Genetics of Renal Disease

Ιωάννης Γ. Γριβέας
Διευθυντής Νεφρολογικού Τμήματος
417 «ΝΙΜΤΣ»

Topics

- Quantitatively important hereditary disorders
  - ADPKD
  - Alport's syndrome/hereditary nephritis
- Novel pathogenetic mechanisms
  - FSGS, nephrotic syndrome, proteinuria
- Novel molecular treatment
  - Fabry's disease
- Tubular disorders

ADPKD genetics

- Autosomal dominant
- 2 genes
- PKD1(encodes polycystin-1)
  - On chromosome 16p13.3
  - Accounts for 85% of disease
- PKD2 (encodes polycystin-2)
  - Chromosome 4q21-22
  - Accounts for 85% of disease
- PKD3? Probably not
- Maybe more PKD2: ascertainment bias

Genetic Testing in PKD

- Mutational analysis ("direct") versus linkage analysis ("indirect")
- Clinical utility
  - Kidney transplant donor evaluation (does potential living related donor have presymptomatic PKD?)
  - Earlier treatment????
    - ACE-I, antioxidants, K+ supplementation....

Genetic diseases linked to structural abnormalities of proteins that are normally present in the glomerular basement membrane

- Type IV collagen (hematuric diseases)

Alport syndrome

Benign familial hematuria with thinning of the glomerular basement membrane

Familial hematuria with retinal arteriolar tortuosity and contractures

- Laminin β2

Pierson syndrome (a recently characterized laminin disease)

Genetic diseases linked to abnormal accumulation of non-glomerular-basement-membrane extracellular components

