ERKNet
The European Rare Kidney Disease Reference Network

european society for paediatric nephrology

WELCOME TO

ESPN/ERKNet
Educational Webinars on Pediatric Nephrology & Rare Kidney Diseases

Date: 02 February 2021

Topic: C3 Glomerulopathies

Speaker: Christoph Licht

Moderator: Marina Vivarelli

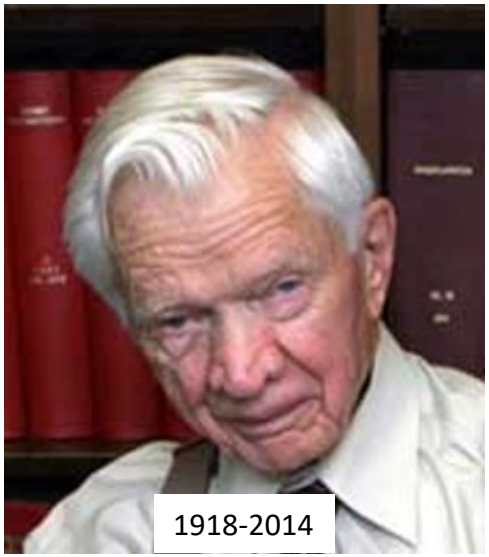
Outline

- MPGN – the traditional classification
- From MPGN to C3G – the new classification
- Pathogenesis – lessons learned from mice and men
- Diagnosis, treatment and outcome
- Summary and perspectives

MPGN – the traditional classification

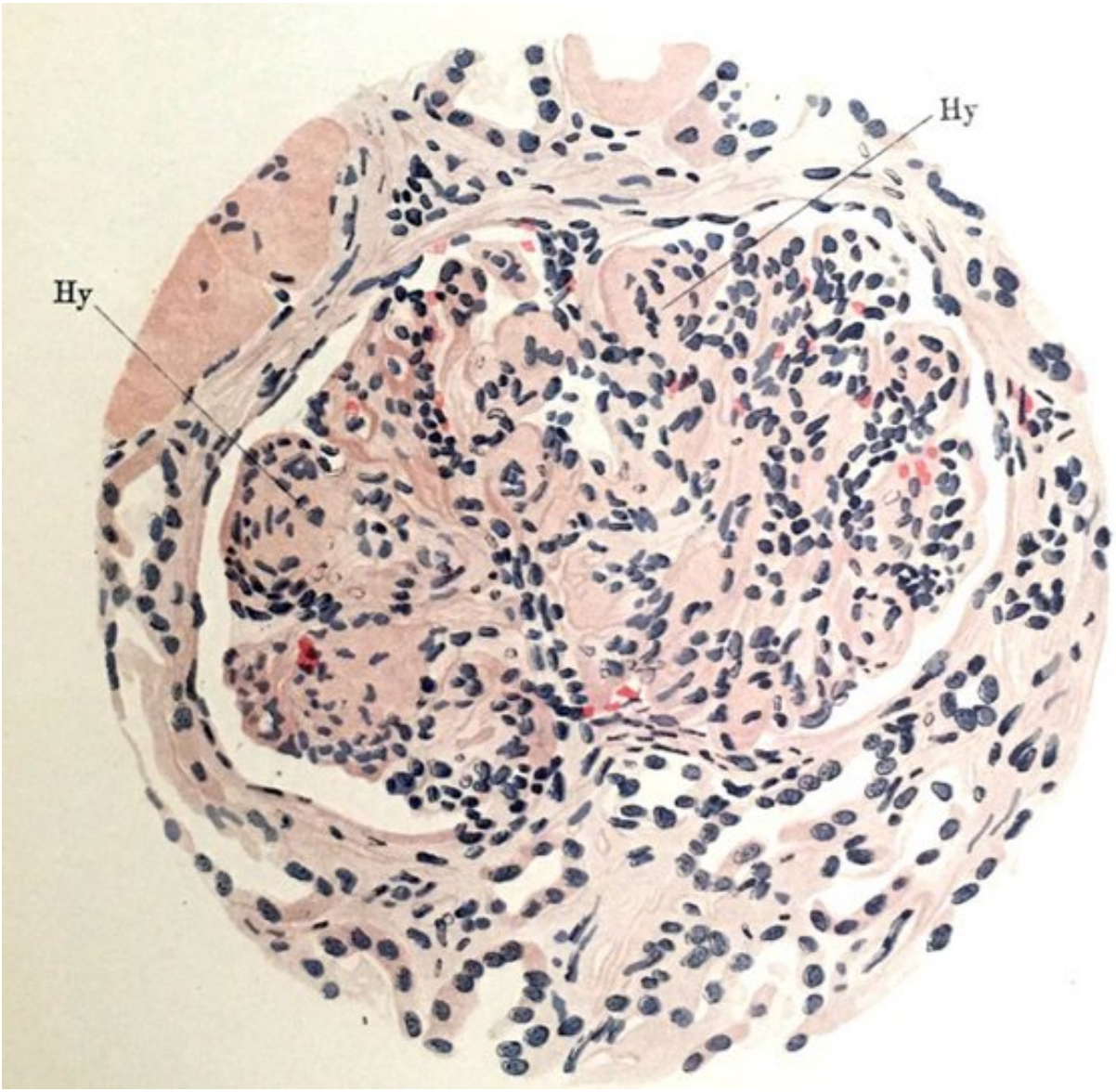
Historical background

- Volhard and Fahr (1914): Die Brightsche Nierenkrankheit
  “Camera lucida” drawing of lobular glomerulonephritis
- Habib and Hamburger (1960):
  At the first worldwide Renal Biopsy Meeting, MPGN is first defined via a variety of findings made in patients with Bright's disease; MPGN becomes a formally named sub-type of glomerulonephritis ("a disease").
- Clinical phenotype (1960's):
  - Mean age 10 (range 2-17)
  - Nephrotic syndrome (70%)
  - Hematuria (90%)
  - Renal function “low” (33%)
- Clark West (1965):
  - Describes hypocomplementemia in MPGN
  - Complement defects (classical pathway) suspected


1918-2014

Wolstenholme and Cameron, Editors (1961). CIBA Foundation Symposium on Renal Biopsy: Clinical and Pathological Significance.
West, J Pediatr 1965

