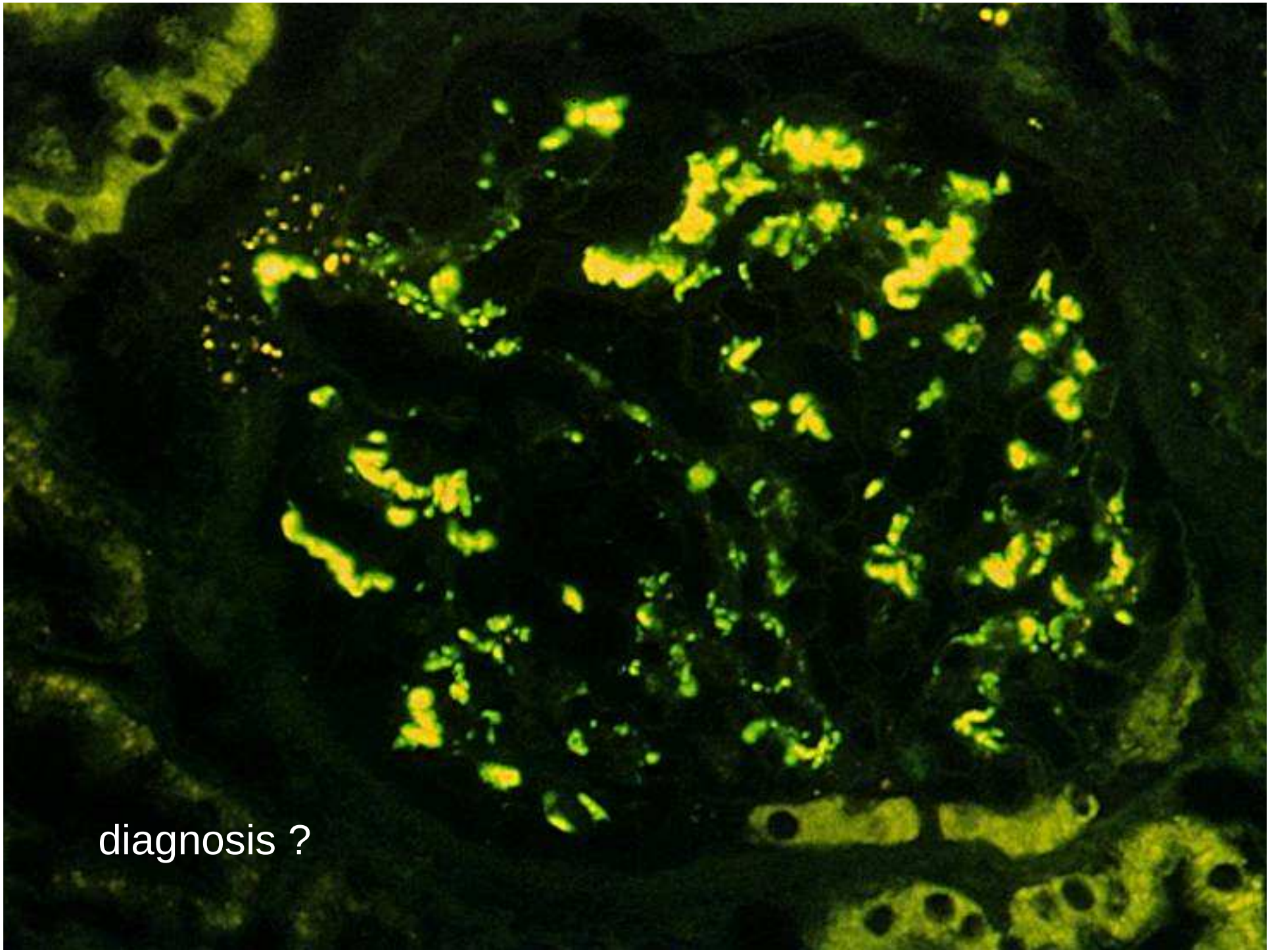
- **(chronic renal failure; CKD)**

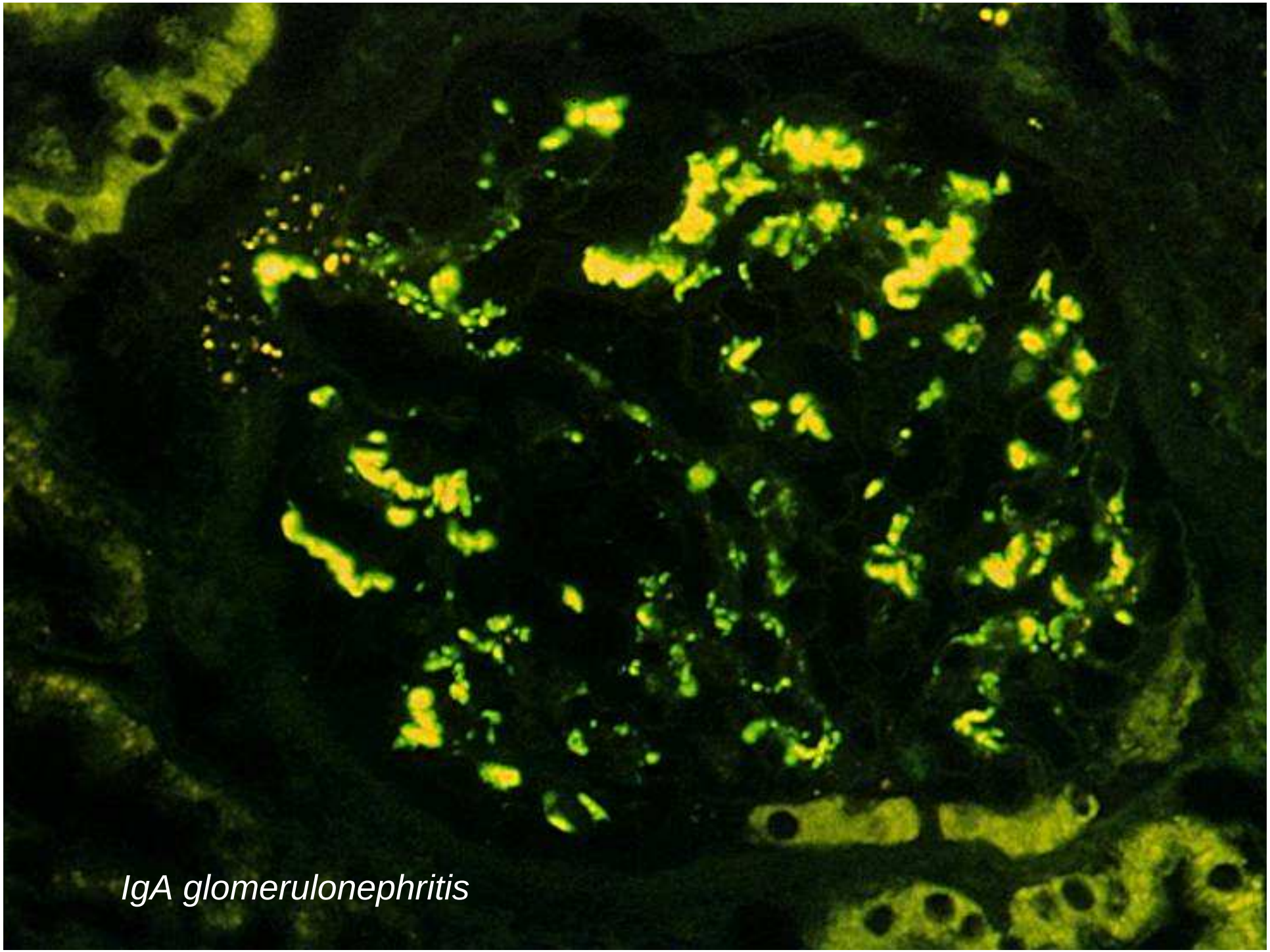
- **(acute renal failure; AKI)**

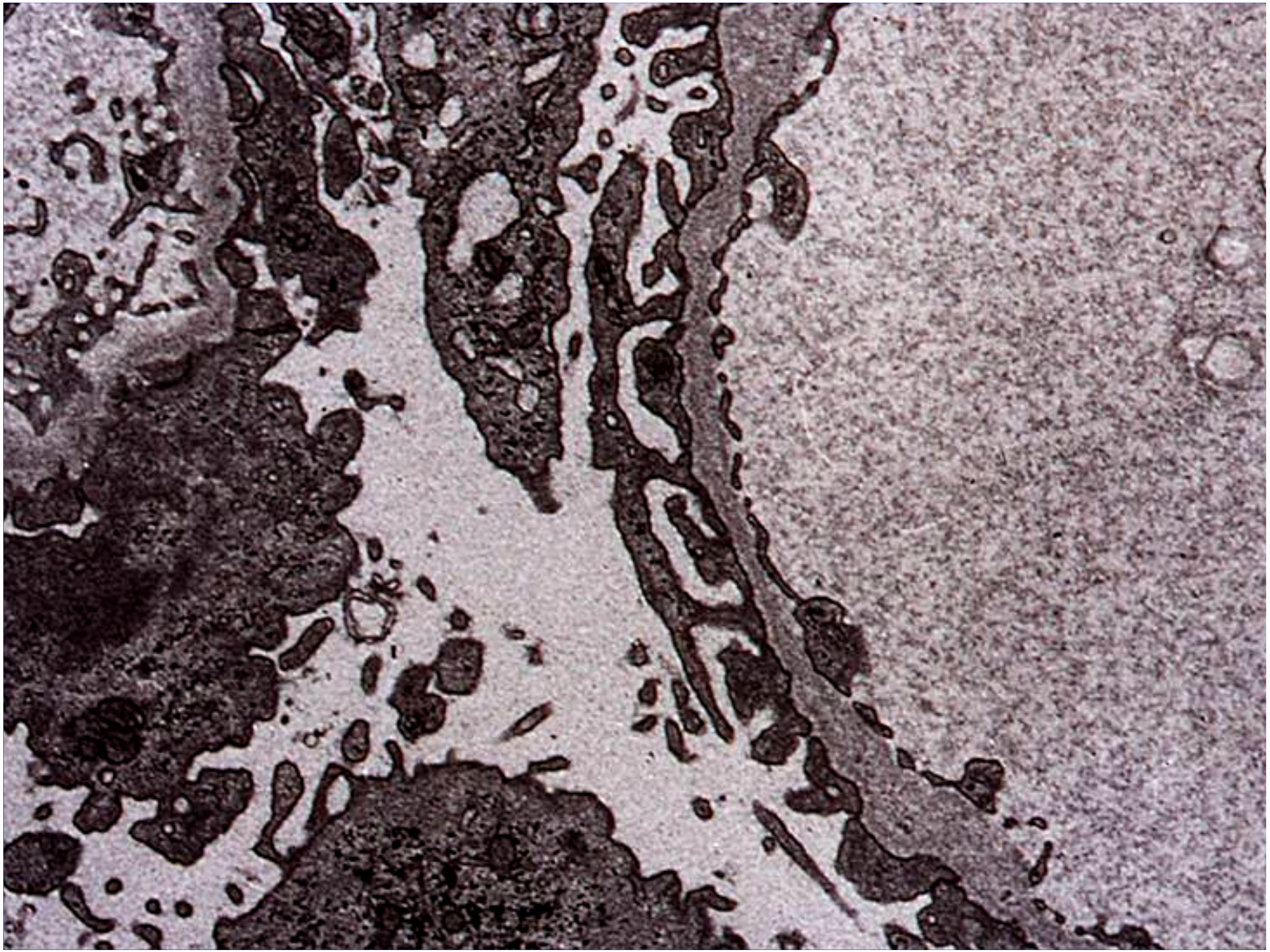
- **renal transplantation**

Differential diagnosis of asymptomatic hematuria ± proteinuria

- *IgA glomerulonephritis*
- *Alport's disease*
- *(thin basement membrane disease)*
- *lupus erythematosus*
- *(initially) isolated renal manifestation of vasculitis*
- *membranoproliferative glomerulonephritis*


diagnosis ?