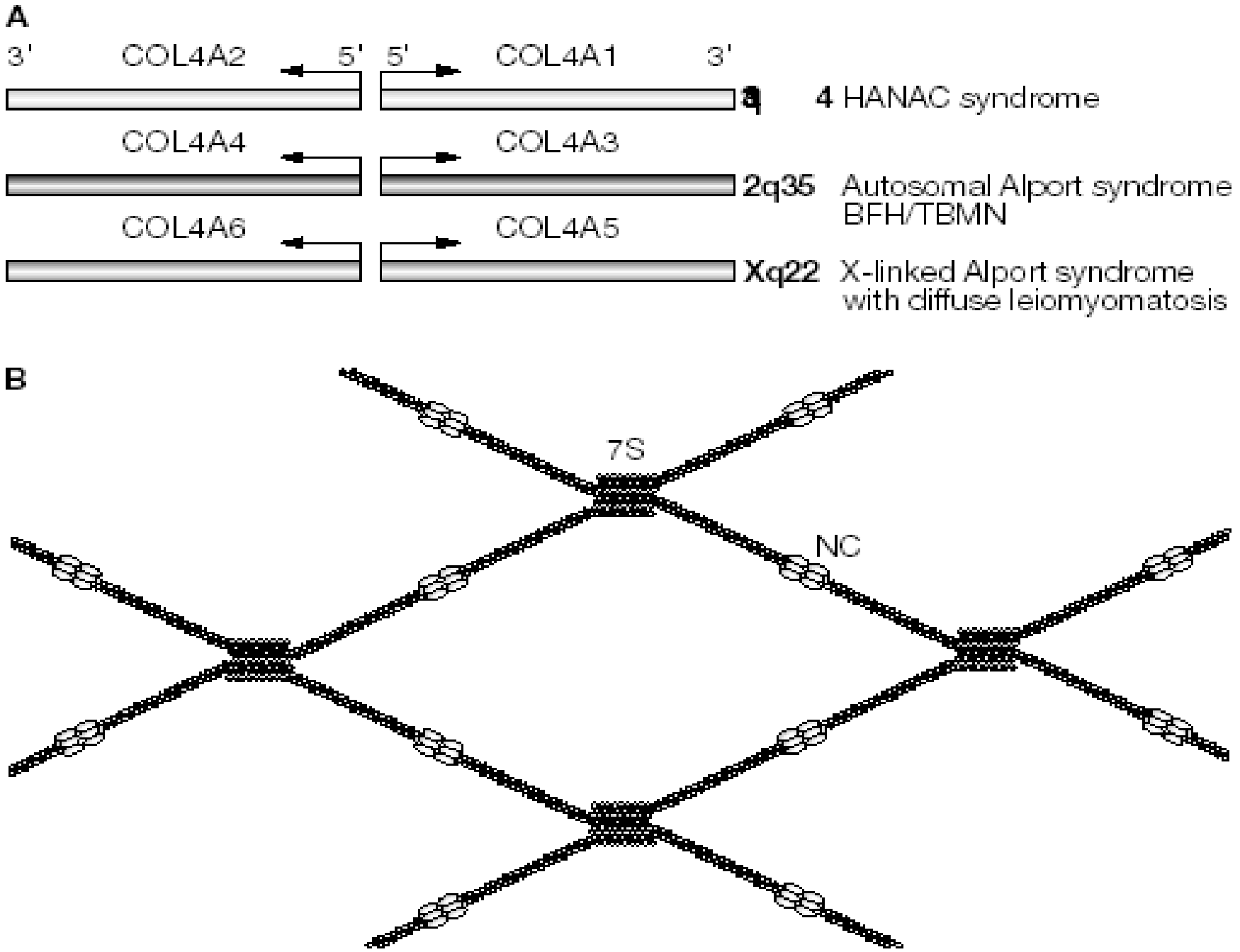
- Type III collagen

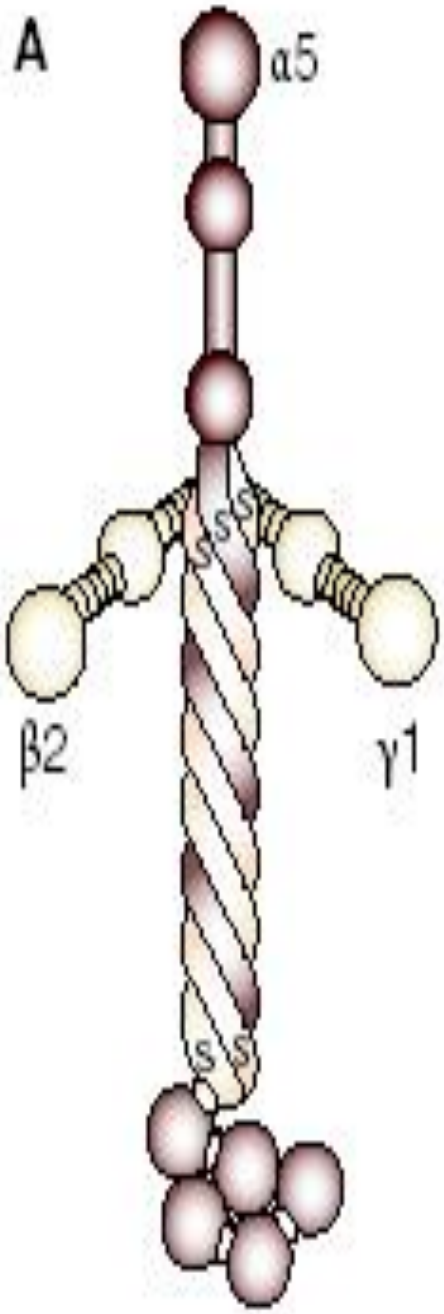
Nail–patella syndrome

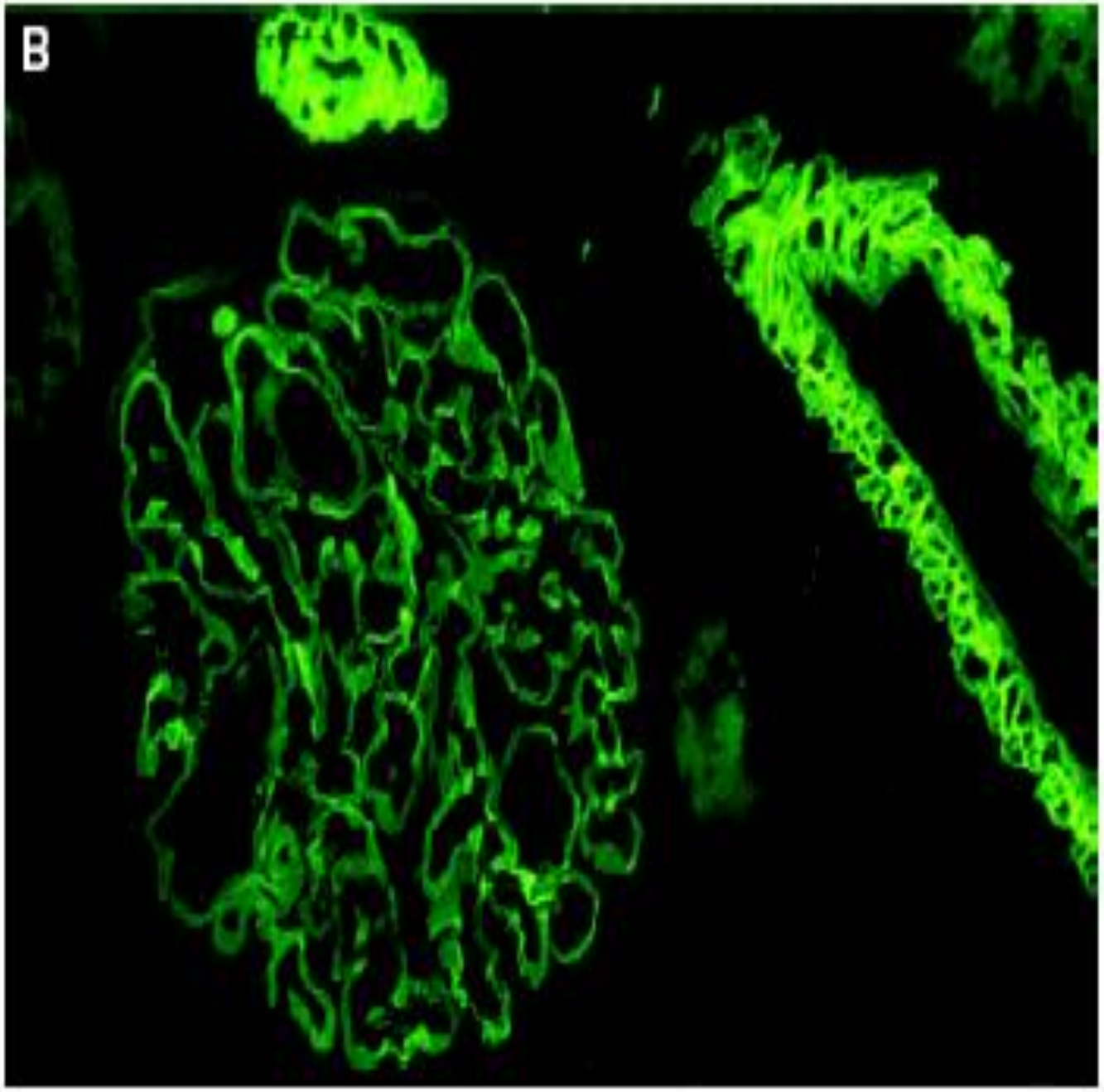
Idiopathic 'collagen type III glomerulopathy'

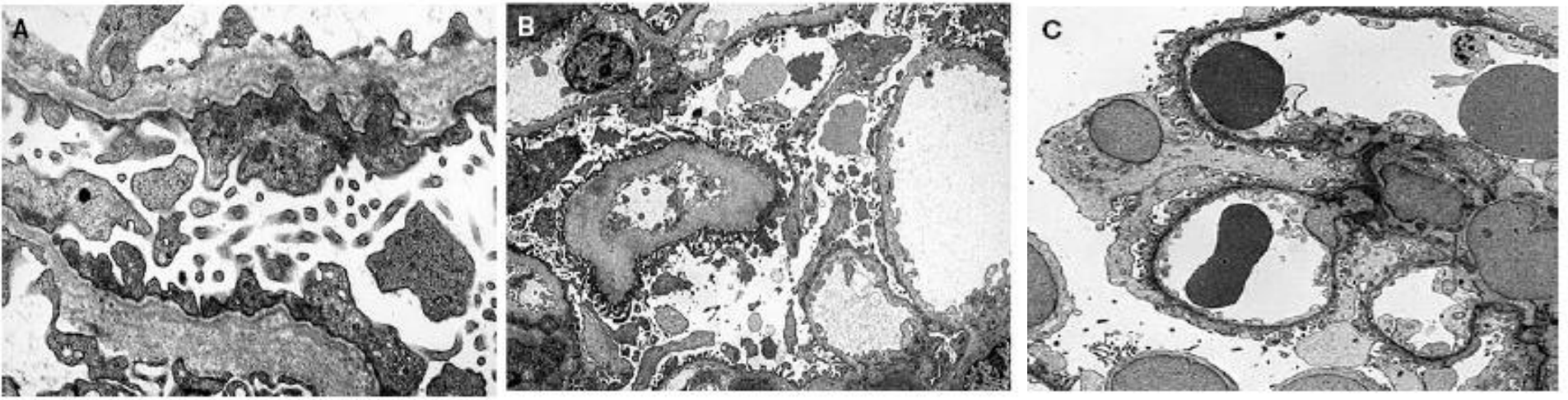
- Fibronectin

Fibronectin glomerulopathy


A
3'
COL4A2
5'
5'
COL4A1
3'
4 HANAC syndrome
COL4A4
COL4A3
2q35
Autosomal Alport syndrome
BFH/TBMN
COL4A6
COL4A5
Xq22
X-linked Alport syndrome
with diffuse leiomyomatosis
B
7S
NC


A
α5
β2
γ1
S
S
S
S
S
S


B



X-linked Dominant Alport Syndrome.

- a 'juvenile' type, characterized by a highly stereotypical course within each affected family and by the occurrence of ESRD in men at about 20 years of age; and
- a'nonprogressive' or 'adult' type in which ESRD develops at approximately 40 years of age and the disease course is much more variable