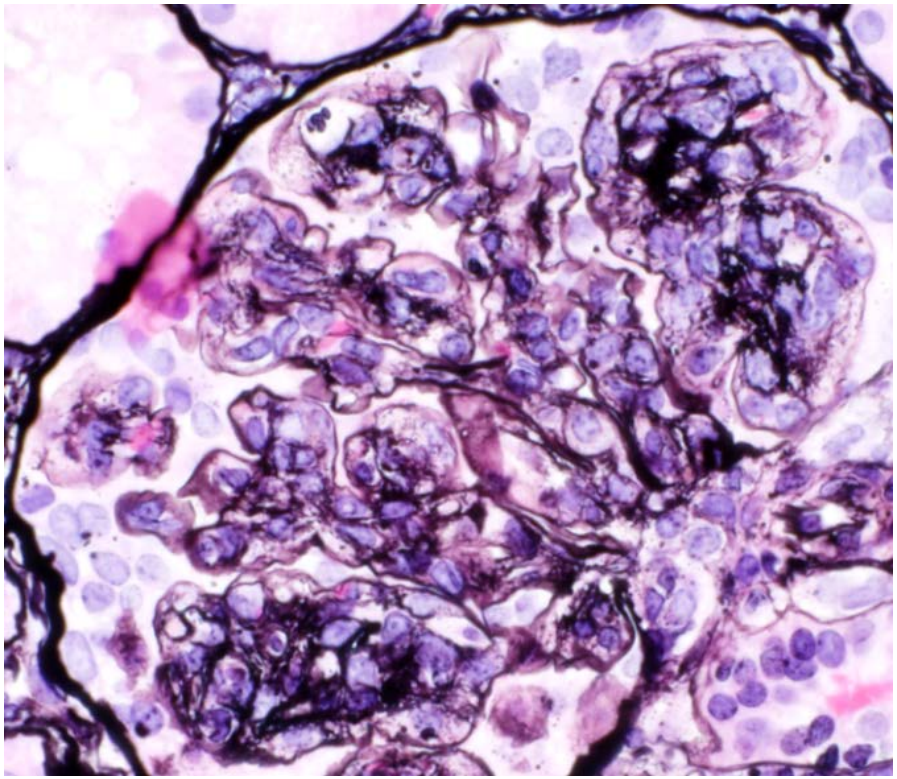
Bright's disease



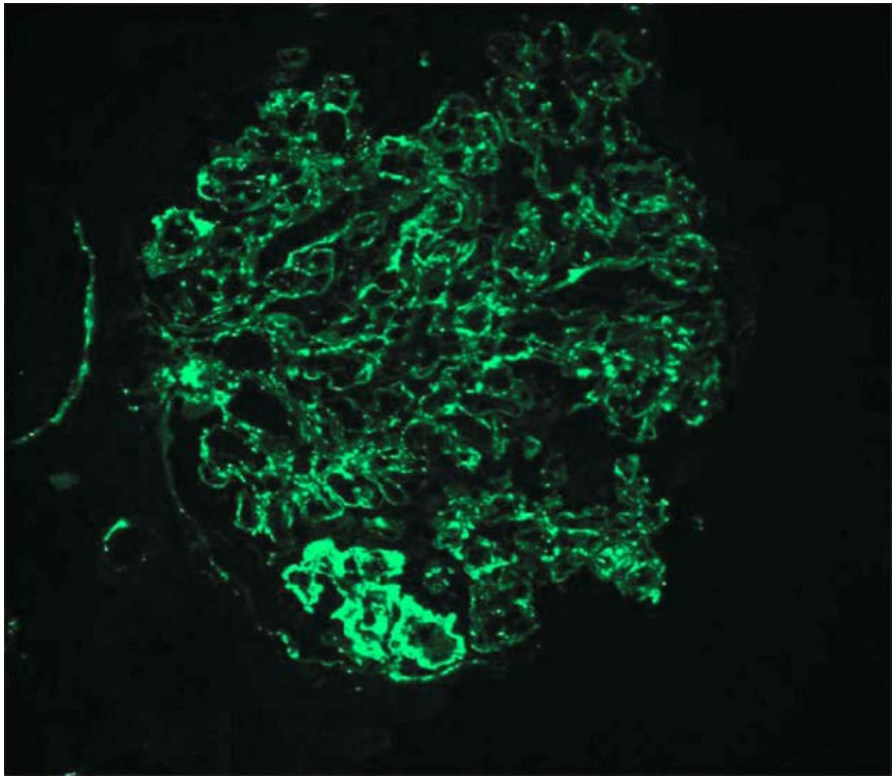
Volhard and Fahr (1914). Die Brightsche Nierenkrankheit