IgA glomerulonephritis



- *85% of X-linked Alport mutation :*
  COL5A5 gene
  (hematuria, proteinuria, renal failure
  sensorineural hearing loss, lenticonus, oil droplet sign)
  alternative to renal biopsy :
  skin biopsy (absent COLI4α5 epitope)

- *autosomal recessive Alport :*
  compound heterozygotes or homozygous for mutations in COL4A3 or COL4A4 genes

- *concomitant mutations in COL4A6 :*
  additional leiomyomatosis

Benign hematuria
~ thin basement disease

- **1-14% of general population**

  Savige, Brit.Med.J. (2001) 322: 942

- **genetically heterogeneous (in 40% mutations in genes COL4A3/COL4A4), 2/3 have autosomal dominant form**

  Tazón,Am.J.Kidn.Dis.(2003) 42:952

- **GBM attenuated diffusely as in early Alport or carriers of autosomal recessive Alport**

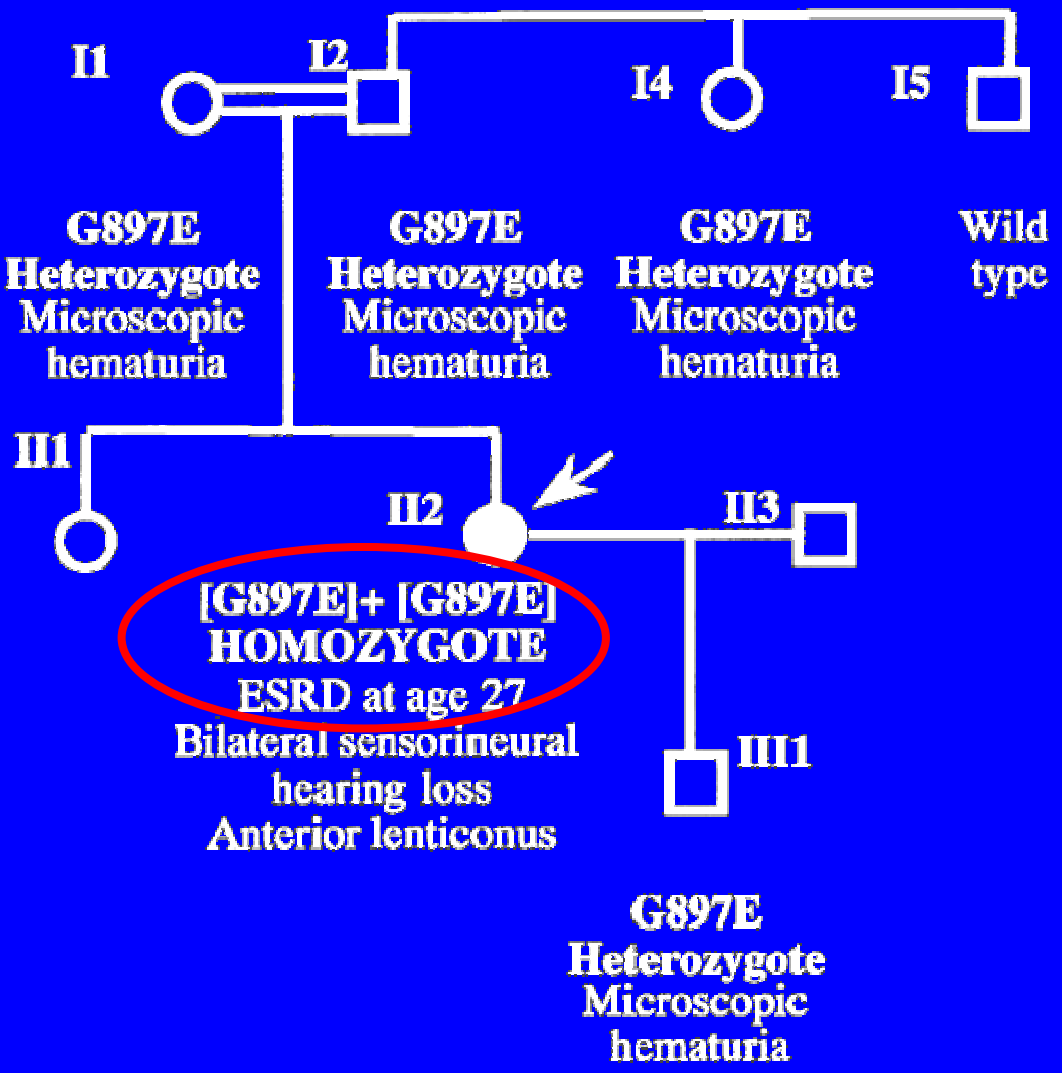
- **some cases develop proteinuria, hypertension and impaired renal function**

  Nieuwhof, Kidn.Intern.(1997) 51:1596

- **benign familial hematuria =**
  may be carrier state for autosomal recessive Alport syndrome

Problems of “benign hematuria“

two heterozygotes may produce offspring with Alport syndrome



Tazón (2003) Am.J.Kidn.Dis. 42: 952

Diagnostic yield of kidney biopsy –

isolated microhematuria vs. microhematuria plus moderate proteinuria

89 pat. isolated microhematuria,
46 pat. microhematuria and moderate proteinuria (<2,5g/Tag)
hypertension in 36% and 56% respectively

isolated microhematuria	
thin basement nephropathy	*43 %*
IgA-glomerulonephritis	*20 %*
nonspecific abnomalities	*19 %*
normal	*18 %*
microhematuria plus proteinuria	
IgA-glomerulonephritis	*46 %*
other forms of GN	*26 %*
nonspecific abnormalities	*17 %*
normal	*4 %*

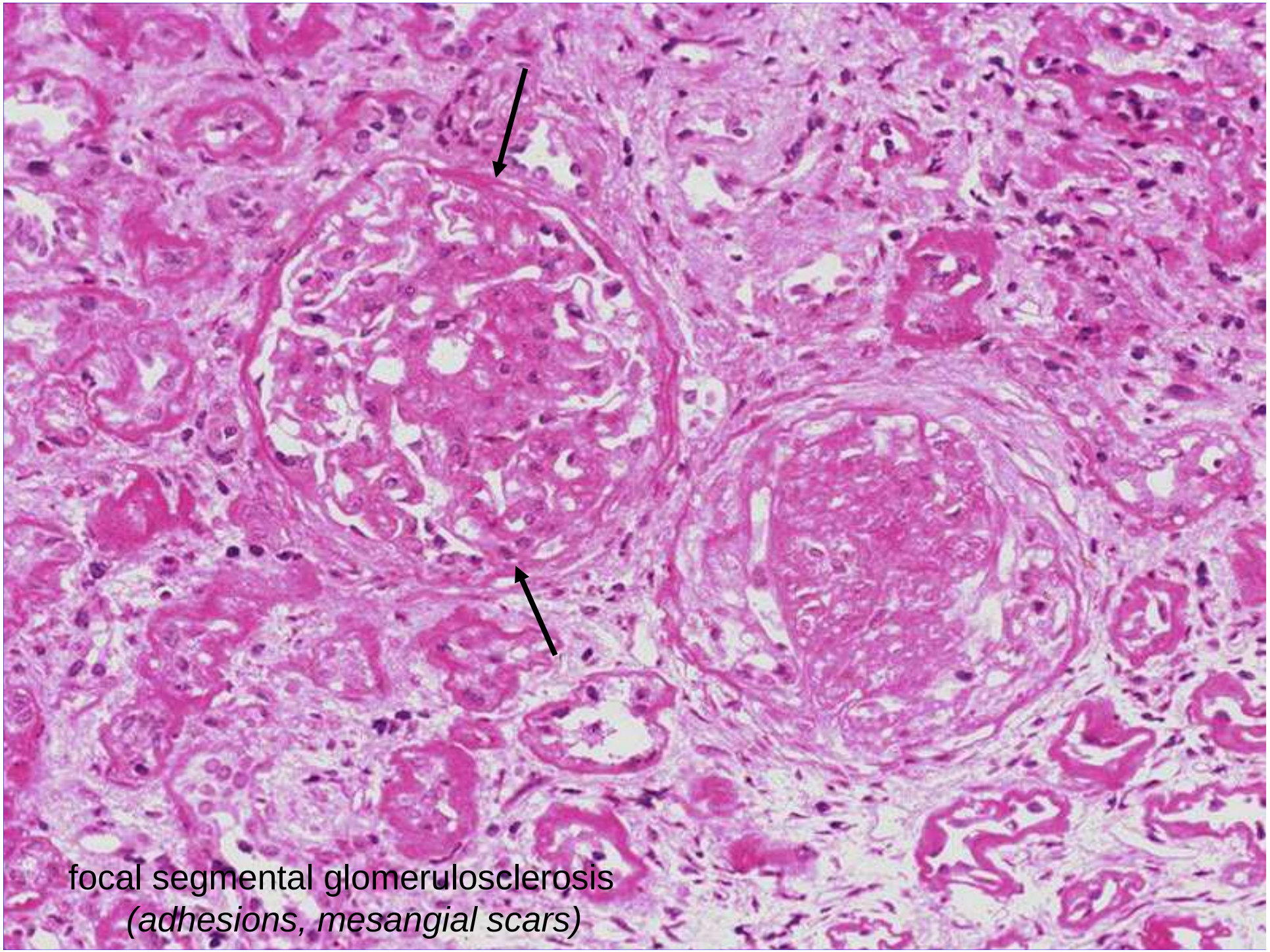
Hall, Clinical Nephrol. (2004) 62:267

For which nephrologic syndromes should renal biopsy be considered :

- asymptomatic hematuria
  (dysmorphic erythrocytes,
  erythrocyte casts) ± proteinuria
- nephrotic syndrome
- acute nephritic syndrome
- rapidly progressive glomerulonephritis !!!
  (nephrological emergency)
- (chronic renal failure CKD)
- (acute renal failure AKI)
- renal transplantation

Differential diagnosis of the nephrotic syndrome

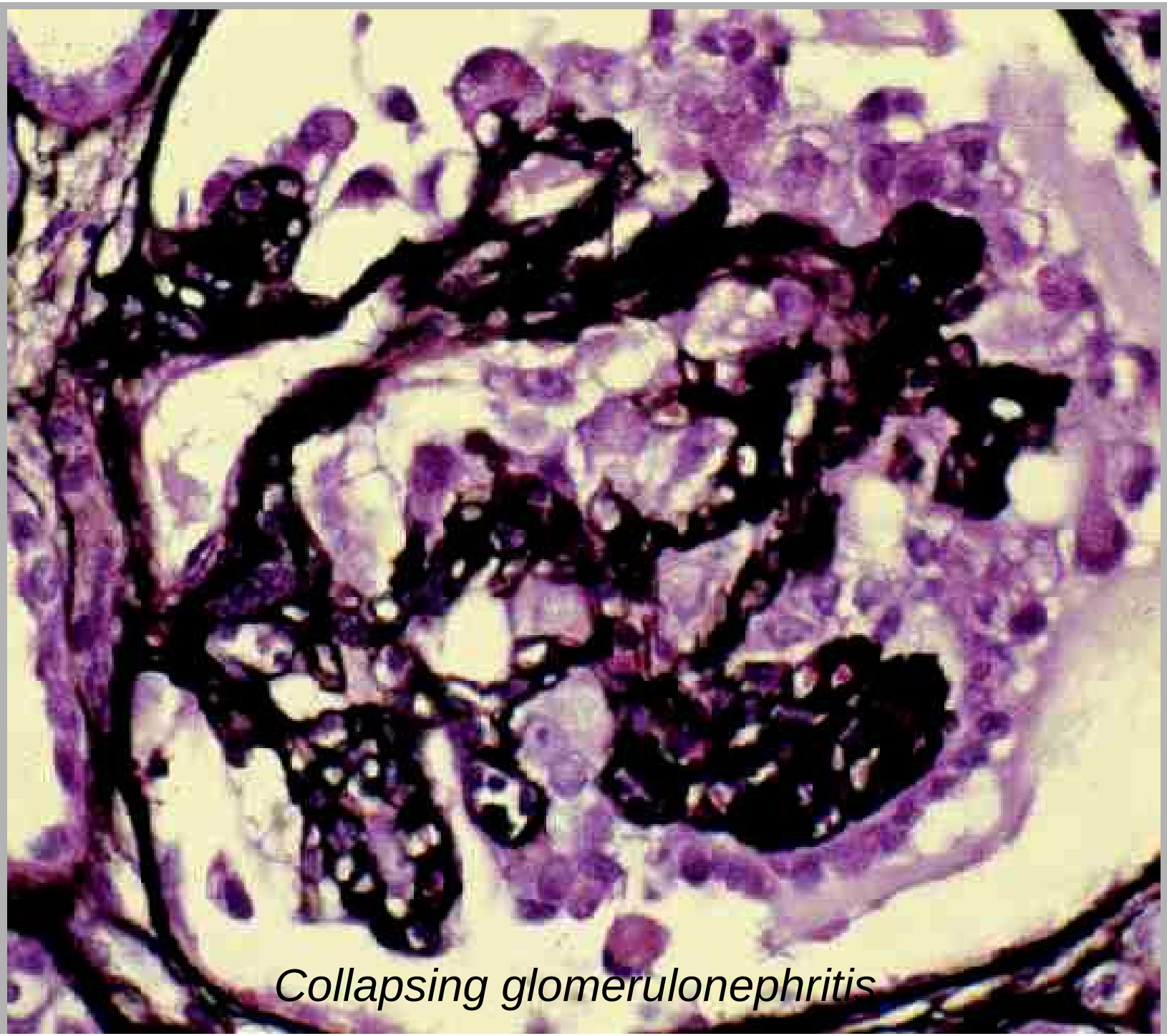
- **minimal change nephropathy/**
  focal segmental glomerulosclerosis (FSGS)
- **primary glomerulonephritis** — **membranous GN**, **membranoproliferative GN**
- **systemic diseases** — **diabetes *(type 1 and 2)***
- **heredofamilial renal disease** — **Alport‘s syndrome**, **nail patella syndrome**
- **protein deposit diseases** — **amyloidosis** *(AL, AA, transthyretin, fibrinogen.....)*, **light chain deposit disease**, **(heavy chain deposit disease)**


focal segmental glomerulosclerosis
(adhesions, mesangial scars)

focal-segmental glomerulosclerosis (FSGS) – potential causes

- ***primary (idiopathic) FSGS***

- ***HIV- or Heroin-associated FSGS***
  (collapsing nephropathy)