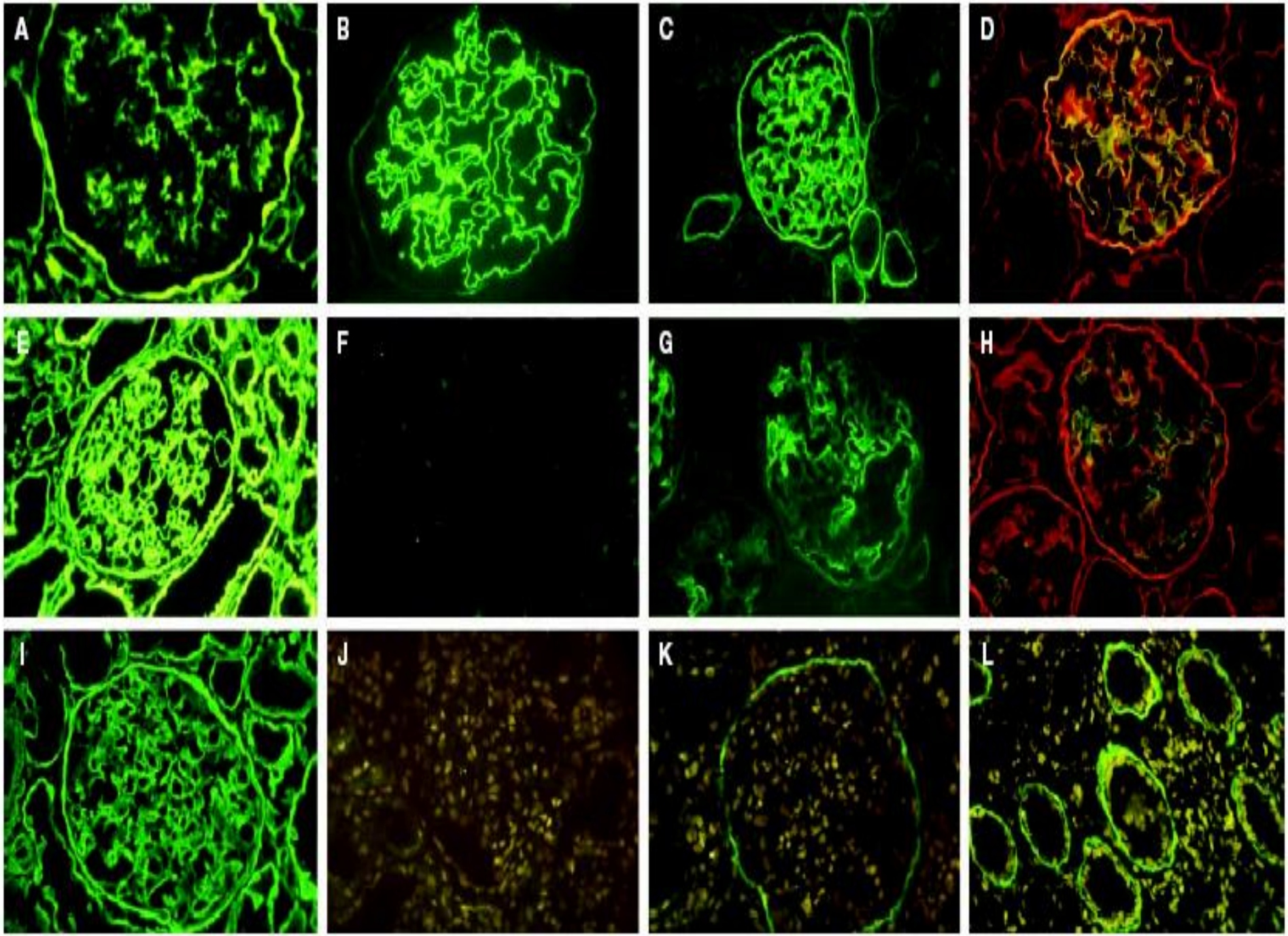
Atkin CL *et al.* (1988) Alport syndrome. In *Diseases of the Kidney,* edn 4, 617–641 (Eds Schrier RW and Gottschalk CW) Boston: Little, Brown, and Company

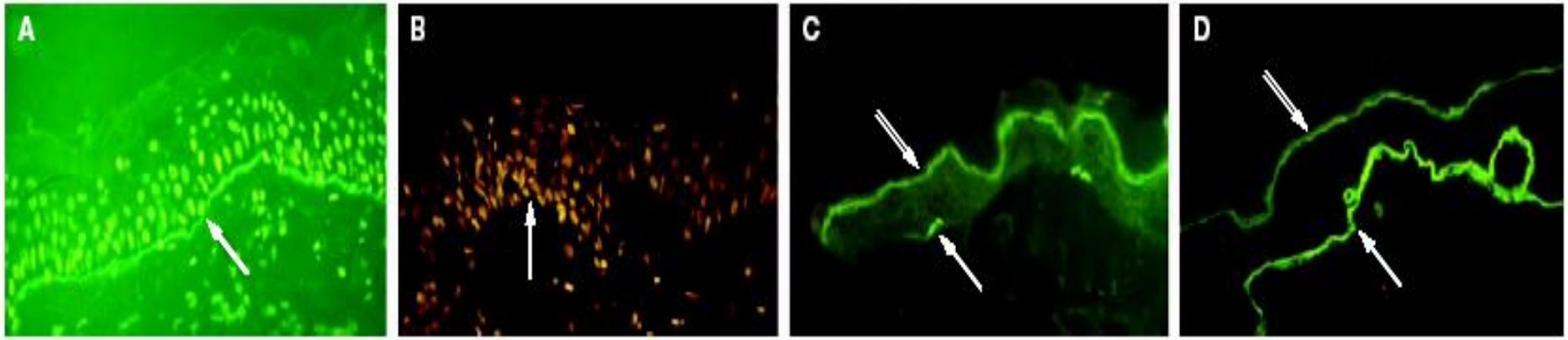
- The risk of developing ESRD before the age of 40 years is 12% in girls and women versus 90% in boys and men.
- The risk of progression in women seems to increase after the age of 60 years.
- Random X-chromosome inactivation might account for the variable clinical course of the disease in female patients.

Jais JP *et al.* (2003) X-Linked Alport syndrome: natural history and genotype-phenotype correlations in girls and women belonging to 195 families: a "European Community Alport Syndrome Concerted Action" study. *J Am Soc Nephrol* **14:** 2603–2610

- **Since the identification of COL4A5 more than 300 mutations have been reported in this gene…**

Myers JC *et al*. (1990) Molecular cloning of α5(IV) collagen and assignment of the gene to the region of the X chromosome containing the Alport syndrome locus. *Am J Hum Genet* **46:** 1024–1033


A
B
C
D
E
F
G
H
I
J
K
L



Autosomal Recessive Alport Syndrome.

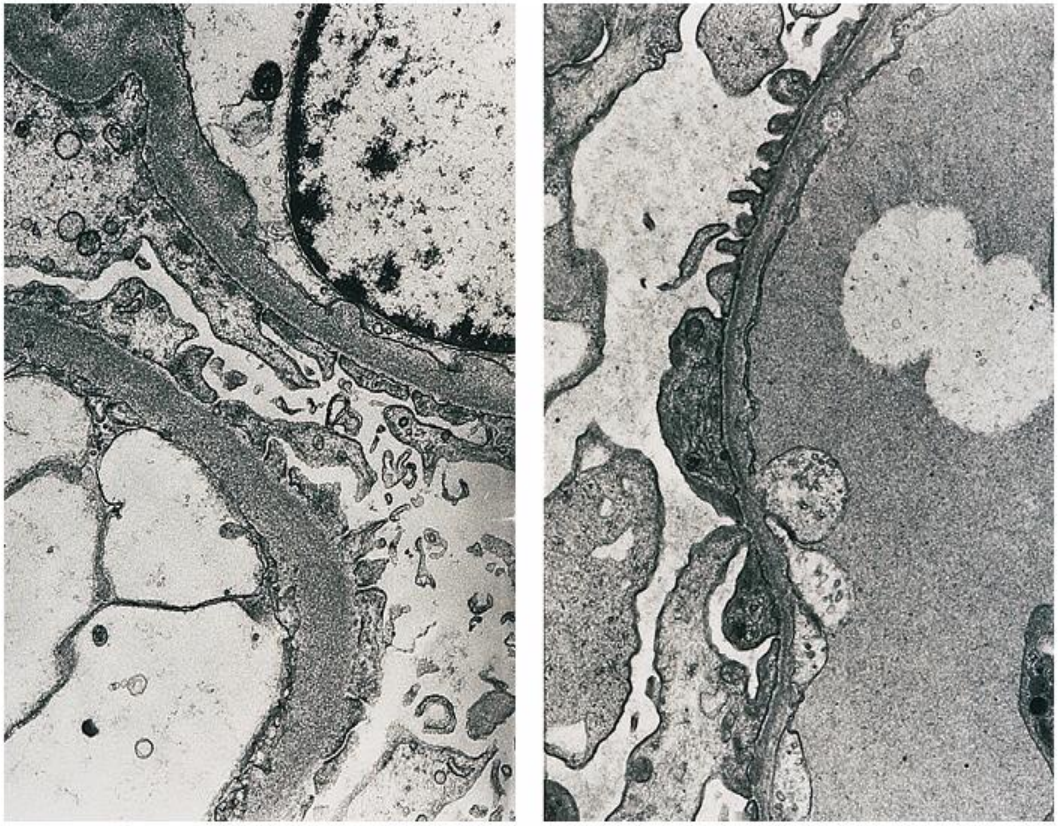
Usually severe; nephritis progresses to early-onset ESRD,

Hearing impairment affects the majority of patients, and ocular lesions may or may not be present.