Membranoproliferative pattern

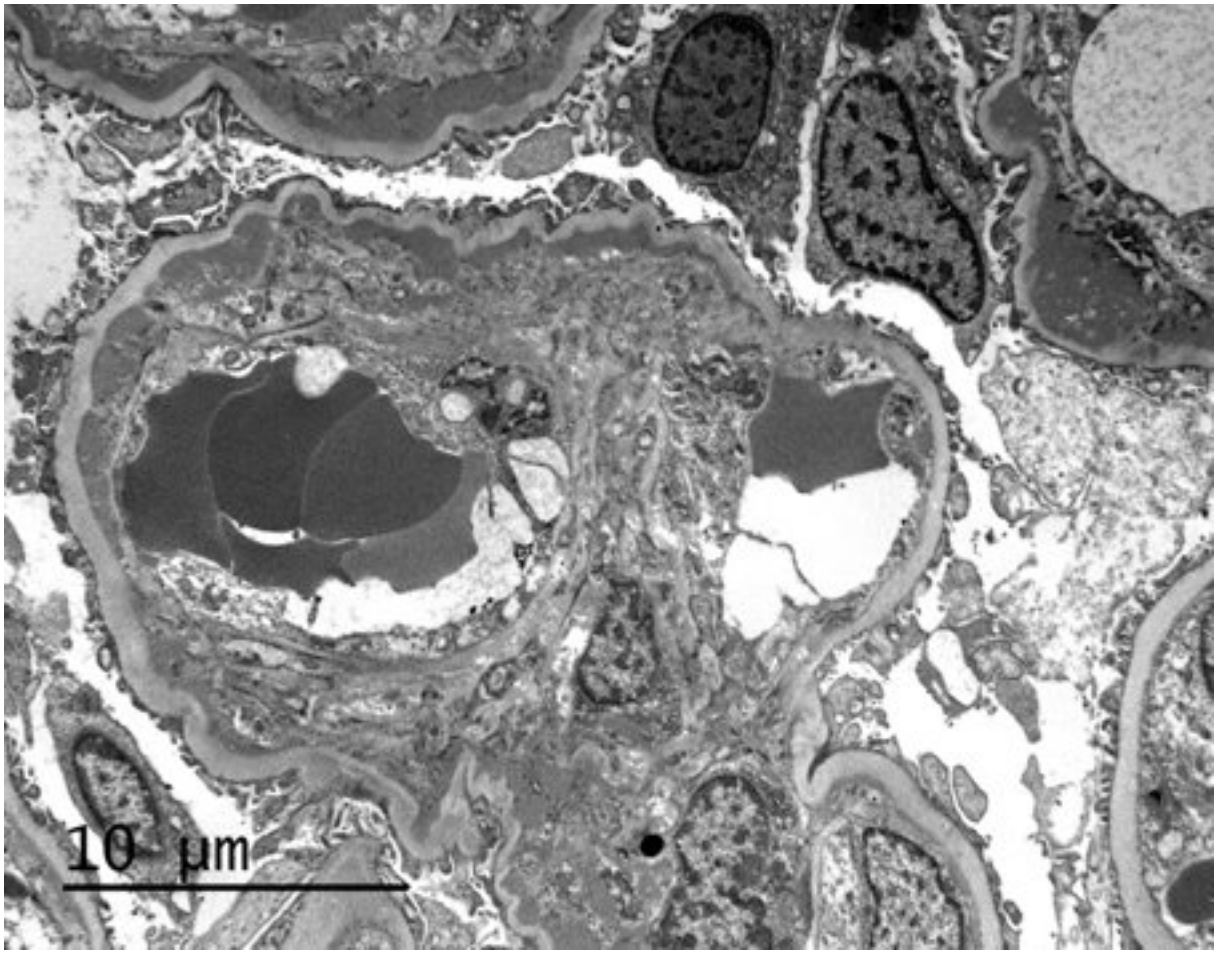
Membranoproliferative pattern



C3 and IgG



Membranoproliferative pattern

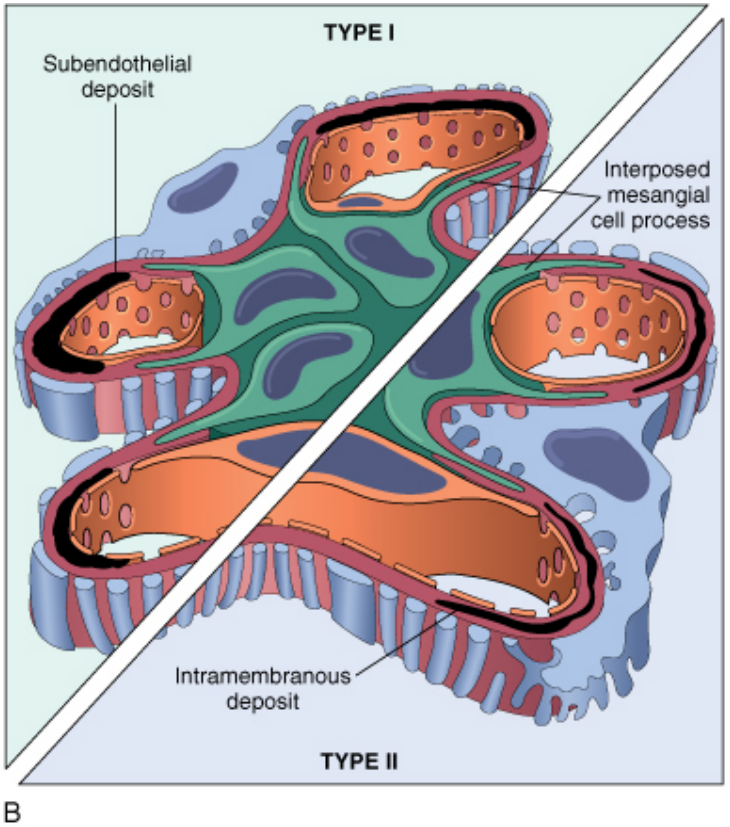


Membranoproliferative pattern (1970's-1980's)

- MPGN Type I:
  Subendothelial deposits
  West et al, J Pediatr 1965

- MPGN Type II / DDD:
  Deposits in lamina densa of glomerular basement membrane
  Galle, Thesis 1962; Habib et al, Kidney Int 1975

- MPGN Type III:
  Subendothelial and subepithelial deposits
  Burkholder et al, Am J Pathol 1969
  Anders et al, Virchows Arch A Pathol Anat Histol 1997
  Strife et al, Clin Nephrol 1984



B

Histological findings in MPGN include … ?

A. Electron dense deposits in various glomerular localizations

B. C3

C. IgG

D. All of the above

Which is not a cause of secondary MPGN?

A. HIV

B. Hepatitis B / C

C. Leukemia / lymphoma

D. Cystic fibrosis

Secondary MPGN (80%)

Condition	Diagnosis
Infections (bacterial / viral / protozoal)	Hepatitis B / C EBV HIV Malaria Mycoplasma Tuberculosis
Systemic immune disease	Cryoglobulinemia Systemic lupus erythematosus Sjögren's syndrome Rheumatoid arthritis
Neoplasmas / dysproteinemias	Plasma cell dyscrasia Light / heavy chain disease Leukemia / lymphoma / other malignancies Waldenstrom macroglobulinemia
Chronic liver disease	Hepatitis / cirrhosis Alpha-1-antitrypsin deficiency
Miscellaneous (null C3 + null IgG)	TMA (aHUS / TTP) Radiation nephropathy Antiphospholipid syndrome Sickle cell disease Transplant glomerulopathy

Riedl / Licht, Pediatr Nephrol 2017 (modified)

From MPGN to C3G – the new classification

ORIGINAL ARTICLE

Primary glomerulonephritis with isolated C3 deposits: a new entity which shares common genetic risk factors with haemolytic uraemic syndrome

Aude Servais, Véronique Frémeaux-Bacchi, Moglie Lequintrec, Rémi Salomon, Jacques Blouin, Bertrand Knebelmann, Jean-Pierre Grünfeld, Philippe Lesavre, Laure-Hélène Noël, Fadi Fakhouri

J Med Genet 2007;**44**:193–199. doi: 10.1136/jmg.2006.045328

New concept of a complement disease spectrum:

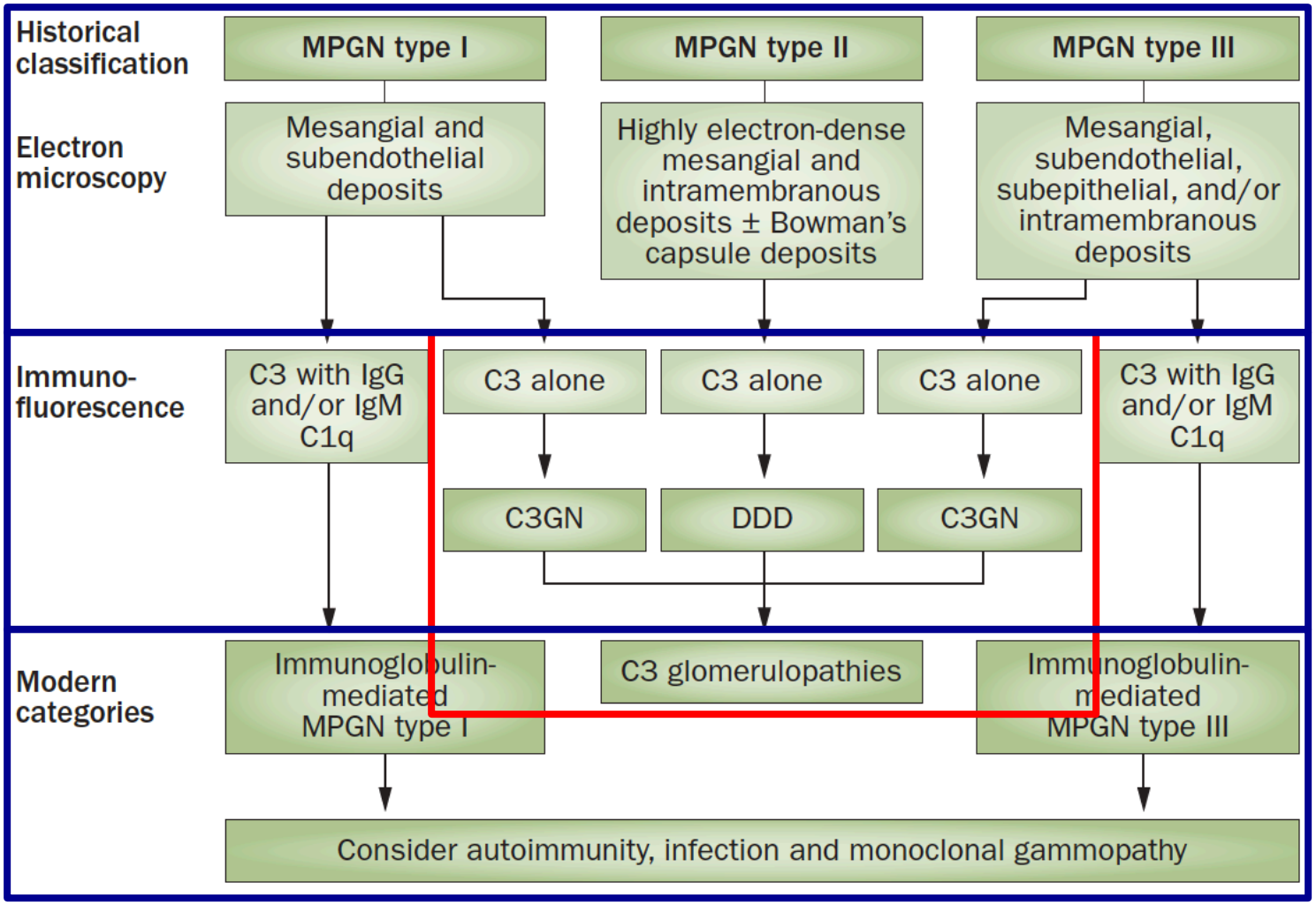
- aHUS and C3G share common genetic risk factors
- Constitutional or acquired risk factors are associated with disease spectrum ranging from aHUS to C3G
- aHUS is caused by local (EC) complement dysregulation
- C3G is caused by systemic complement dysregulation

Local activation ? Systemic activation ?

Unidentified factors

Servais et al, J Med Genet 2007

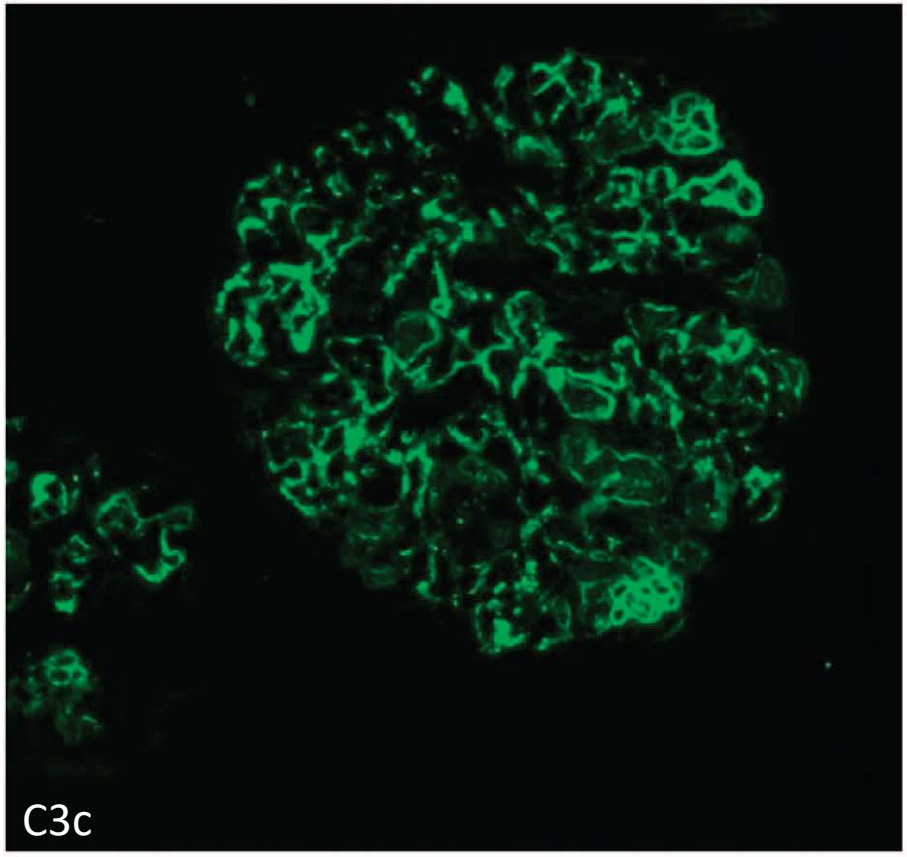
Historical vs. current classification of *MPGN*