Collapsing glomerulonephritis

Secondary focal-segmental glomerulosclerosis
potential causes

reduced renal mass

oligomeganephronia
unilateral renal agenesis } *HNF1, OFD,Wt1,ROBO2*
renal dysplasia
reflux nephropathy
massive surgical loss of renal mass
chronic renal graft failure

normal renal mass

diabetes
obesity
cyanotic cardiac disease

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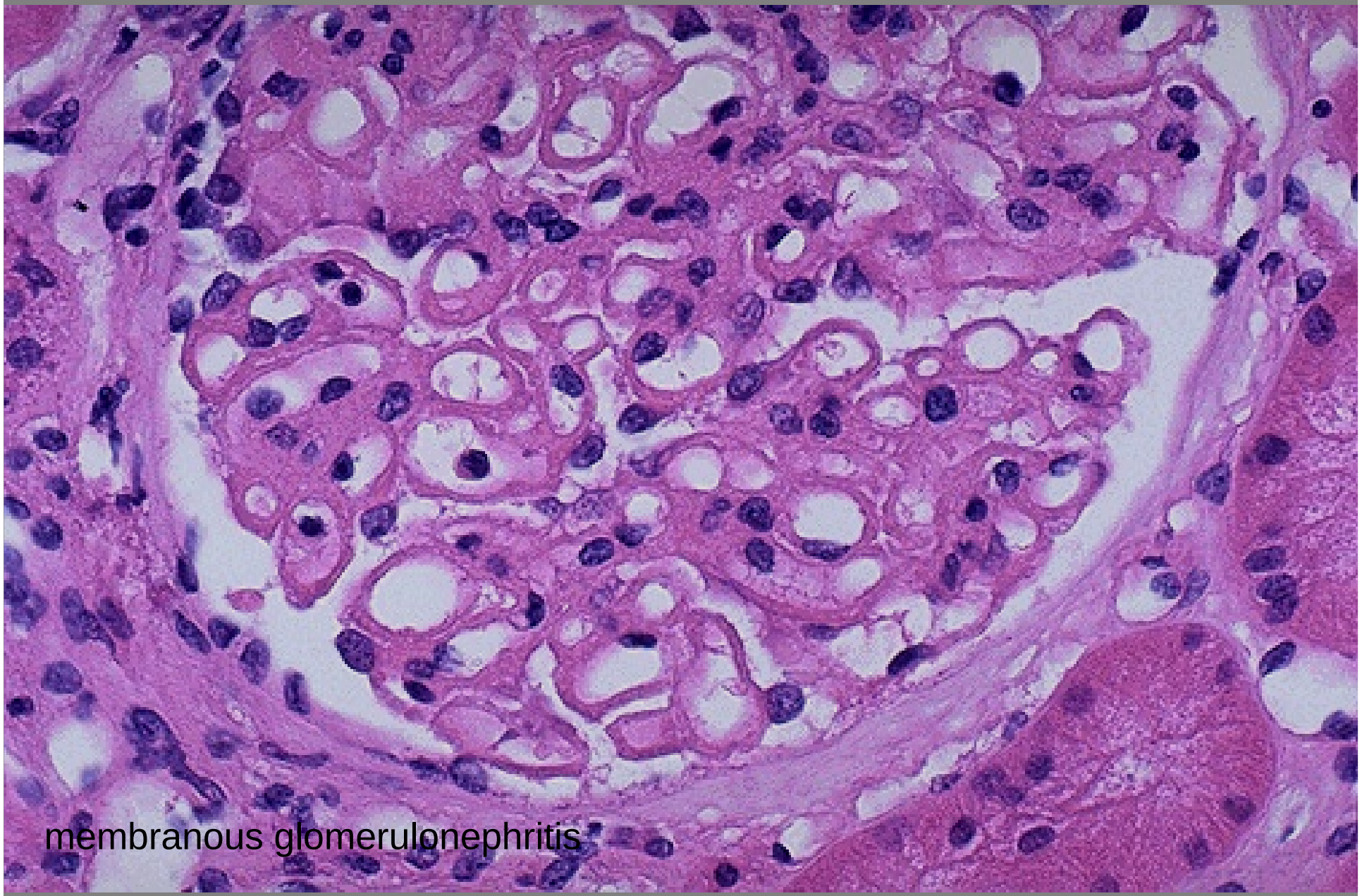
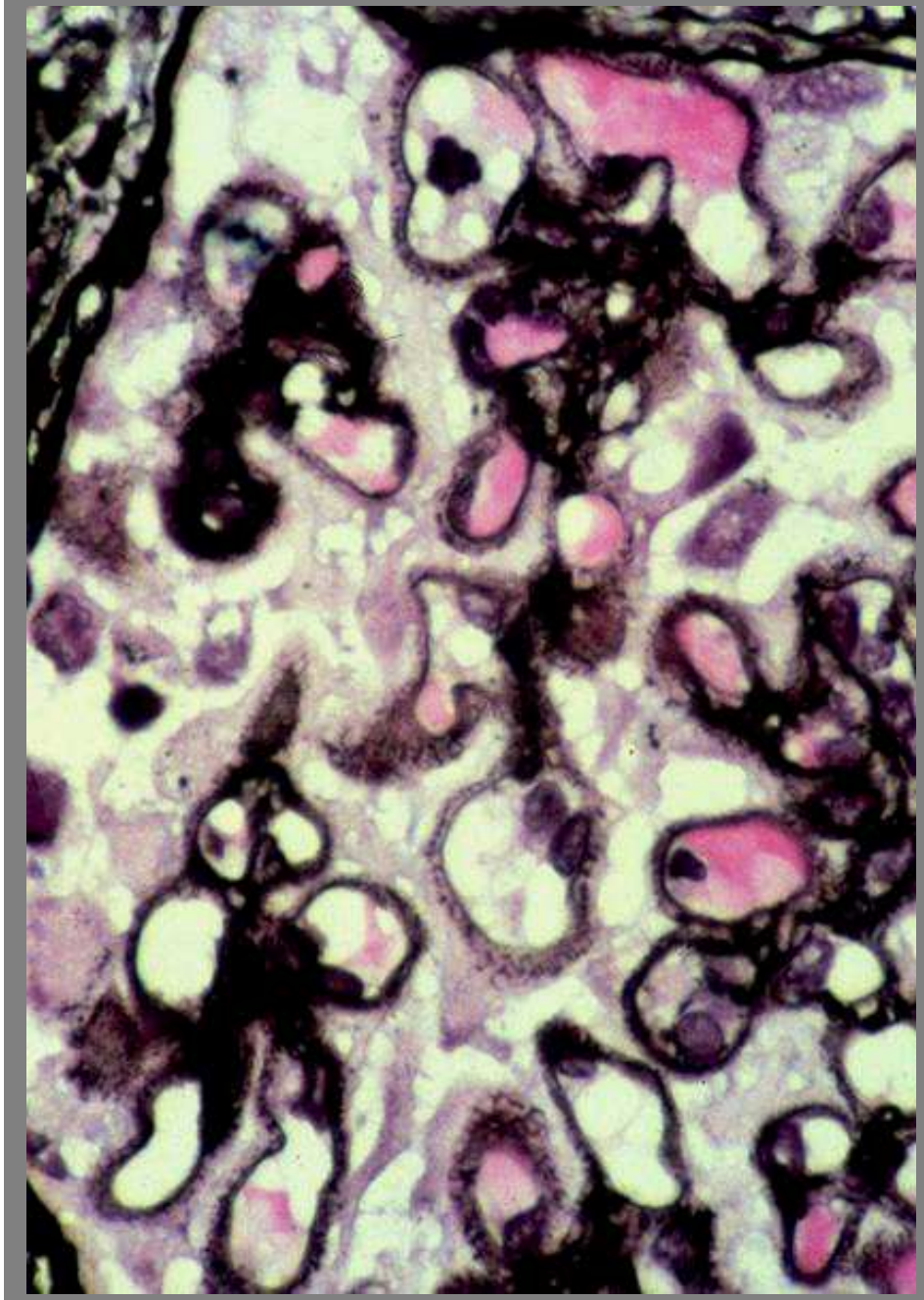
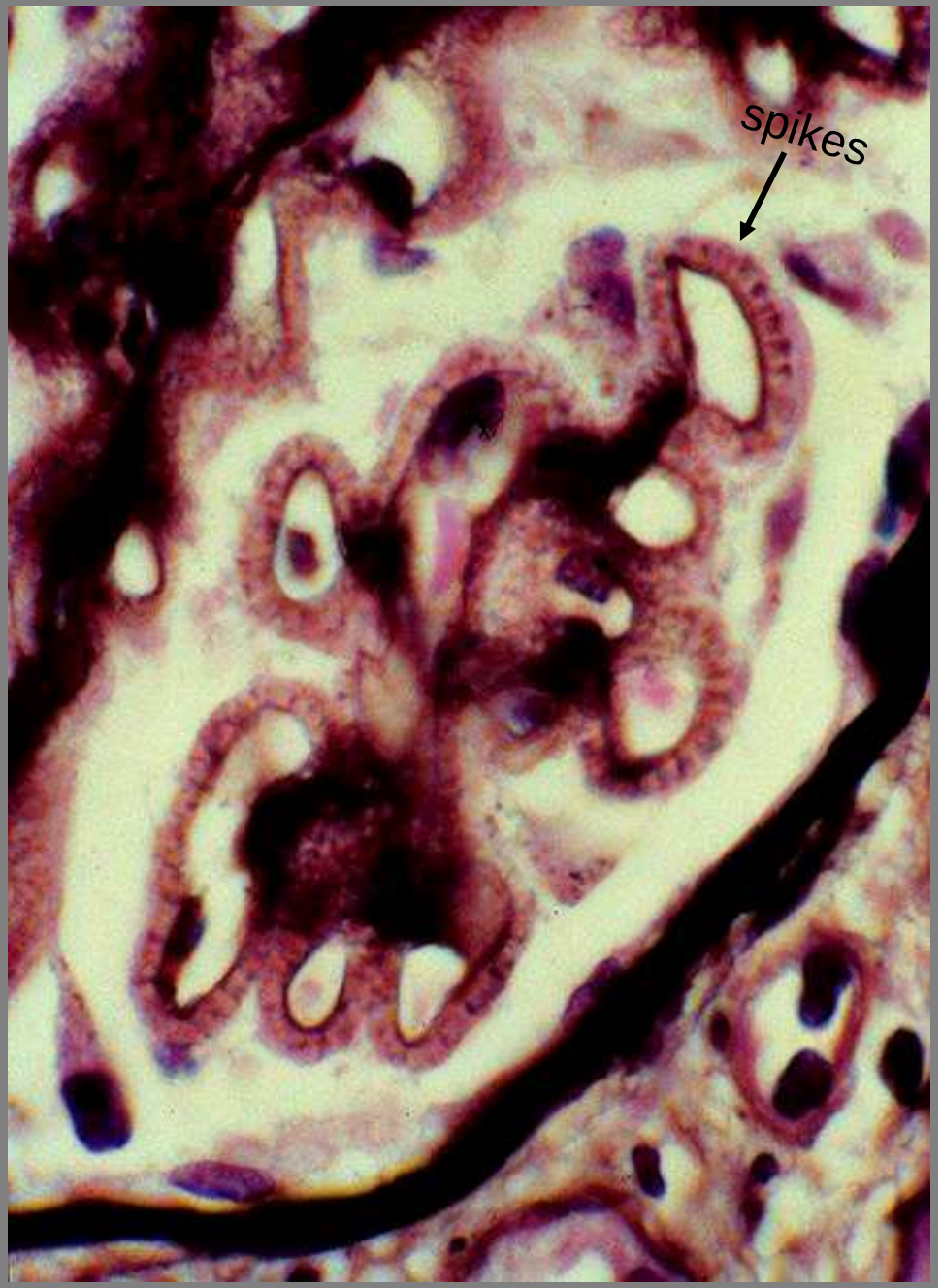


membranous glomerulonephritis




spikes

Membranous glomerulonephritis

- **poor prognosis –**
  only nephrotic proteinuria

  deBuf-Vereijken, Am.J.Kidn.Dis.(2005) 46:1012

- **predictors of progression**
  - ***male gender***
  - ***age***
  - ***nephrotic range proteinuria***
  - ***IgG/α1 microglobulin ratio in urine***
  - ***focal segmental lesions***