Autosomal Dominant Alport Syndrome.

The clinical phenotype is variable and milder than that of the X-linked dominant form.

Progression to ESRD and hearing defect are not always seen and usually occur after 50 years of age; no ocular involvement has been reported.



By definition, no significant proteinuria, no progression to renal failure and no extrarenal symptoms are observed, and the prognosis is excellent.

Rogers PW *et al.* (1973) Familial benign essential hematuria. *Arch Intern Med* **131:** 257–262

A heterozygous COL4A4 mutation was detected in a large family.

Lemmink HH *et al.* (1996) Benign familial hematuria due to mutation of the type IV collagen α4 chain. *J Clin Invest* **98:** 1114–1118

…Spectrum of Phenotypes Associated With Heterozygous COL4A3 or COL4A4 Mutations…

Torra R *et al*. (2004) Collagen type IV (α3–α4) nephropathy from isolated haematuria to renal failure. *Nephrol Dial Transplant* **19:** 2429–2432

Πρόληψη

Η ανεύρεση της συγκεκριμένης μετάλλαξης του γονιδίου.

Frances Flinter and Kate Plant. Why are mutations in COL4A5 not detectable in all patients with Alport's syndrome? Nephrol Dial Transplant 1998,13:1348-1351

Προγεννητική διάγνωση

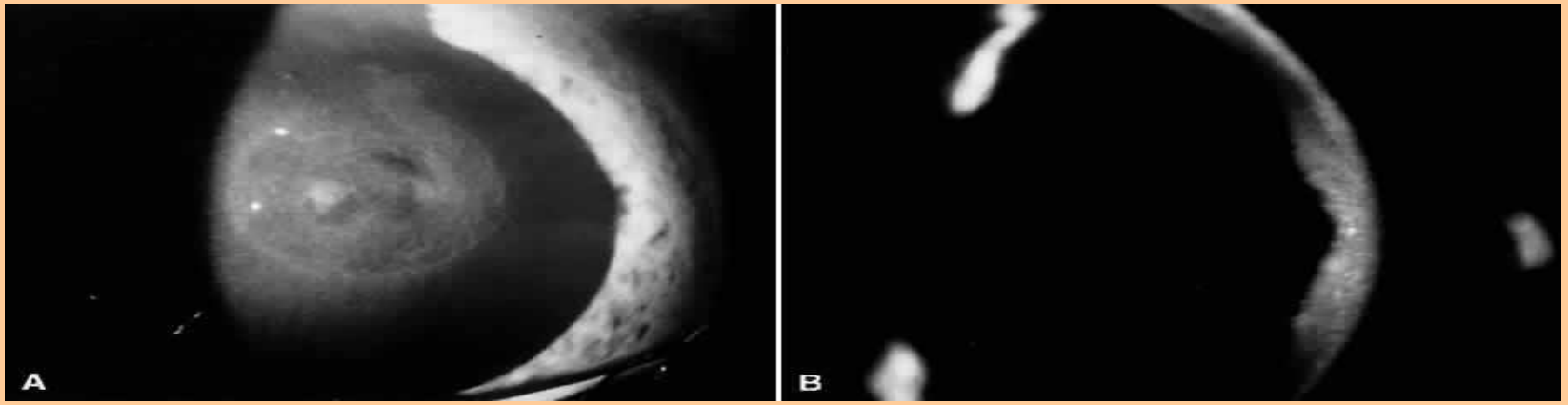
Pre-implantation diagnosis (Θαλασσαιμία)

…Γονιδιακή θεραπεία…

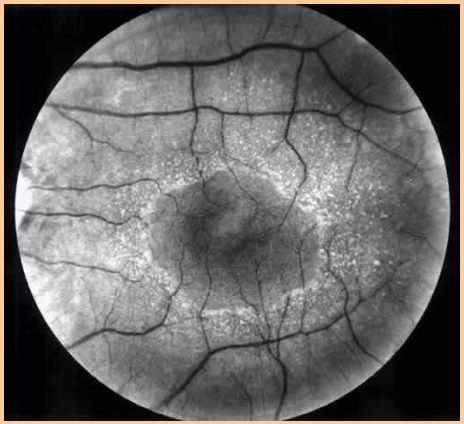
Karl Tryggvason , Pirkko Heikkila, Erna Petterson, Annika Tibeli and Paul Thorner. Can Alport syndrome be treated by gene therapy? Kidney Int 1997,51:1493-1499

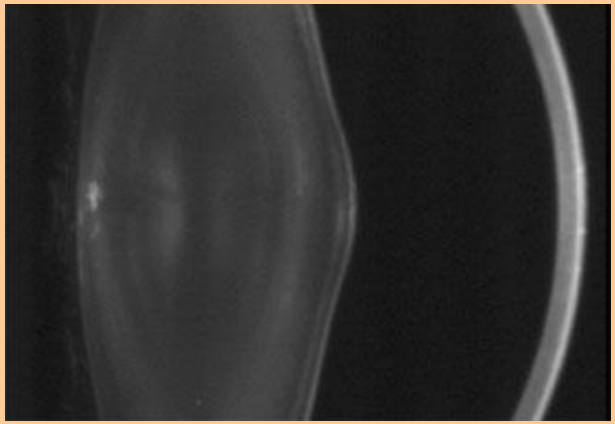
- Γενικά μέτρα αντιμετώπισης Χρονίας Νεφρικής Νόσου (άγνωστος ο ακριβής παθογενετικός μηχανισμός, TGF-β1).

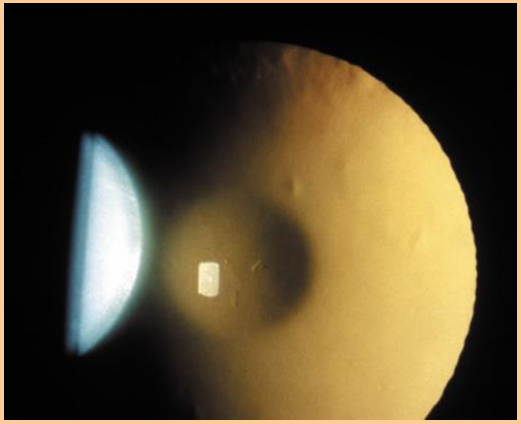
- Η βαρηκοία αντιμετωπίζεται με τη βοήθεια ακουστικών.



- **Οι οφθαλμικές διαταραχές που συνοδεύονται με επηρεασμό της όρασης (καταρράκτης, φακόκωνος) αξιολογούνται και αντιμετωπίζονται συντηρητικά ή χειρουργικά.**







- Alport Syndrome Diffuse Leiomyomatosis (ASDL) (φυλοσύνδετος τύπος).
- Μεγαθρομβοκυτταροπενία (αυτόσωμος επικρατούντας τύπος).
- Μακροκεφαλία και Διανοητική καθυστέρηση.
- Πρώιμη απώλεια οδόντων.
- Ανίχνευση αντιθυρεοειδικών αντισωμάτων.

- Progression to ESRD is ineluctable in males with X-linked Alport syndrome and in all patients with the autosomal recessive form of the disease.

- Renal transplantation is generally a satisfactory treatment, but about 2.5% of patients develop anti-GBM glomerulonephritis leading to rapid graft loss.

Gubler MC *et al.* (2006) Alport syndrome, familial benign hematuria, nail–patella syndrome and type III collagen nephropathy. In *Heptinstall's Pathology of the Kidney*, edn 6, 487–515 (Eds Jennette JC *et al.*) Baltimore: Lippincott, Williams & Wilkins

- Temporary or prolonged alleviation of proteinuria has been achieved by blockade of the renin–angiotensin system, but no data are available on the long-term evolution of renal function in response to this treatment.

- There has been controversy over the effects of cyclosporin in these patients.

Callis L, Vila A, Camera M, Nieto J. Long term effects of cyclosporine A in Alport's syndrome. Kidney Int 1999;55:1051

- Several therapeutic approaches have been tested on canine or murine models of X-linked and autosomal Alport syndrome.