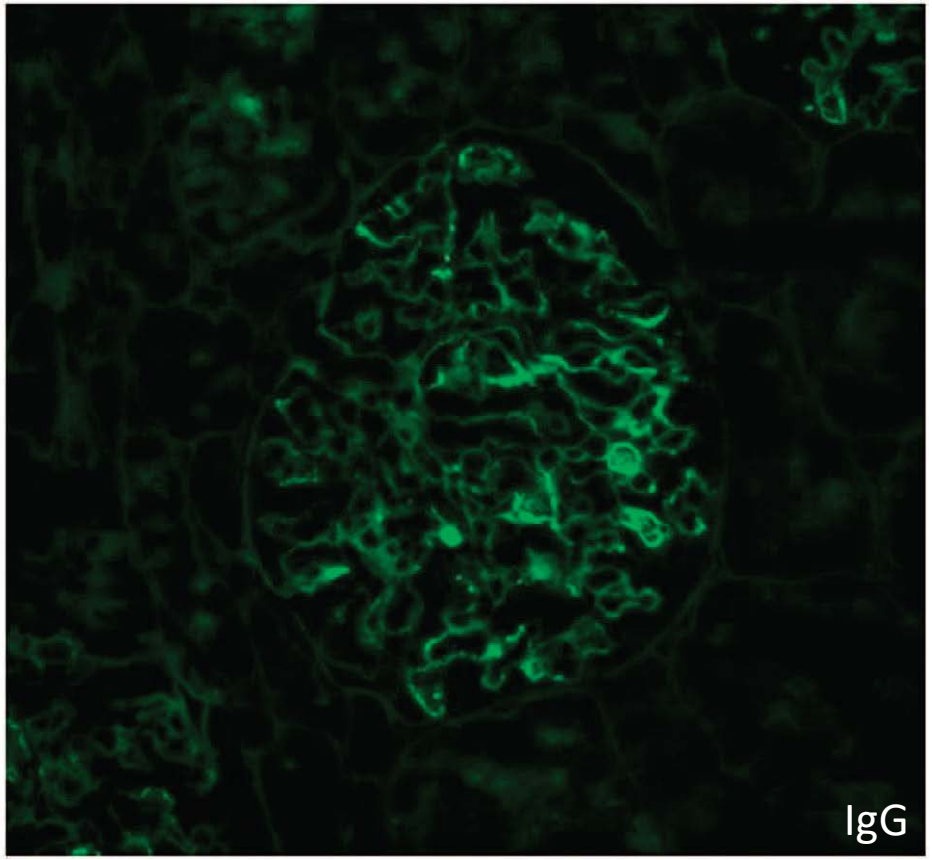


Cook and Pickering, Nat Rev Nephrol 2015

C3 Glomerulopathy
Consensus

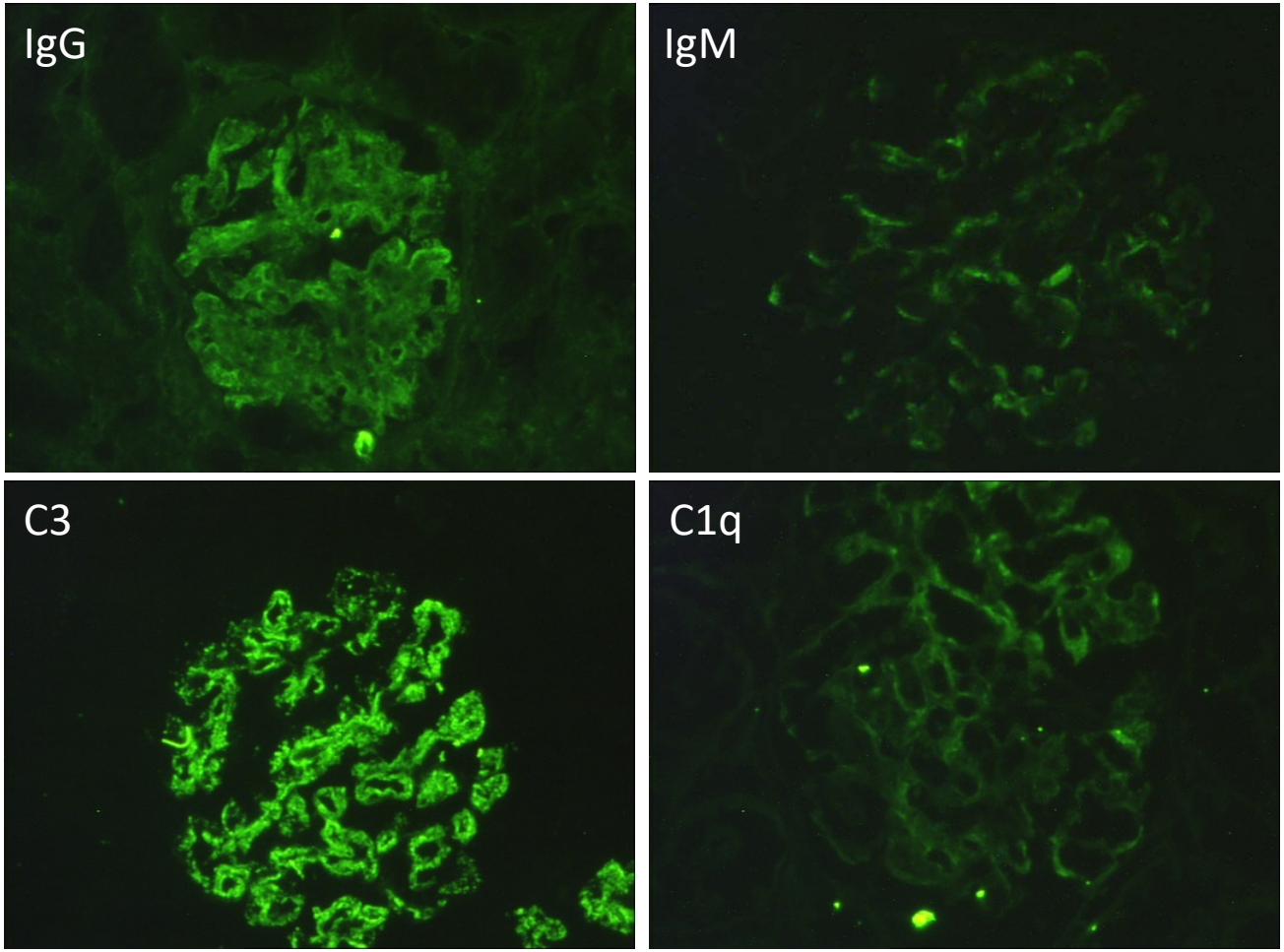
Dominant C3 staining:
C3 staining > 2 magnitudes stronger