  Lai,Kidn.Internat.(2007) 71:841

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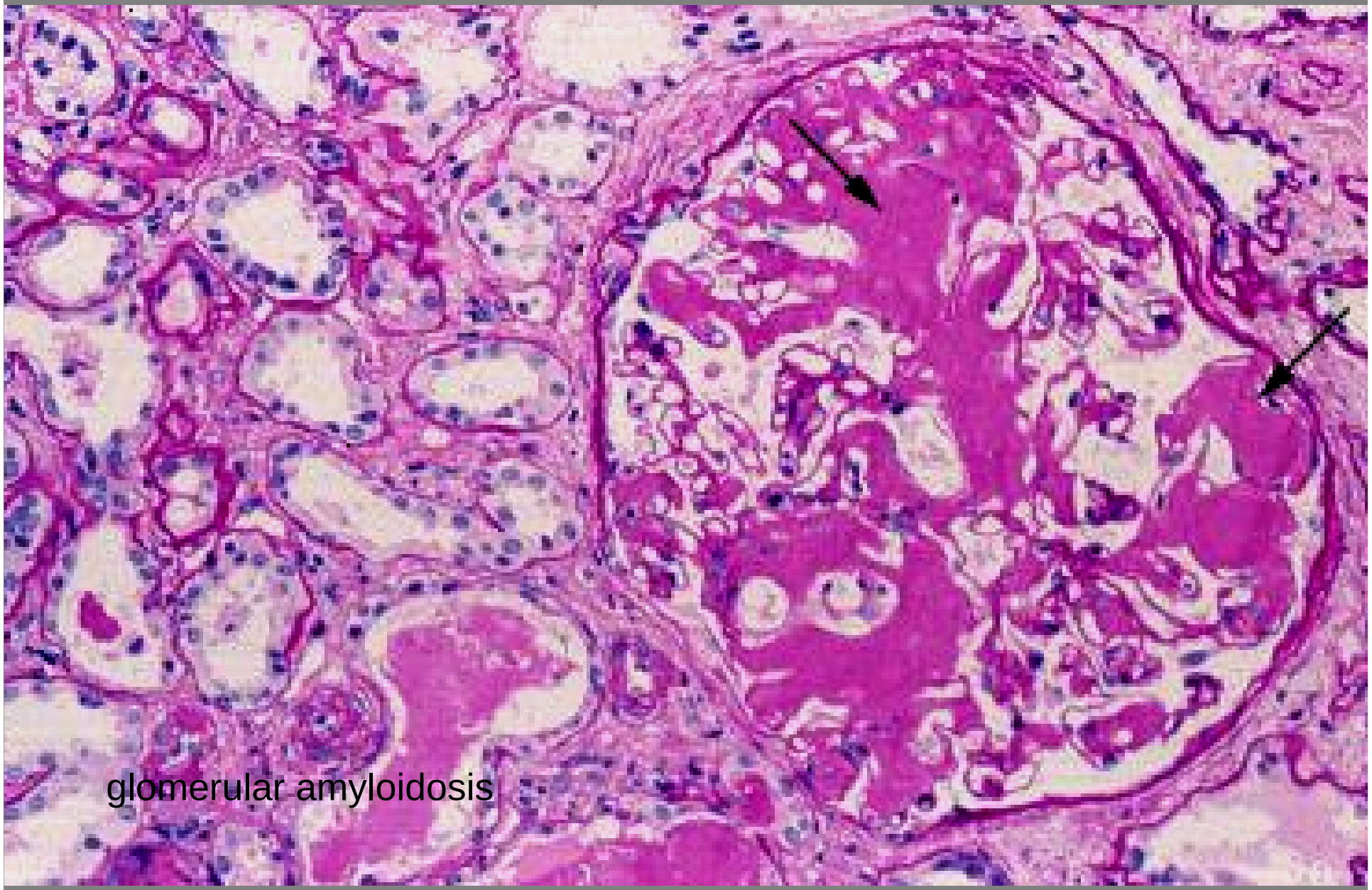
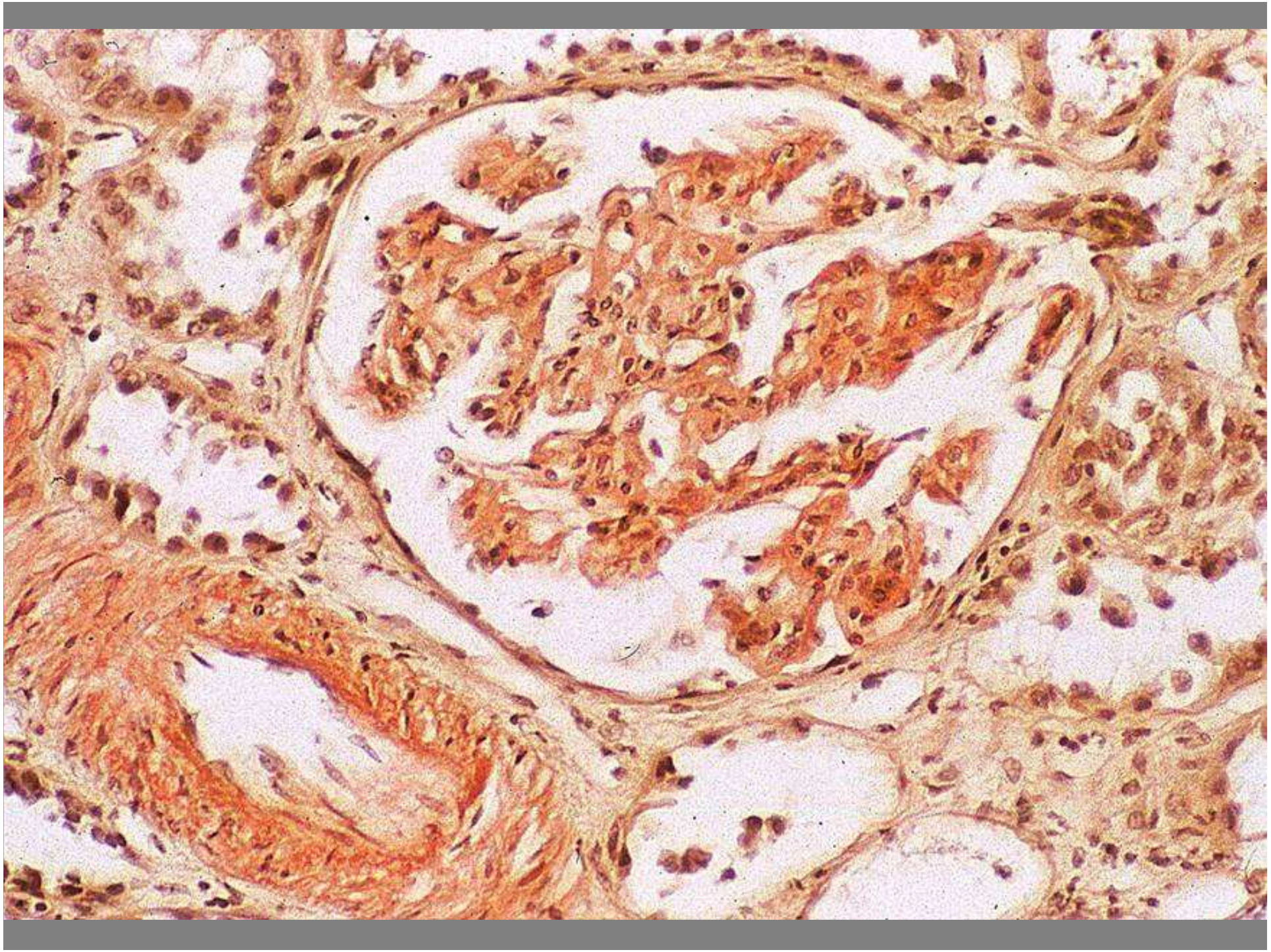
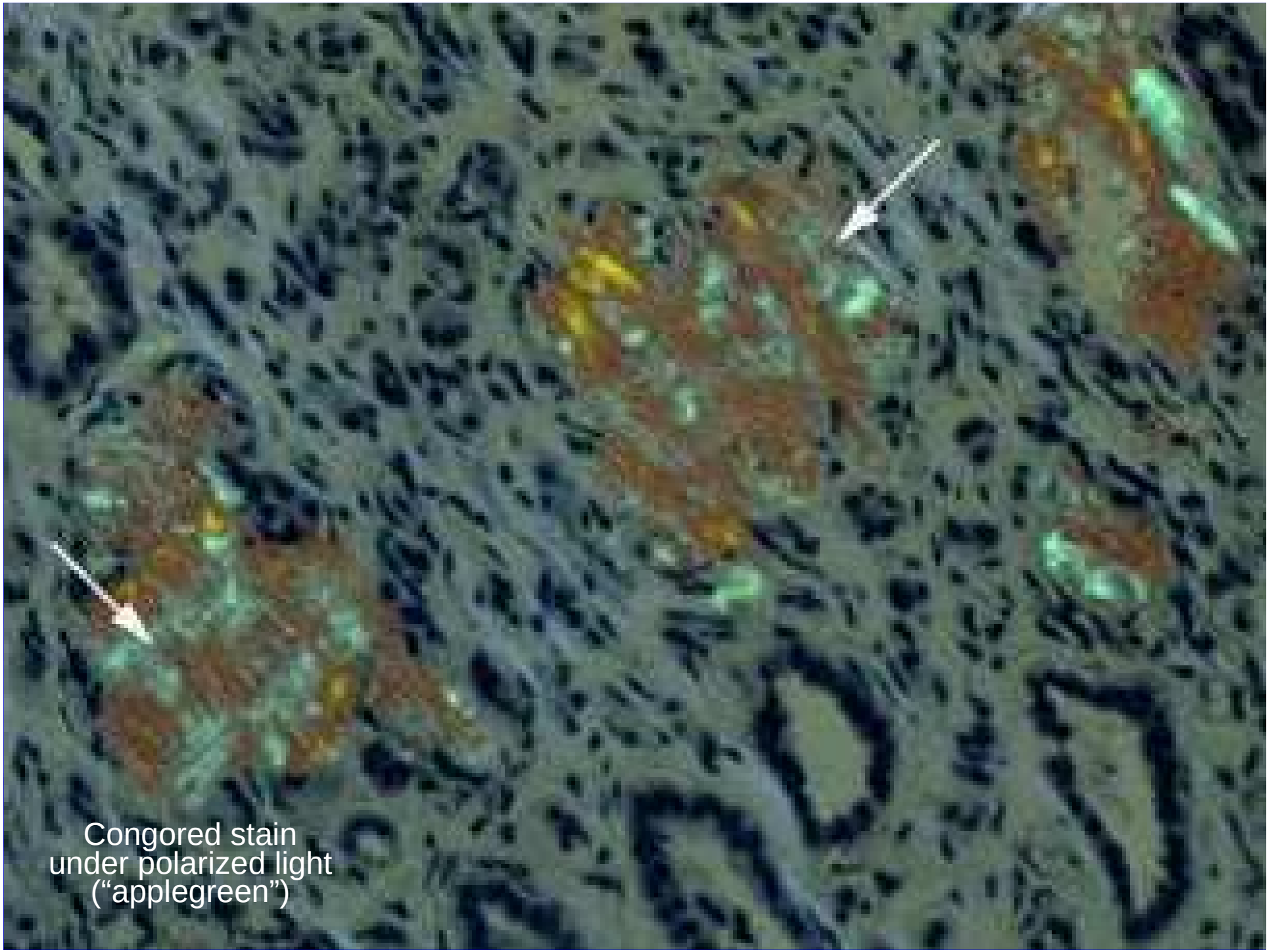
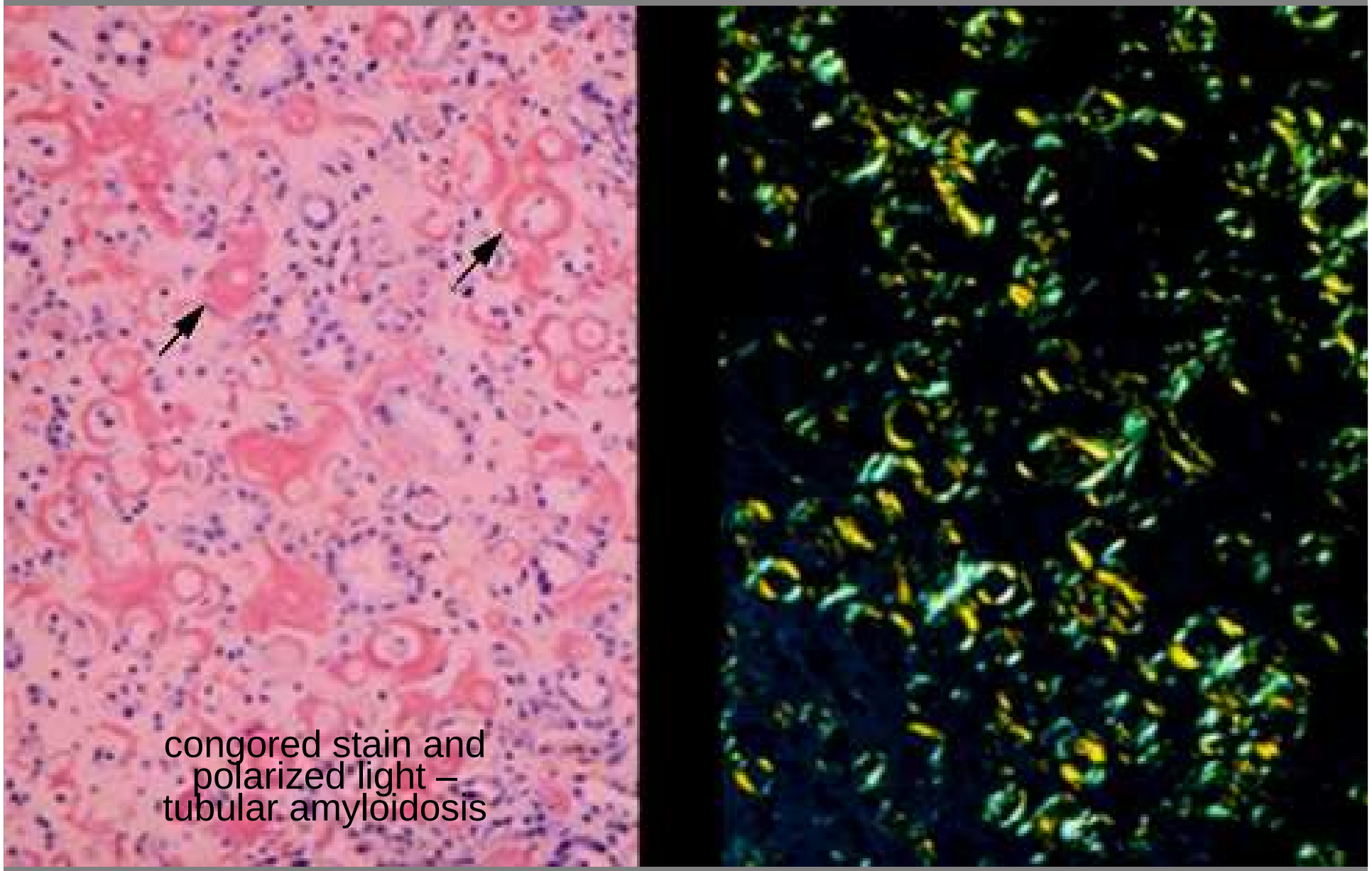
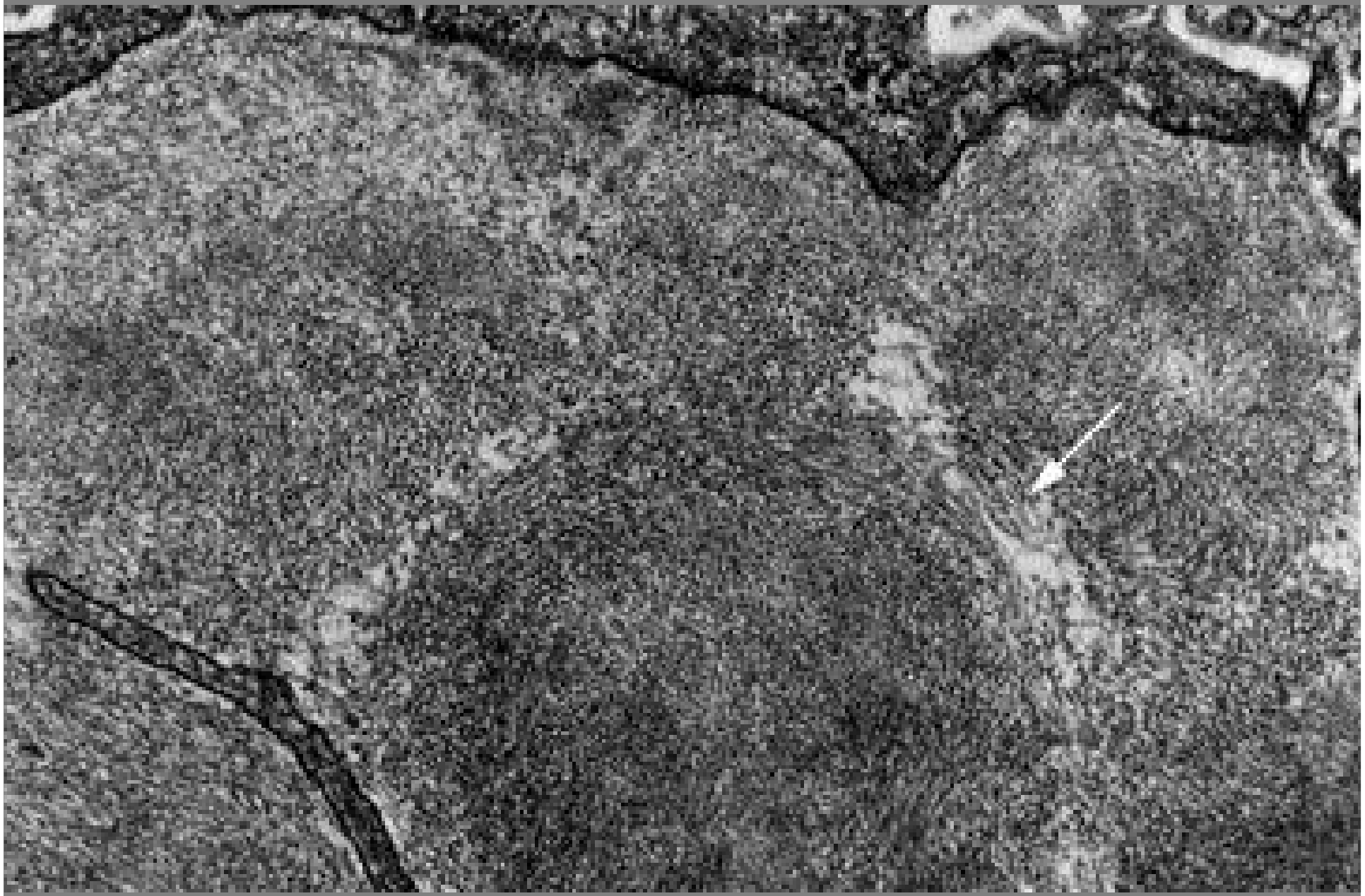