- In Col4a3–/– mice, transgenesis of the human *COL4A3– COL4A4* locus restores the expression of type IV collagen chains and rescues the Alport phenotype.

Heidet L *et al.* (2003) A human-mouse chimera of the α3α4α5(IV) collagen protomer rescues the renal phenotype in Col4a3$^{-/-}$ mice. *Am J Pathol* **163**: 1633–1644

- Transplantation of wild-type bone-marrow-derived stem cells into irradiated Col4a3−/− mice results in the recruitment of wild-type podocytes and mesangial cells, partial restoration of α3(IV) expression in the GBM, and improvement in glomerular structure and function.

Sugimoto H *et al.* (2006) Bone-marrow-derived stem cells repair basement membrane collagen defects and reverse genetic kidney disease. *Proc Natl Acad Sci USA* **103:** 7321–7326

Pharmacologic treatments have also been tested, and it seems that

- angiotensin- converting-enzyme inhibitors and angiotensin-receptor-1 antagonists,
- chemokine receptor 1 blockade,
- statins and
- metalloprotease inhibitors

alleviate proteinuria and prolong the survival of treated animals, if administered before severe renal lesions develop.

Gross O *et al.* (2004) Antifibrotic, nephroprotective potential of ACE inhibitor vs AT1 antagonist in a murine model of renal fibrosis. *Nephrol Dial Transplant* **67:** 1716–1723

Ninichuk V *et al.* (2005) Delayed chemokine receptor 1 blockade prolongs survival in collagen 4A3-deficient mice with Alport disease. *J Am Soc Nephrol* **16:** 977–985

Koepke ML *et al.* (2007) Nephroprotective effect of the HMG-CoA-reductase inhibitor cerivastatin in a mouse model of progressive renal fibrosis in Alport syndrome. *Nephrol Dial Transplant* **22:** 1062–1069

- Accordingly, it will be necessary to be able to recognize Alport syndrome early in the course of disease development if trials of similar therapies in humans are to prove successful.

Thin basement membrane disease (BFH)

- The long-term prognosis is excellent in most patients with thin basement membrane disease.
- Slowly progressive renal insufficiency can occur and is often manifested on renal biopsy by focal and segmental glomerulosclerosis.
- It has been unclear whether thin basement membrane disease was responsible for the progressive glomerular injury or was an incidental finding in patients with underlying focal glomerulosclerosis.

Tryggvason K and Patrakka J (2006) Thin basement membrane nephropathy. *J Am Soc Nephrol* **17:** 813–822

- **A study of 19 normotensive and nonazotemic patients with biopsy-proven thin basement membrane disease (in whom clinical and histologic criteria tended to exclude hereditary nephritis) suggested that focal glomerulosclerosis and slowly progressive renal insufficiency may be more common than previously suggested.**

Nienwhof CM, de Heer F, de Leeuw P, van Breda Vriesman PJ. Thin GBM nephropathy. Premature glomerular obsolescence is associated with hypertension and late onset renal failure. Kidney Int 1997;51:1596

The following findings were noted in the patients and their families:

- Increased incidence of focal global sclerosis on renal biopsy compared to matched controls with IgA nephropathy (13.5 versus 5.1 percent of glomeruli).

- Family history revealed that 6 of 89 first-degree relatives (all of whom were elderly) had renal failure compared to 1 of 129 with IgA nephropathy.

- At the end of 12 year follow-up, three of seven subjects over age 50 had a modest decline in glomerular filtration rate (64 to 79 mL/min).

- **The frequency of thin basement membranes in the general population means that it will often be found as an incidental finding in other renal diseases:**

In diabetic nephropathy, for example, thin basement membrane disease may be responsible for otherwise unexplained hematuria

Matsumae T, Fukusaki M, Sakata N, et al. Thin glomerular basement membrane in diabetic patients with urinary abnormalities. Clin Nephrol 1999;42:221

It has been suggested that thin basement membrane disease may occur with slightly increased frequency in patients with IgA nephropathy.

It is unclear if there is a true relation between these disorders or if thin membranes might increase the severity of concurrent IgA nephropathy.

Chystomou A, Kincaid-Smith P, Becker G. Clinical features and prognosis of thin basement membrane (abstract). J Am Soc Nephrol 1993;4:261.

- **As noted above, occasional patients have recurrent episodes of gross hematuria and/or flank pain. The administration of an angiotensin converting enzyme inhibitor may be beneficial in this setting, perhaps by lowering the intraglomerular pressure.**

Hebert LA, Betts JA, Sedmak DP, et al. Loin pain-hematuris syndrome associated with thin glomerular membrane disease and hemorrhage into renal tubules. Kidney Int 1996;49:168

- **In addition, treating hypercalciuria or hyperuricosuria, if present, has reduced hematuria in other patients.**

Praga M, Alegre R, Hernandez E, et al. Familiar microscopic hematuria caused by hypercalciuria and hyperuricosuria. Am J Kidney Dis 2000;35:141

COL4A1 and Familial Hematuria

- No mutations of *COL4A1* or *COL4A2* have been detected in patients with Alport syndrome or isolated BFH/TBMN.
- *COL4A1* mutations have been identified in a mouse model of, and in families affected by, porencephaly and in a novel rare autosomal dominant syndrome known as hereditary angiopathy with nephropathy, aneurysms and cramps(HANAC).

Zhang KW *et al.* (2007) Do mutations in *COL4A1* or *COL4A2* cause thin basement membrane nephropathy (TBMN)? *Pediatr Nephrol* **22:** 645–651

Laminin β2 Disease
Pierson Syndrome

- In 1963, Pierson et al. reported the curious association in siblings of eye abnormalities with microcoria and congenital nephrotic syndrome progressing rapidly to ESRD.
- The glomerular lesions were classified as mesangial sclerosis, with diffuse alteration of the GBM.
- Hypotonia and psychomotor retardation developed in the few patients who survived for several months after birth.

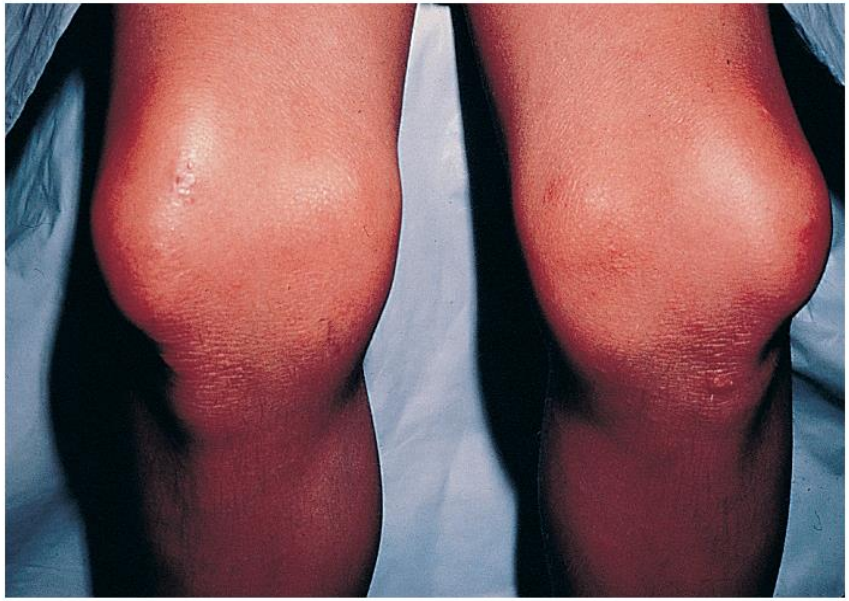
Zenker M *et al*. (2004) Human laminin β2 deficiency causes congenital nephrosis with mesangial sclerosis and distinct eye abnormalities (Pierson syndrome). *Hum Mol Genet* **13:** 2625–2632

Hereditary Renal Diseases With Type III Collagen Deposits

Nail-patella Syndrome

- osteo-onychodysplasia
- rare autosomal dominant disorder (affecting 1 in 50,000 individuals) characterized by an association between nail hypoplasia or dysplasia and bone abnormalities that primarily affect the knees, elbows and pelvis.
- Normal-tension glaucoma and sensorineural hearing impairment have been recognized as additional features of the disease.