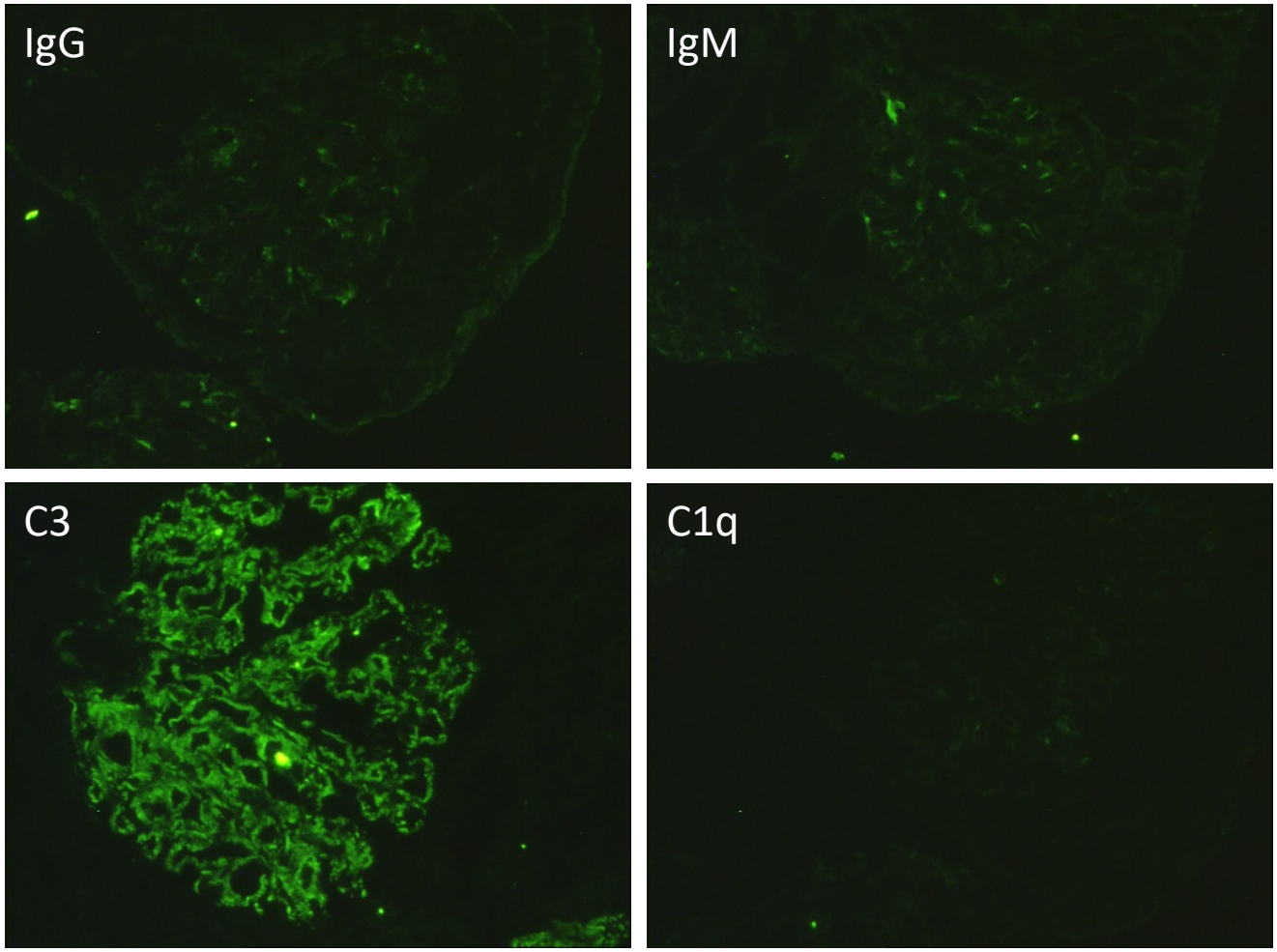
Pickering et al, Kidney Int 2013; Cook and Pickering, Nat Rev Nephrol 2015

First biopsy: IC-MPGN



Courtesy of Francesca Diomedi, Pathology, Bambino Gesu Pediatric Hospital, Rome, Italy

Second biopsy (+10 months): C3GN



Courtesy of Francesca Diomedi, Pathology, Bambino Gesu Pediatric Hospital, Rome, Italy