glomerular amyloidosis




Congored stain
under polarized light
(“applegreen”)



congored stain and
polarized light –
tubular amyloidosis



mesangial amyloid deposits
electron microscopy

Amyloidoses

- **acquired forms :**

  # AA amyloidosis – chronic infection or inflammation

  # AL amyloidosis – plasma cell dyscrasia
  (κ/λ 1:3 vs 3:2 in controls;
  usually the rare λVI and κI subgroups)

- **hereditary forms (with renal involvement) :**

  transthyretin (binding thyroxin and retinol binding protein),
  apo-Lipoprotein A I,II ,
  lysozym,
  fibrinogen A α,

Lachmann, New Engl.J.Med.(2002) 346:1786

Causes of (secondary) AA amyloidosis

- ***rheumatoid arthritis or juvenile RA 56 %***
- ***chronic infection 14 %***
- ***ankylosing spondylitis 8%***
- ***psoriatic arthritis 5 %***
- ***familial mediterranian fever 3 %***
- ***Crohn‘s disease 3 %***
- ***others 11 %***

Gorevic, update patient information Oct 2007,

misclassification of hereditary amyloidoses

- *350 patients with systemic amyloidosis diagnosed as AL type and had no known family history (only 85% of AL amyloidoses have monoclonal proteins in serum or urin, therefore confirmation by biopsy indispensable)*

- *amyloidogenic mutations in 34/350 patients (9,7% !!) mostly fibrinogen A α or transthyretin*

- *failure to make the diagnosis of hereditary amyloidosis explained by fortuitous combination of low grade monoclonal gammopathy in 8/34 patients (24%!!)*
  ► *erroneous therapy : cytotoxic treatment instead of liver transplantation*

Lachmann, New Engl.J.Med.(2002) 346:1786

alternative possibilities biopsy (less invasive?)

- **abdominal fat**
- **rectal mucosa**
- **bone marrow**

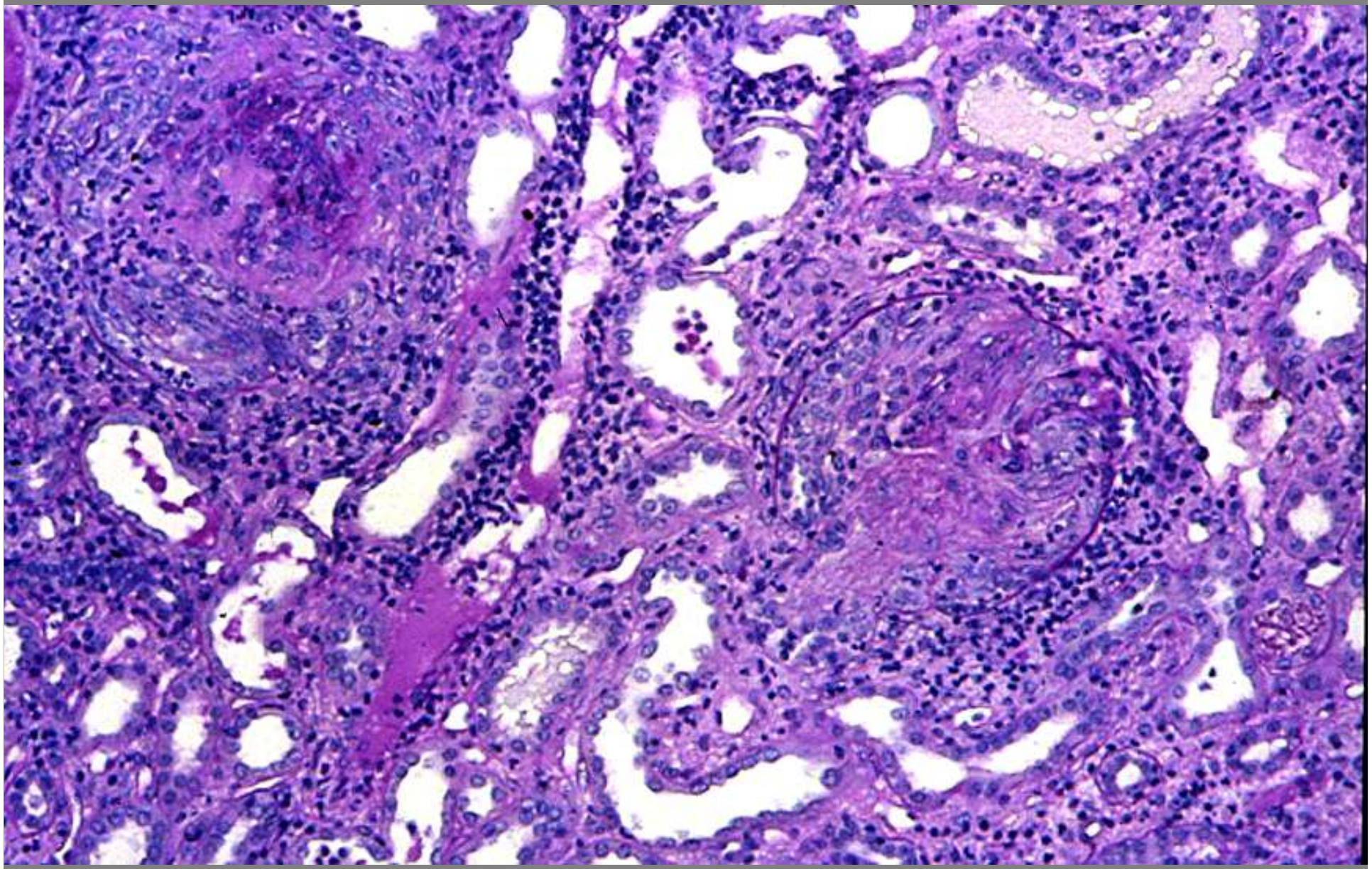
- **renal biopsy :**

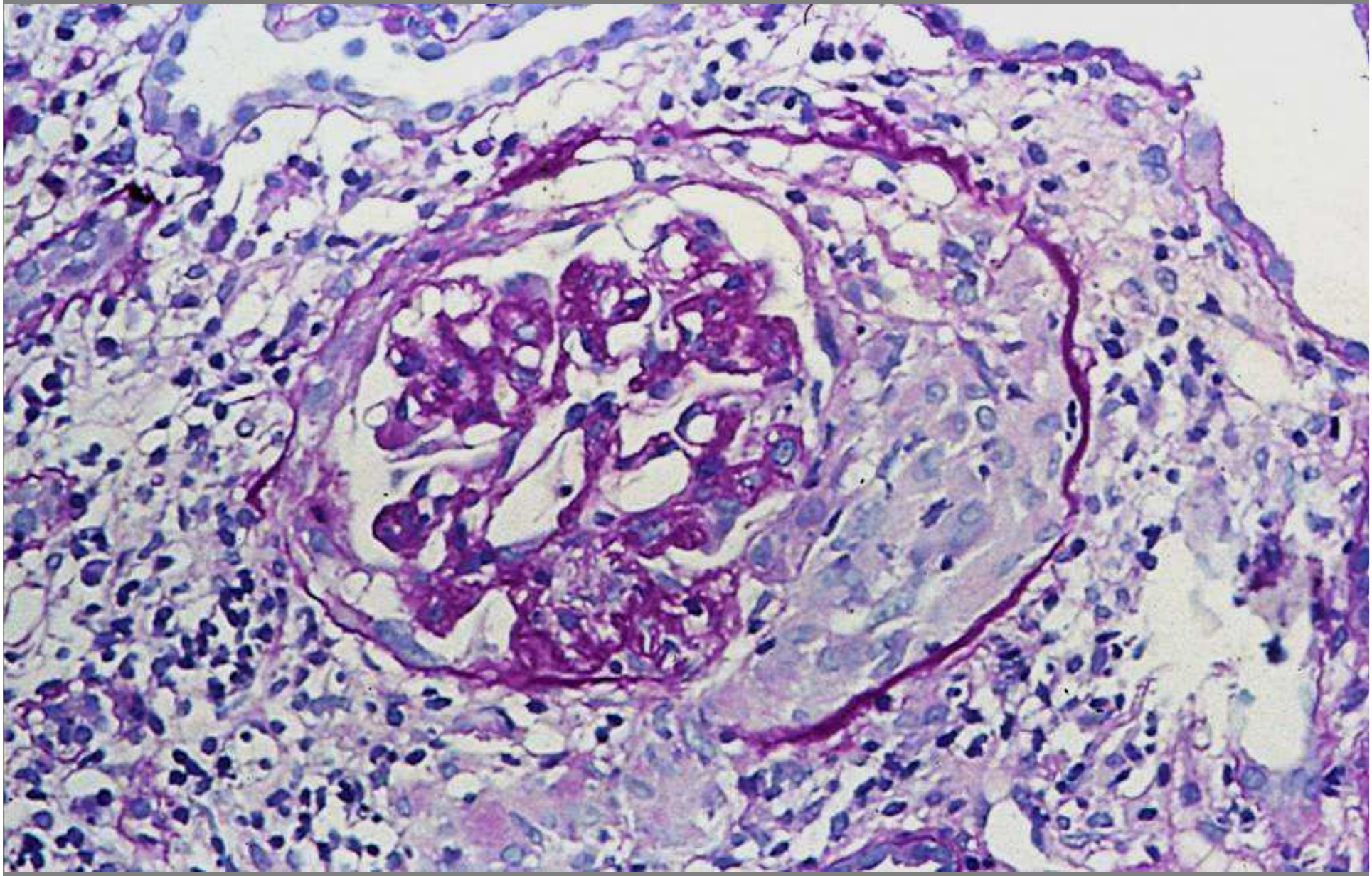
  vascular amyloidosis →

  vessel contraction↓ → bleeding risk↑

For which nephrologic syndromes should renal biopsy be considered :