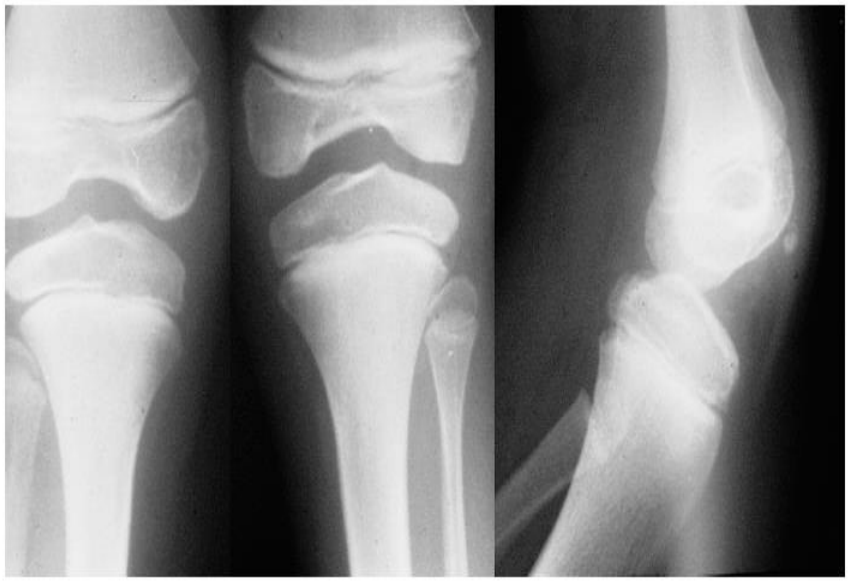
Bongers EM *et al*. (2005) Genotype-phenotype studies in nail–patella syndrome show that LMX1B mutation location is involved in the risk of developing nephropathy. *Eur J Hum Genet* **13:** 1019–1024

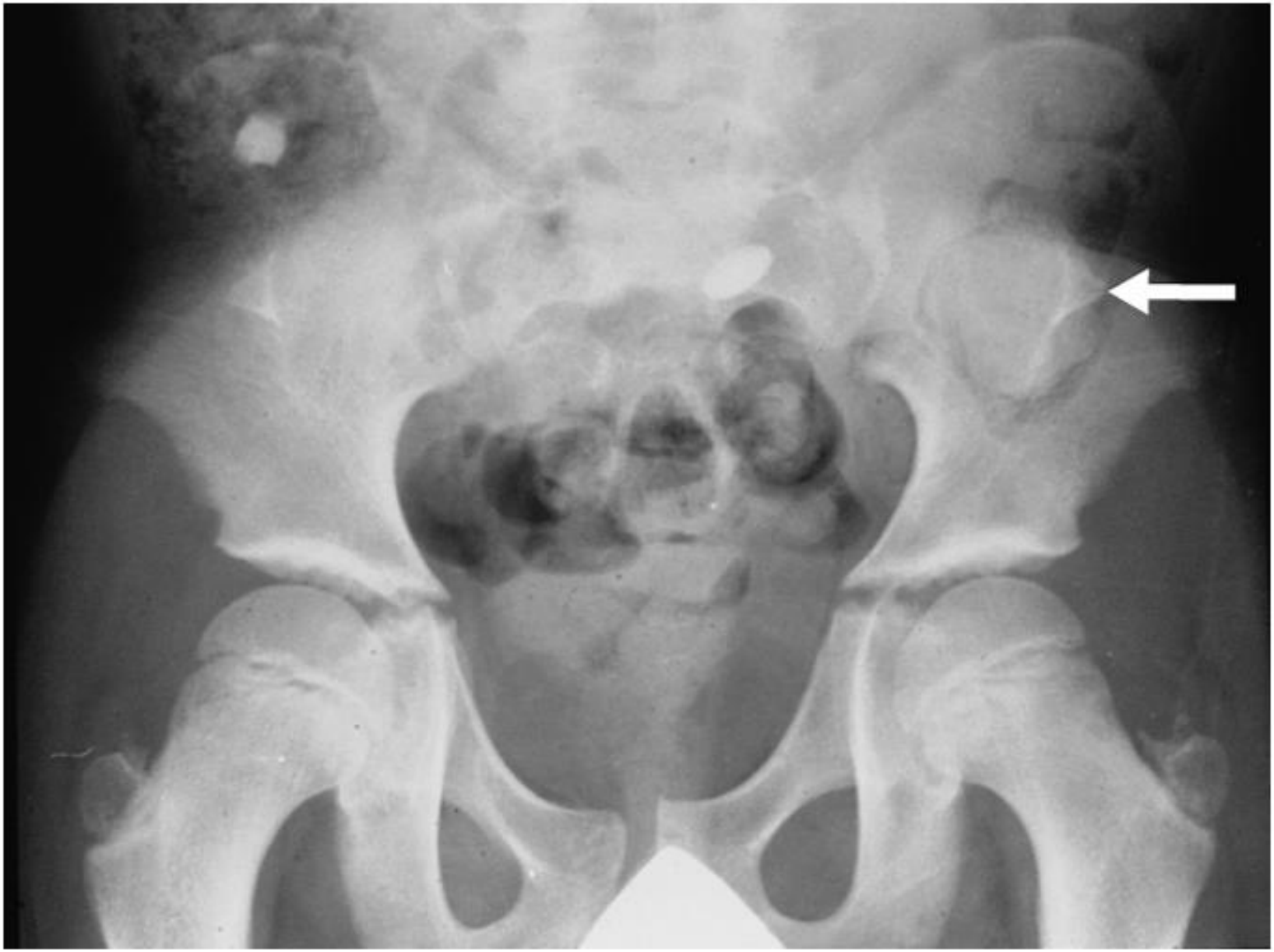


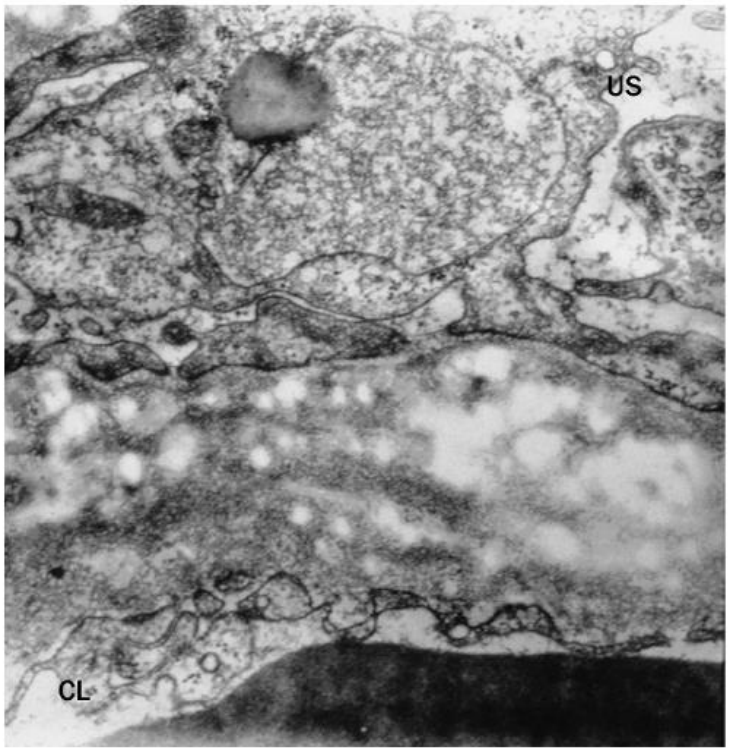
Renal involvement usually manifests as proteinuria, sometimes with hematuria.