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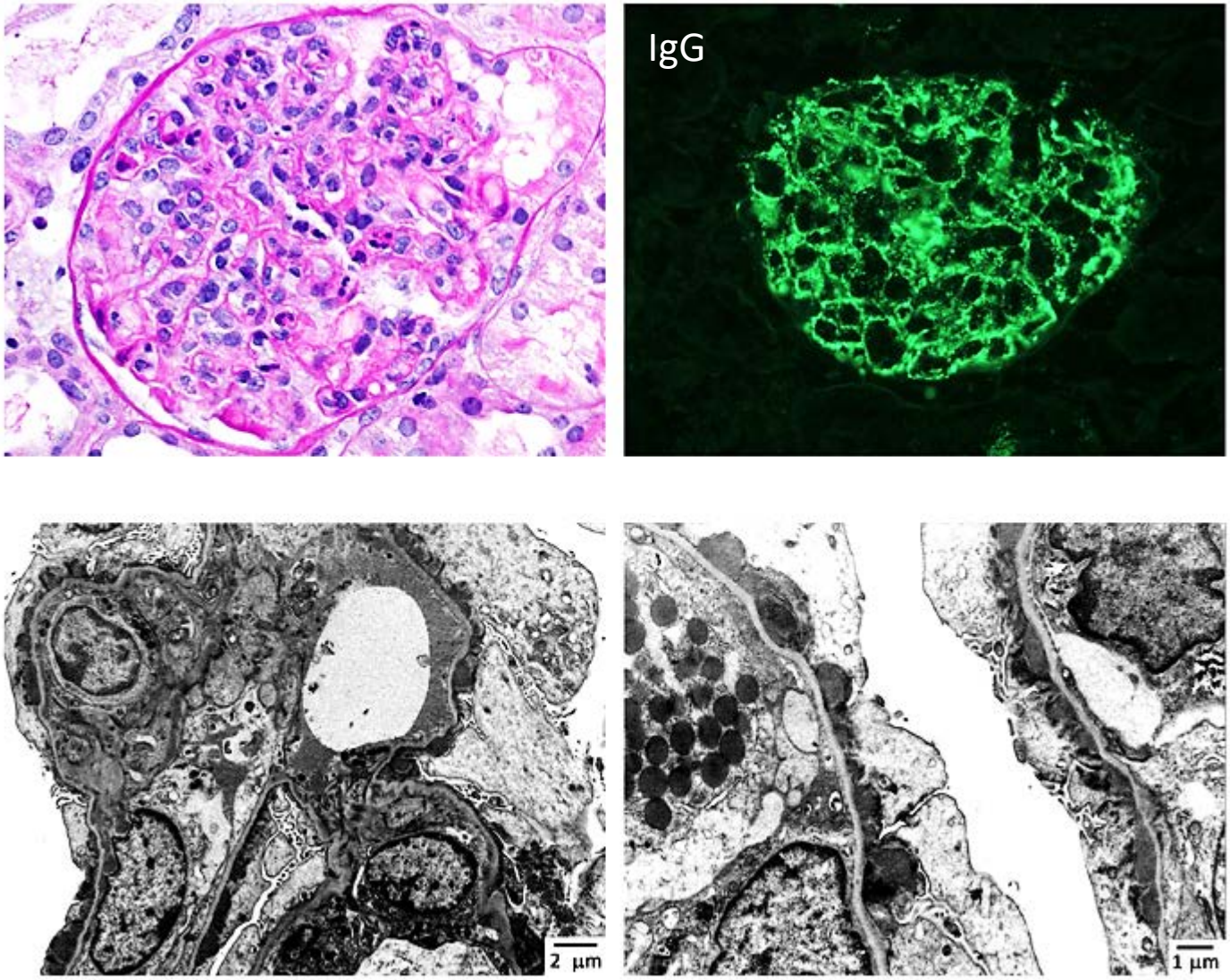
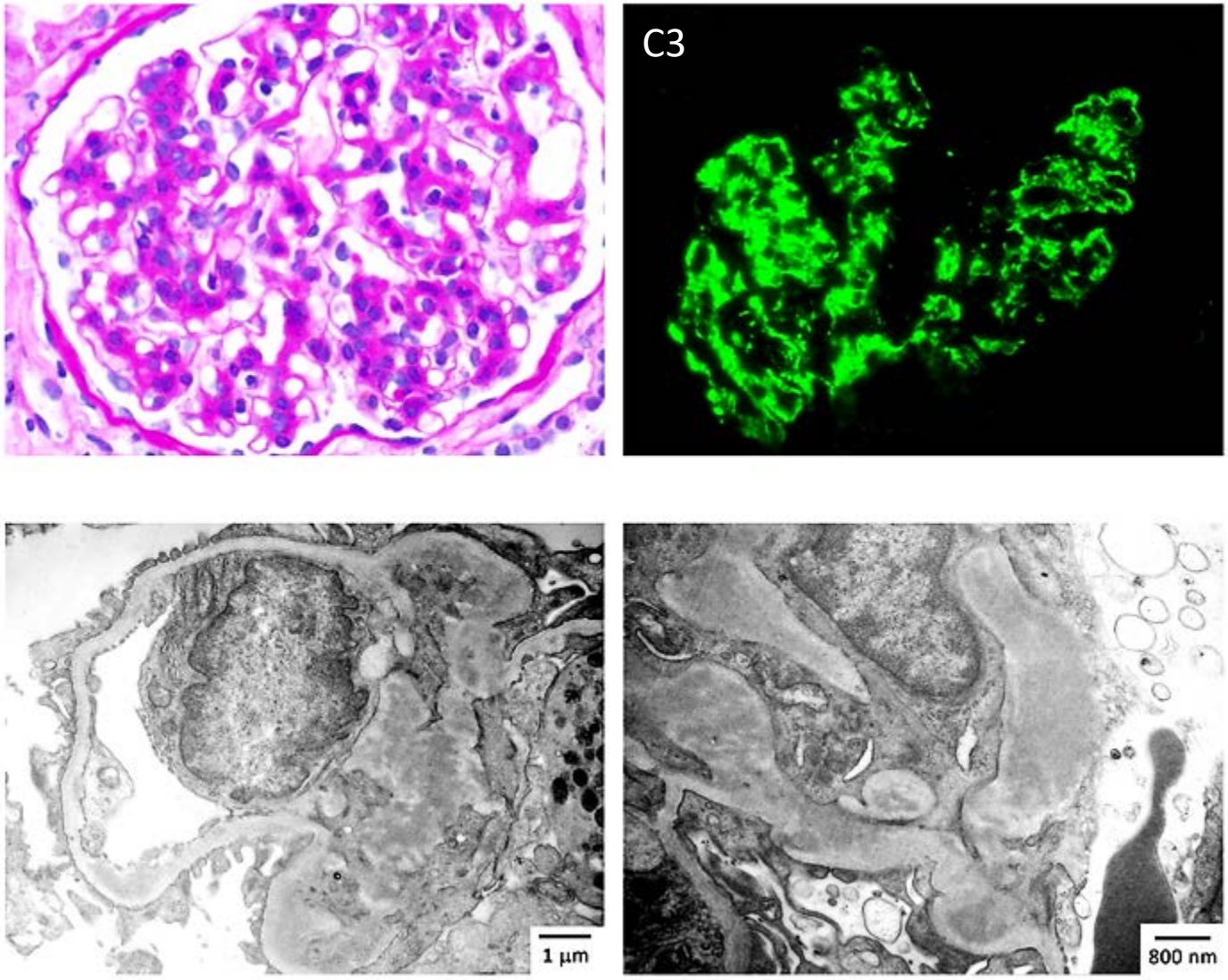


Courtesy of Patrick Walker, Arkana Laboratories, Little Rock, AR

Second biopsy (+6 months): C3GN



Courtesy of Patrick Walker, Arkana Laboratories, Little Rock, AR

SickKids®

Pathogenesis

Lessons learned from mice and men

Which complement pathway is mainly involved in the pathogenesis of MPGN?

A. Classical pathway

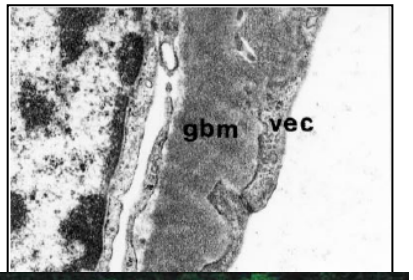
B. Lectin pathway

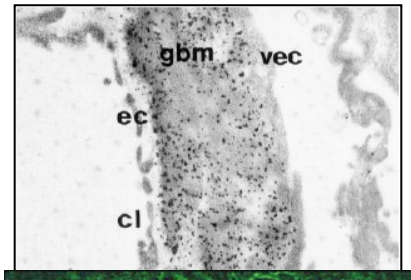
C. Alternative pathway

D. Terminal pathway

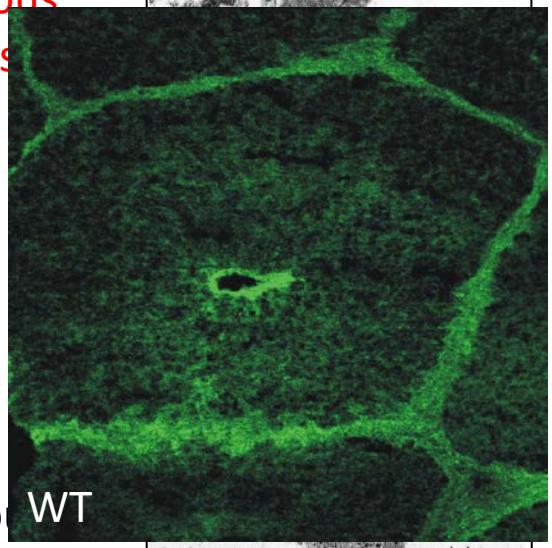
Cfh-/- pigs

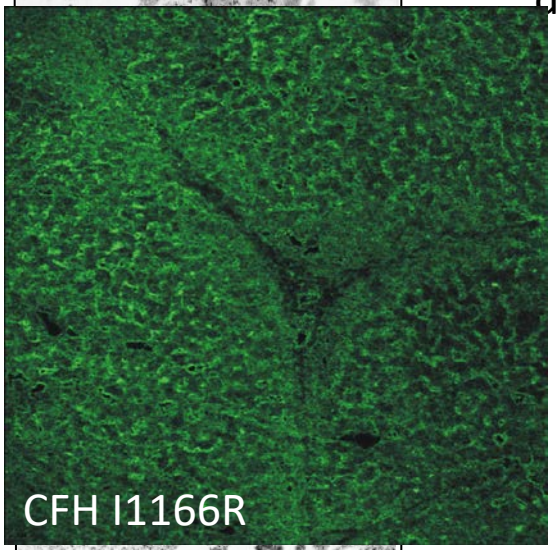
Thickened GBM with intramembraneous dense deposits