- asymptomatic hematuria
  (dysmorphic erythrocytes,
  erythrocyte casts) ± proteinuria
- nephrotic syndrome
- acute nephritic syndrome
- rapidly progressive glomerulonephritis !!!
  (nephrological emergency)
- (chronic renal failure CKD)
- (acute renal failure AKI)
- renal transplantation





Differential diagnosis of rapidly progressive glomerulonephritis (RPGN)

immunecomplex-GN	**postinfectious**
	Schönlein Henoch purpura
	SLE (lupus)
antibasalmembrane Ab GN	**isolated GN**
	Goodpasture syndrome (with hemoptyses)
	combination with pauciimmune GN
pauciimmune RPGN	**Wegener's granulomatosis**
	microscopic polyangiitis

differential diagnosis:
HUS (EHEC, idiopathic), malignant hypertension, scleroderma-crisis
Cave: combination HUS plus glomerulonephritis

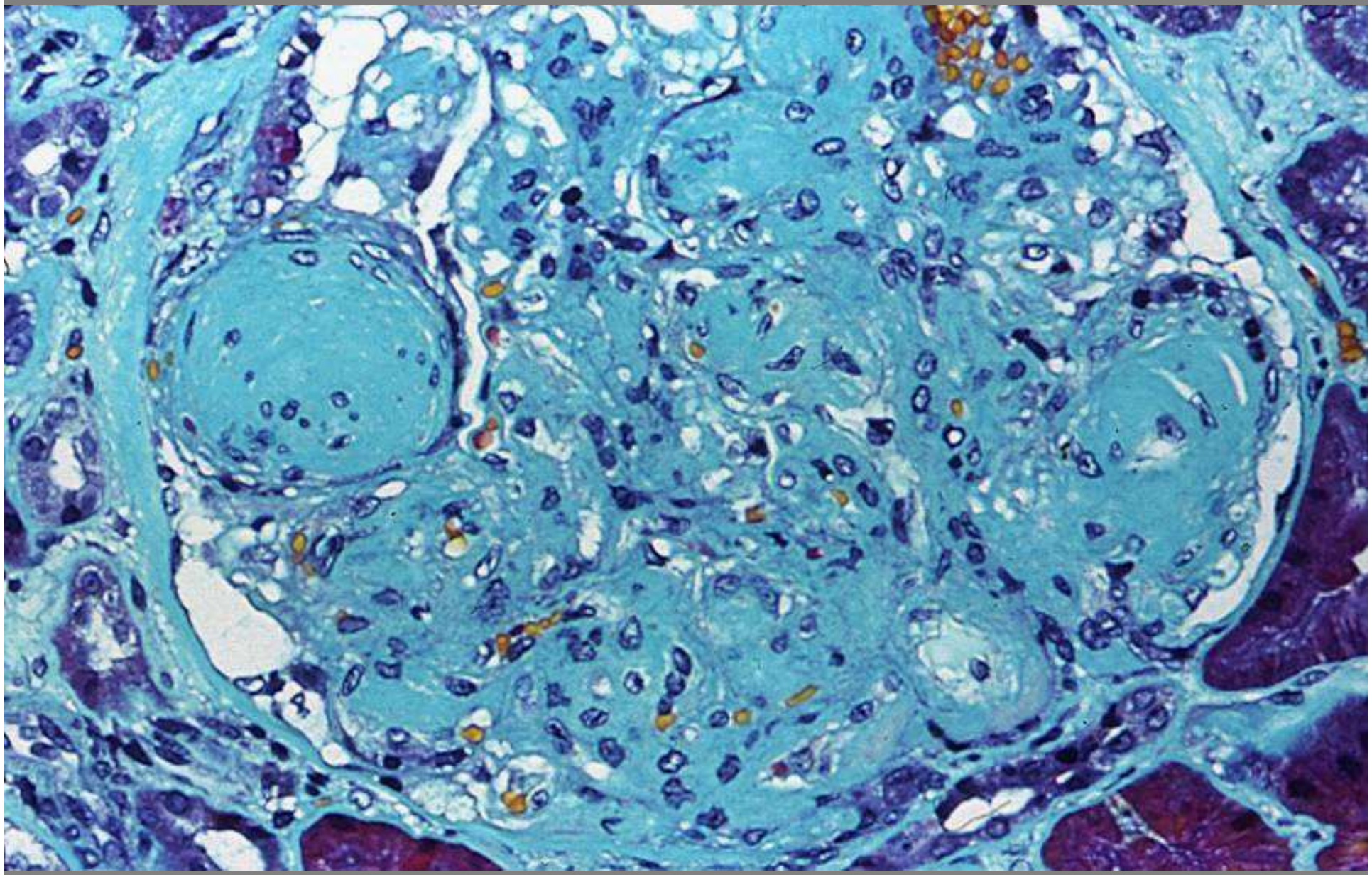
Renal biopsy in diabetes

 # *no* biopsy if :

- *typical evolution of renal disease*
- *accompanied by retinopathy (not reliable in type 2)*

 # *consider* biopsy if :

- *atypically early onset of renal abnormalities (< 5-10 years in type 1 diabetes)*
- *dysmorphic erythrocytes/erythrocyte casts*
- *rapid deterioration of renal function*
- *increased S-creatinine without urinary abnormalities or rapid increase of S-creatinine*
- *extremely high proteinuria (~7g/day) or rapid increase in proteinuria*



Histologic findings in patients with type 2 diabetes and abnormal renal findings

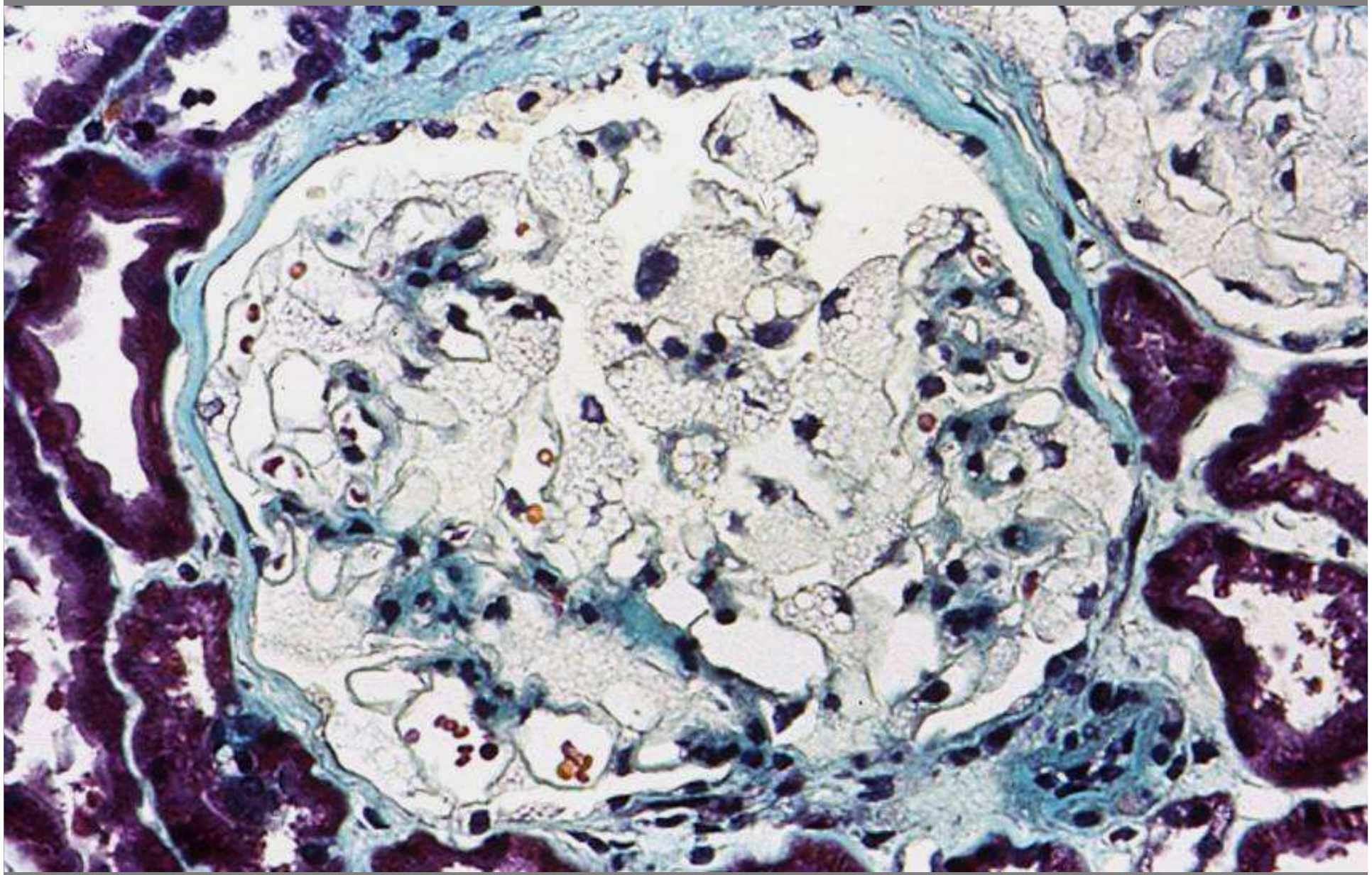
303 biopsies

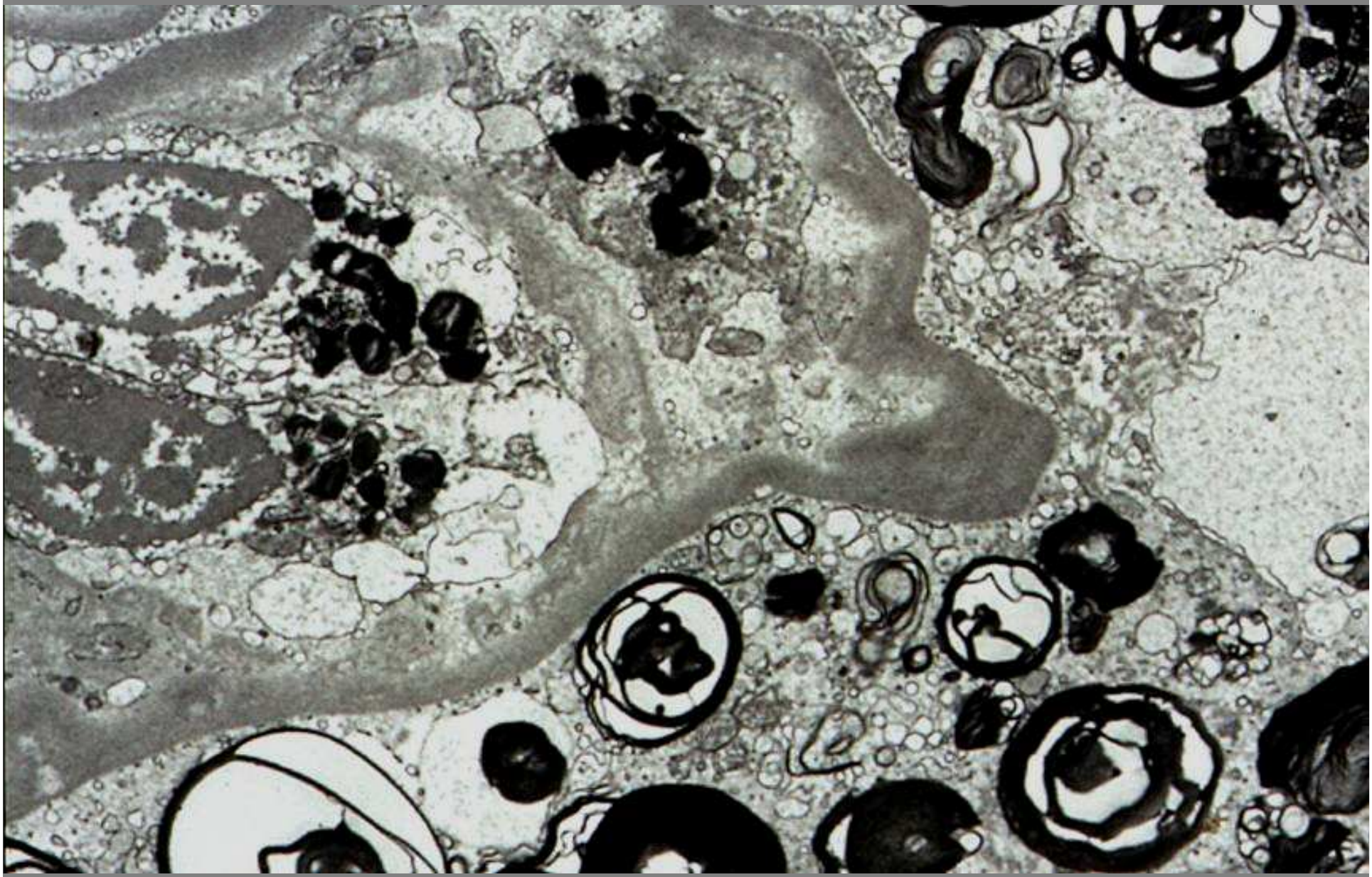
- **diabetic glomerulosclerosis** **51%**
- **atherosclerotic/ischemic lesions** **16%**
- **primary nephropathy** *(isolated or superimposed)* **33%**