The prognosis depends on the presence and severity of renal involvement, which is observed in 30–40% of patients.

Progression to kidney failure occurs in about 30% of patients.







The hallmark of the disease, observed by electron microscopy, is the presence of clusters of fibrillar type III collagen irregularly distributed within thick GBM segments and the mesangial matrix.

Collagen Type III Glomerulopathy

- ✓ Accumulation of type III collagen in the glomerular extracellular matrix has been detected in some proteinuric patients.
- ✓ Diffuse glomerular changes (i.e. Marked expansion of the mesangial matrix and thickening of the capillary walls) can be observed under the light microscope.

- At the ultrastructural level, the mesangial matrix and the subendothelial aspect of the GBM are enlarged and have a mottled appearance due to the presence of fibrillar collagen (which can be visualized after staining with phosphotungstic acid).

- Unlike the lesions observed in NPS, the lamina densa is usually preserved.

Conclusions

- **Alport syndrome was the first characterized, and is the most frequent hereditary hematuric disorder of the GBM that progresses to ESRD.**
- **Therapeutic progress is expected to result from extension of the promising results obtained in animal models to human trials.**

Thinning of the GBM is not a marker of a specific disease entity, and does not guarantee a benign disease course.

- Long-term follow-up and repeat investigations might be needed before a definitive diagnosis can be made.

- The wide spectrum of phenotypes observed in patients that carry a heterozygous *COL4A3* or *COL4A4* mutation heralds a need for caution during risk assessment and genetic counseling.

- *The recent identification of LAMB2 mutations in congenital and infantile nephrotic syndromes indicates that hereditary disorders resulting from defects in genes encoding noncollagen*

  components of the GBM might not yet have been identified.

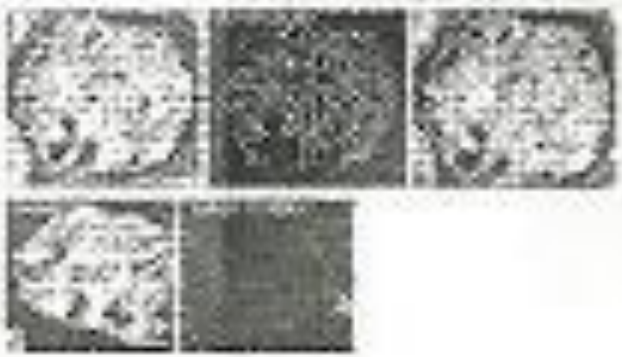
- **Clinical investigation and documentation of atypical symptoms are the basis for the recognition of new syndromes.**

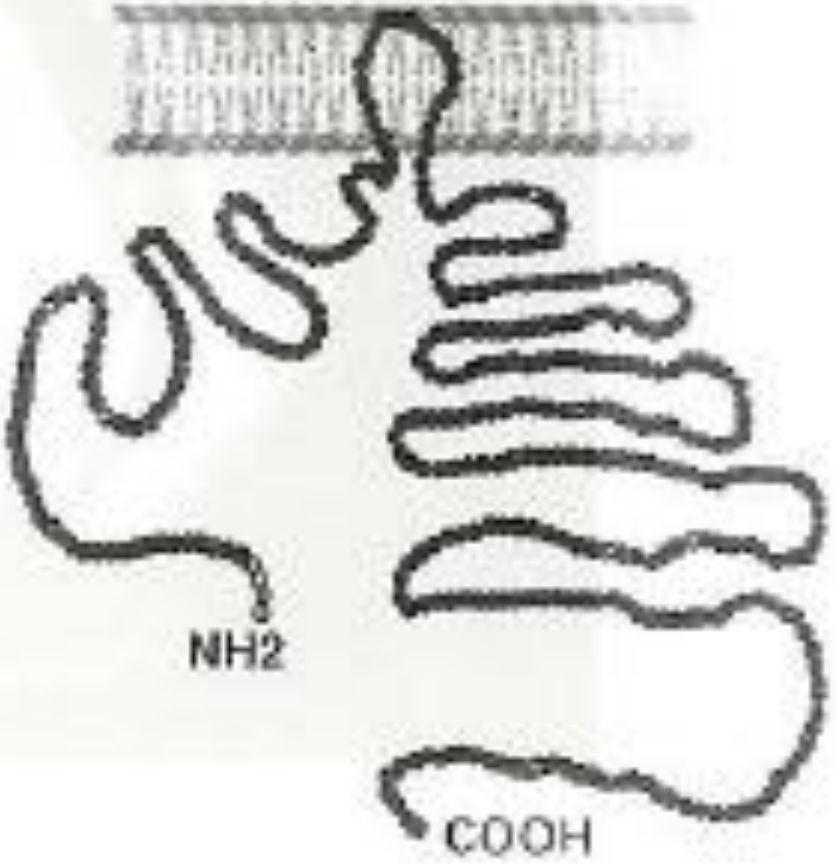
- No specific treatments are available for inherited diseases in which mutation of genes that encode components of the glomerular basement membrane (GBM) perturb its structure.

Familial FSGS

- 1/3 of pediatric steroid-resistant nephrotic syndrome or FSGS is due to mutations in the podocin gene NPHS2
- Recessive: absence of FH does not rule out "familial" form of disease