Mazzuccho, AJKD (2002) 38: 713

For which nephrologic syndromes should renal biopsy be considered :

- **asymptomatic hematuria**
  (dysmorphic erythrocytes,
  erythrocyte casts) ± proteinuria
- **nephrotic syndrome**
- **acute nephritic syndrome**
- **rapidly progressive glomerulonephritis !!!**
  (nephrological emergency)
- **(chronic renal failure CKD)** ***exceptions:*** *Fabry, Balkan nephritis…*
- **(acute renal failure AKI)**
- **renal transplantation**





For which nephrologic syndromes should renal biopsy be considered :

- asymptomatic Hematuria
  (dysmorphic erythrocytes,
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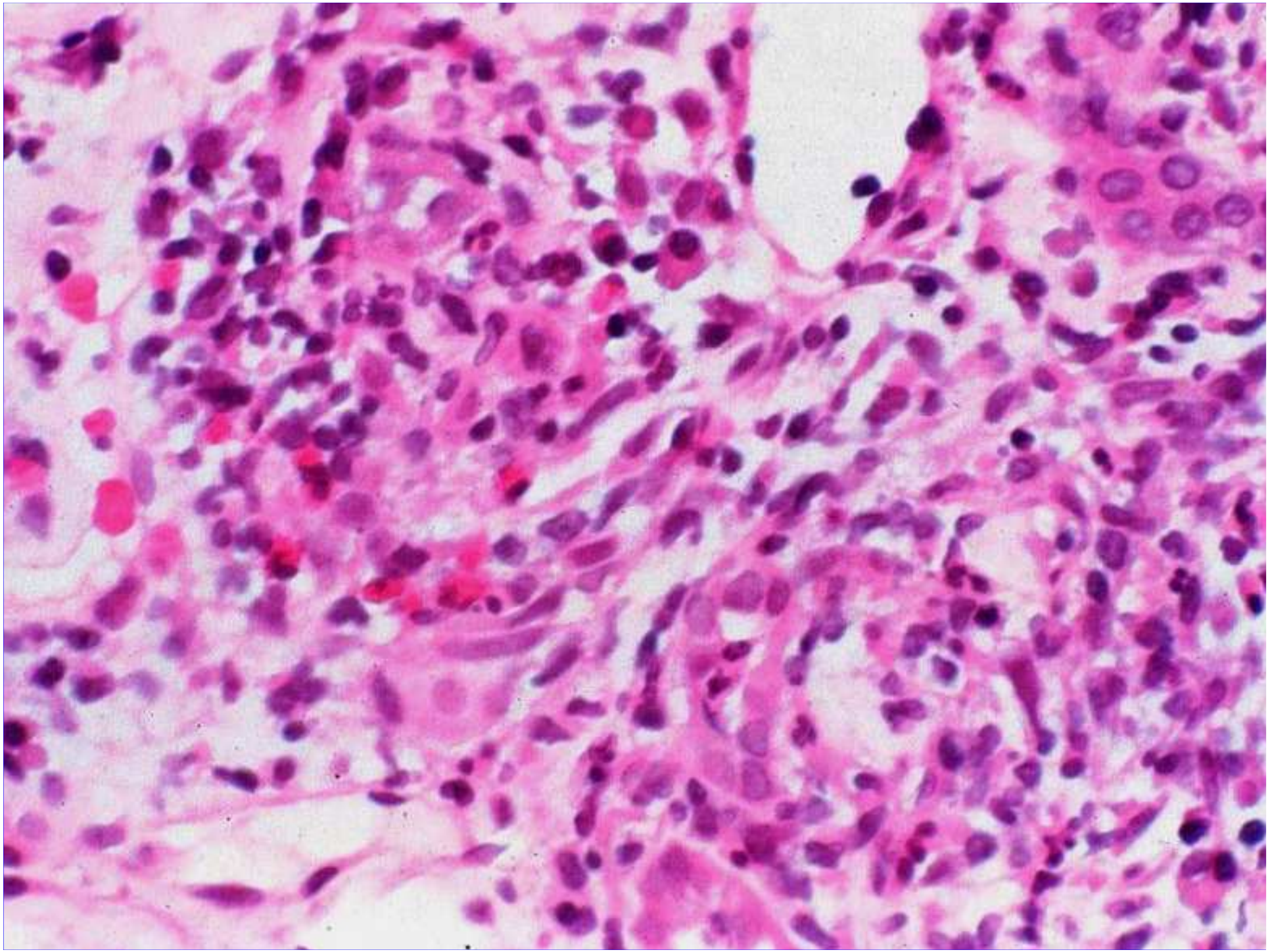
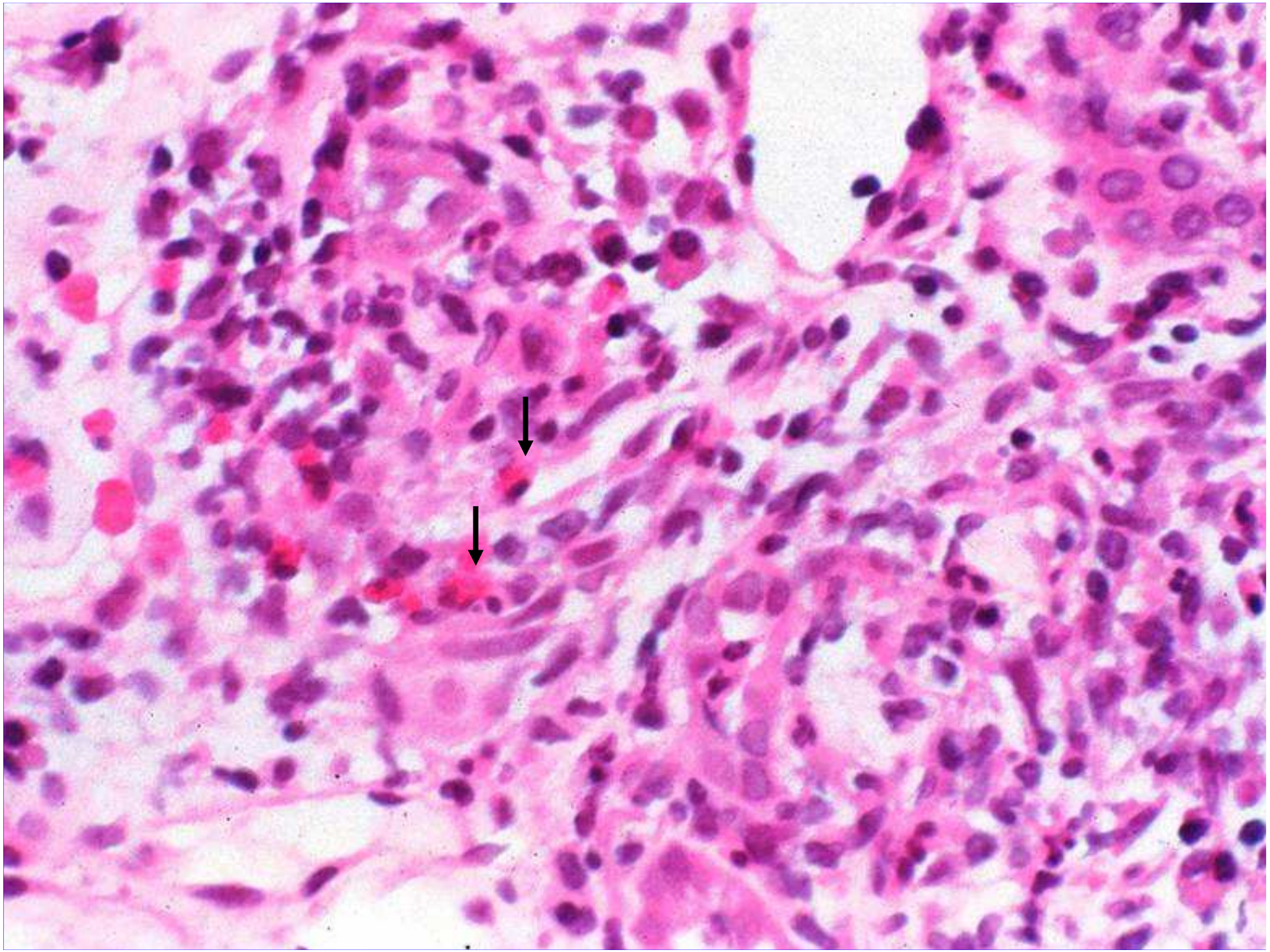
- nephrotic syndrome
- acute nephritic syndrome
- rapidly progressive glomerulonephritis !!!
  (nephrological emergency)
- (chronic renal failure CKD)
- (acute renal failure AKI)
- renal transplantation

Reasons to consider renal biopsy in patients with acute renal failure (AKI)

- *progressive loss of renal function*
- *persisting oliguria*
- *(absence of obvious causes of ARF)*
- *pronounced proteinuria/hematuria*
  (consider Hanta and exclude vasculitis !!!)
- *pronounced hypertension*
  (exclude HUS, malignant hypertension, scleroderma crisis)
- *history of glomerulonephritis*
- *suspicion of drug allergy or*
  tubular obstruction by drug crystalluria





No value of urinary eosinophils to diagnose interstitial nephritis

- *62/534 quantitative urinary eosinophil counts positive*
- *acute interstitial nephritis diagnosed in 13 patients during this period*