NPHS2: Podocin

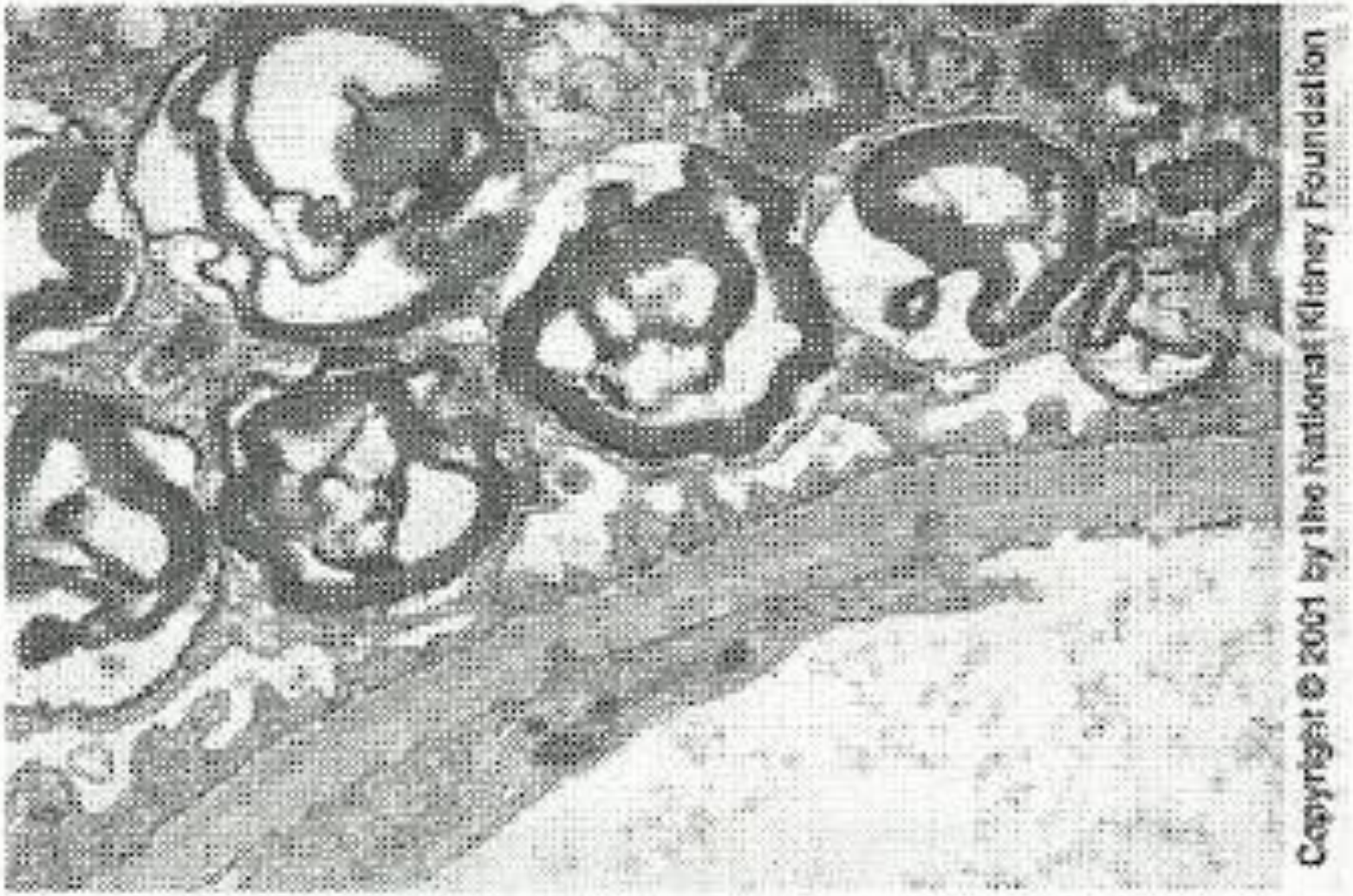




Fabry's Disease

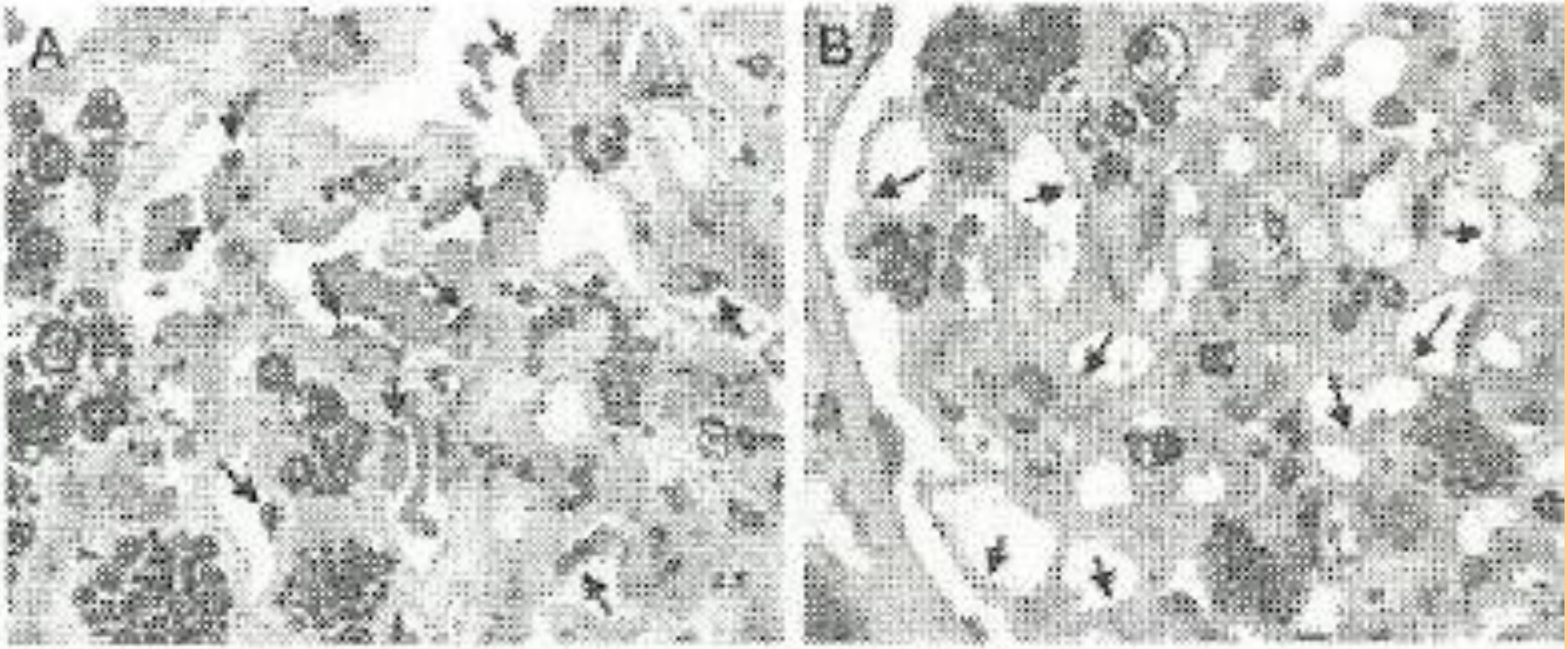
- X-linked deficiency of lysosomal α-galactosidase → accumulation of neutral sphingolipids, mostly globotriasylceramide (GL3)
- Classically, symptoms begin ~ 10 years of age
  - Characteristic angiokeratomas, groin, hips, umbilical region
  - Characteristic corneal opacity, which does not affect vision
  - Peripheral neuropathy with severe acroparasthesias
  - Mixed glomerular and tubular renal disease, typically ESRD by 3-4th decade
  - Cardiac involvement – LVH, conduction abnormalities, arrhythmias
  - Vascular disease (endotheliopathy)

Lamellated Lysosomal Inclusions of GL3 in Podocytes of a Patient with Fabry Disease



Atlas of Renal Pathology, *AJKD*, 2001

Reduced Glomerular Endothelial GL3 After Enzyme Replacement



Pre-treatment **5 mos. treatment**

↑ – GL3 deposits

Thurberg et al, *KI*, 2002

Diagnosis

- Affected males → α-galactosidase activity in peripheral WBC. Atypical variants may have residual activity.
- Female carriers may have normal enzyme activity → DNA analysis
- Female carriers may have significant disease burden, due to random X-inactivation

Treatment

- Enzyme replacement therapy, with Replagal or Fabrazyme
- Reduces plasma GL3 to undetectable, in addition to reduced tissue accumulation
- Indications, dosage, endpoints, and cost-effectiveness are still being clarified by clinical studies

Summary and "take-home" points

- Most common Mendelian form of renal failure: ADPKD
- Hematuria: consider familial disease (Alport syndrome, thin basement membrane)
- Electrolyte, BP disorders: consider familial renal tubule defects

Chronic metabolic alkalosis/hyperkalemia

- Diuretic abuse
- Bulemia
- Hyperaldosteronism
- Bartter/Gitelman syndromes
  - Dx: Urinary chloride
  - Family history
  - Molecular genetics

Bartter's vs Gitelman's syndromes

	BARTTER's	GITELMAN's
Location of defect	Ascending limb of Henle	Distal tubule
Age of presentation	Prenatal, infancy, early childhood	Late childhood or adulthood
Biochemical differences	Serum Mg sometimes low Increased urine Ca excretion	Serum MG low Decreased urine Ca excretion
Molecular etiology	Na-K-2Cl cotransporter, apical K channel (ROMK), basolateral Cl channel (CLCNKB) Henle	Na-Cl cotransporter in distal tubule

Question #3

- You are asked to evaluate a 26 y.o. woman with mental retardation because a recent USG showed bilateral renal cysts. Study was performed because of RUQ pain and ? GB disease. Other USG findings are normal GB and somewhat irregularly-sized kidneys measuring 11.5 cm bilaterally. No FH of renal disease, but father was told that he had fatty tumors in both kidneys two years ago, after he underwent colectomy for colon CA.
- Exam is unremarkable. 51 kg. Creatinine 1.3 mg/dL. UA normal.

Question #3

Which of the following is the most likely diagnosis?

- (A) ADPKD
- (B) ARPKD
- (C) von Hippel-Lindau disease
- (D) Tuberous sclerosis
- (E) Multiple simple cysts

Question #3

Which of the following is the most likely diagnosis?

- (A) ADPKD
- (B) ARPKD
- (C) von Hippel-Lindau disease
- (D) Tuberous sclerosis
- (E) Multiple simple cysts