- *in 6 patients urine negative for eosinophils*
- *in others not performed*

⟹ **sensitivity 25%**

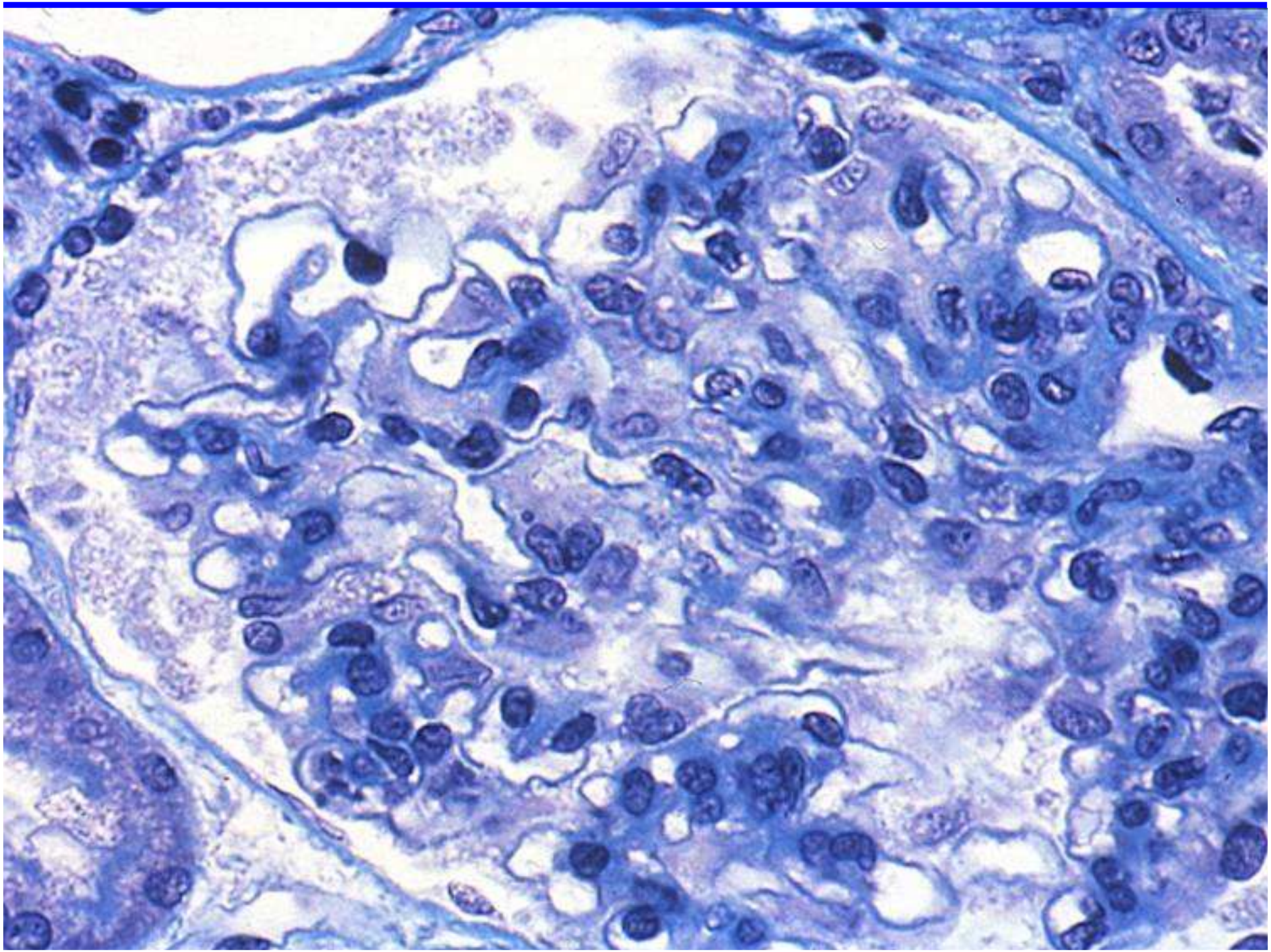
predictive value 3%

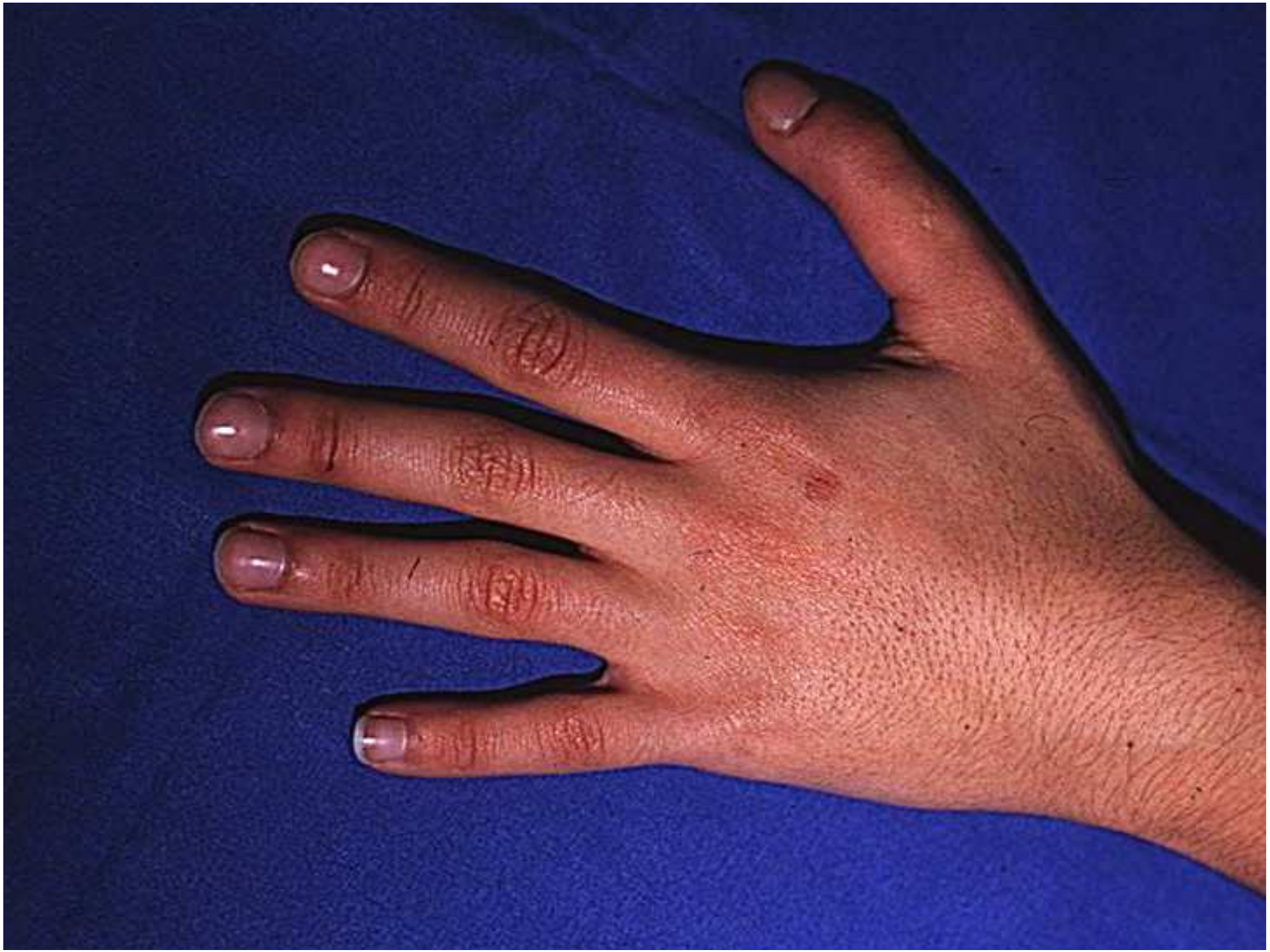
Fletcher,New Engl.J.Med.(2008) 358:1760

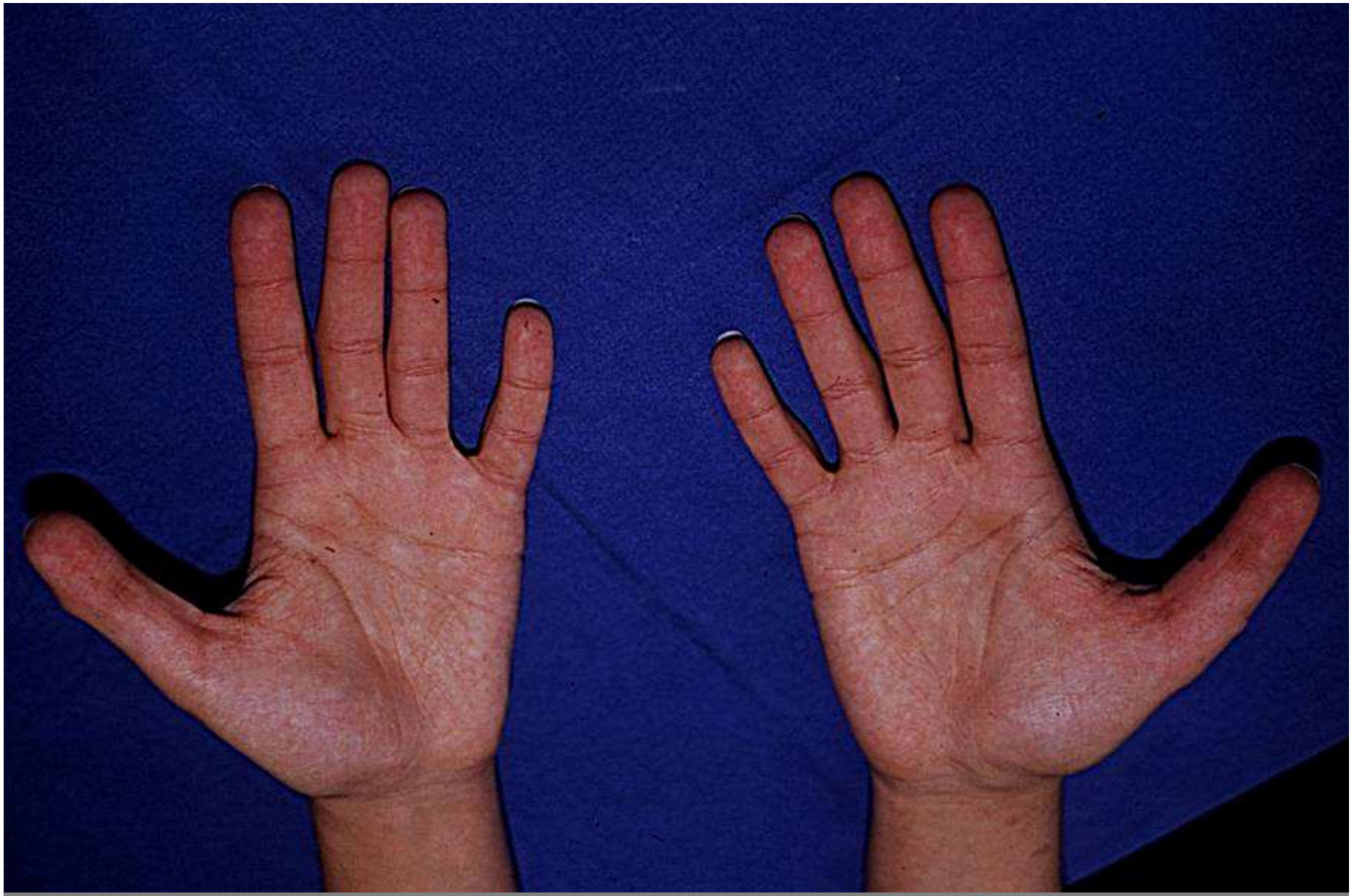
Thank you for your attention

NPHS2 mutations :

- *familial and sporadic steroid-resistant focal segmental glomerulosclerosis*
  - *- pediatric age*
  - *- late onset*
- *modifier of progression in some patients with primary glomerulonephritis*







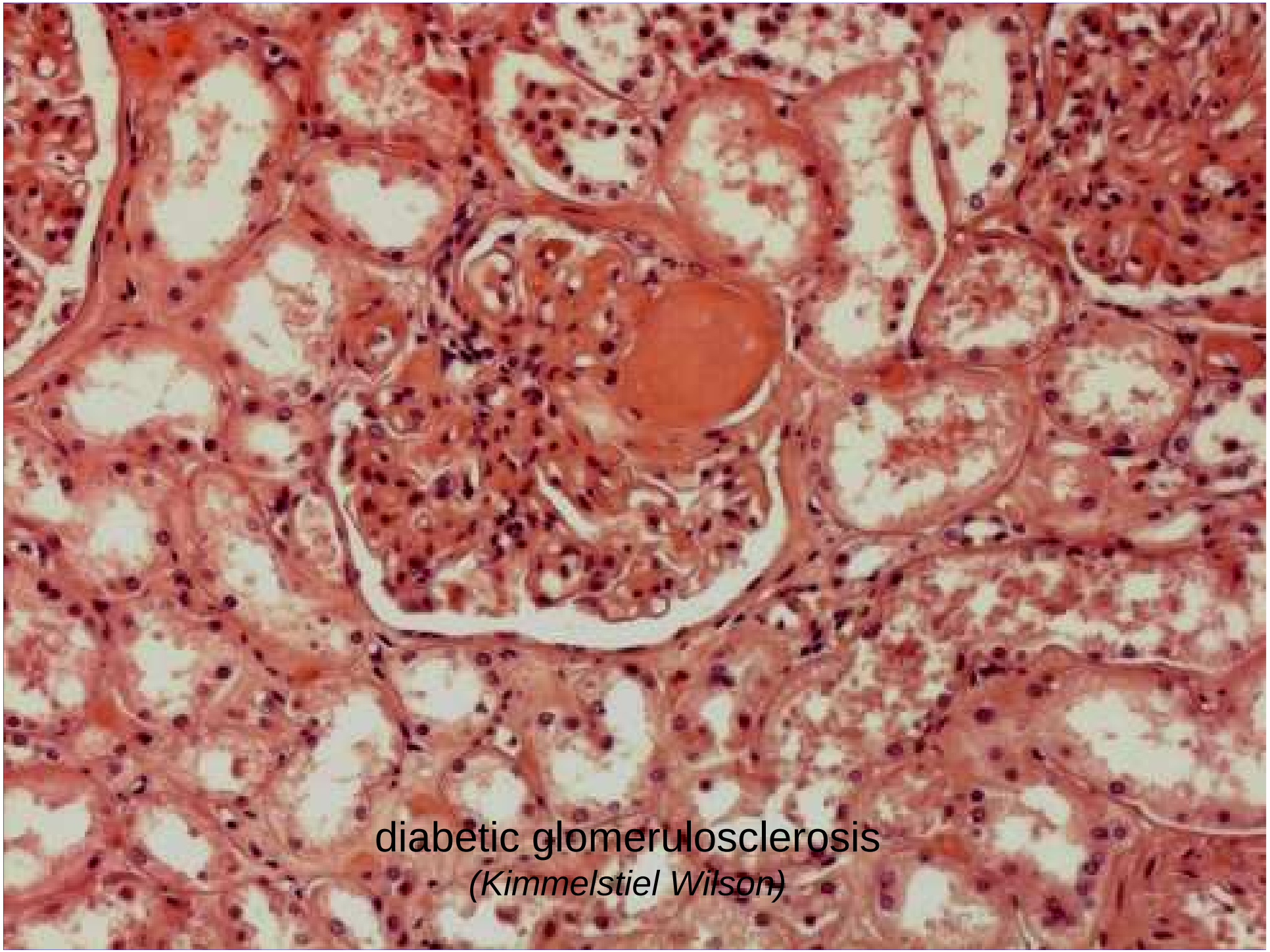
nail-patella syndrome:mutation of transcription factor **LMX1B** controlling expression of COL4A3,COL4A4,nephrin,podocin …

- characteristic lesions of glomerular basement membrane
  - rarefications, lucent areas (“moth eaten”)
  - with cross-striated collagen fbrils (type III)

Del Poso (1970) Am J Pathol 54: 845

- disease does not recur in transplant
- kidney of donors with nail patella syndrome suitable for transplantation

Vega (1997) Nephrol.Dial.Transplant. 12: 1742



diabetic glomerulosclerosis
(Kimmelstiel Wilson)