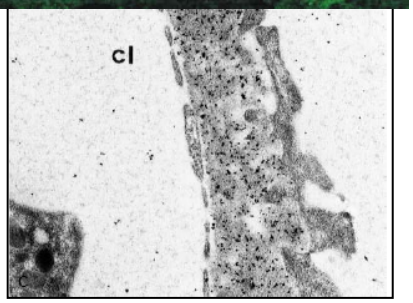


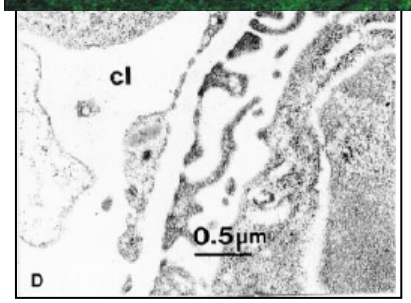
Intramembraneous dense deposits contain C3





Intramembranous dense deposits contain C5b-9

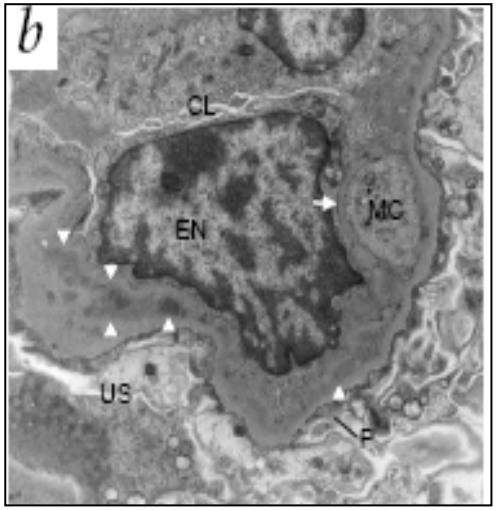




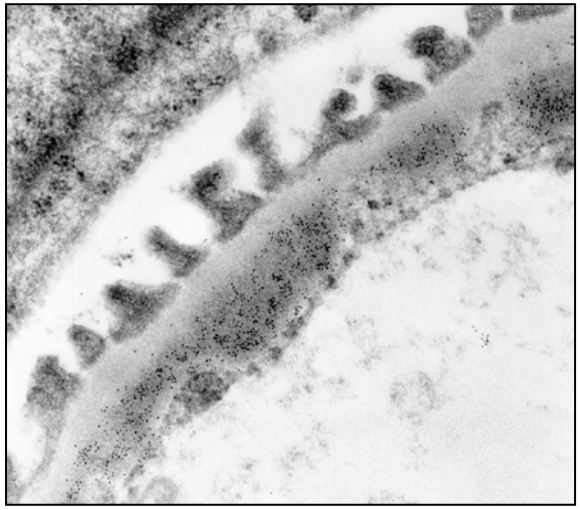
Control

Hogasen et al, J Clin Invest 1995; Jansen et al, Kidney Int 1998; Hegasy et al, Am J Pathol 2002

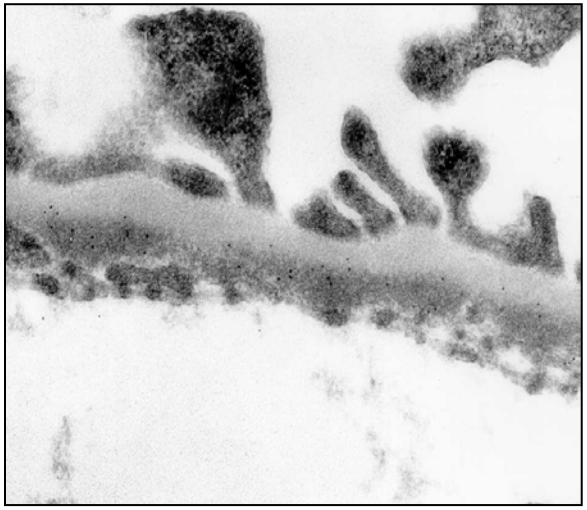
Cfh-/- mice



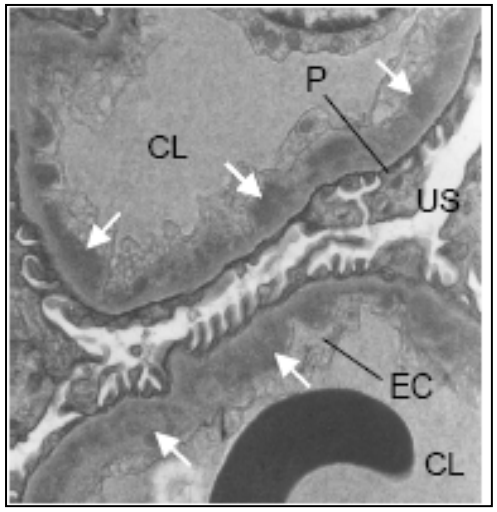
Cfh-/-



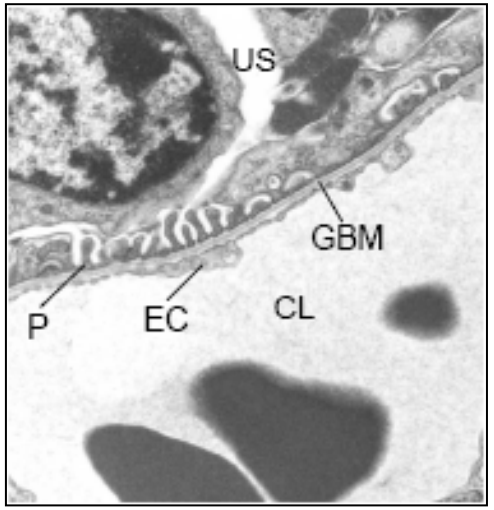
C3



C9



Cfh-/-Cfb+/-



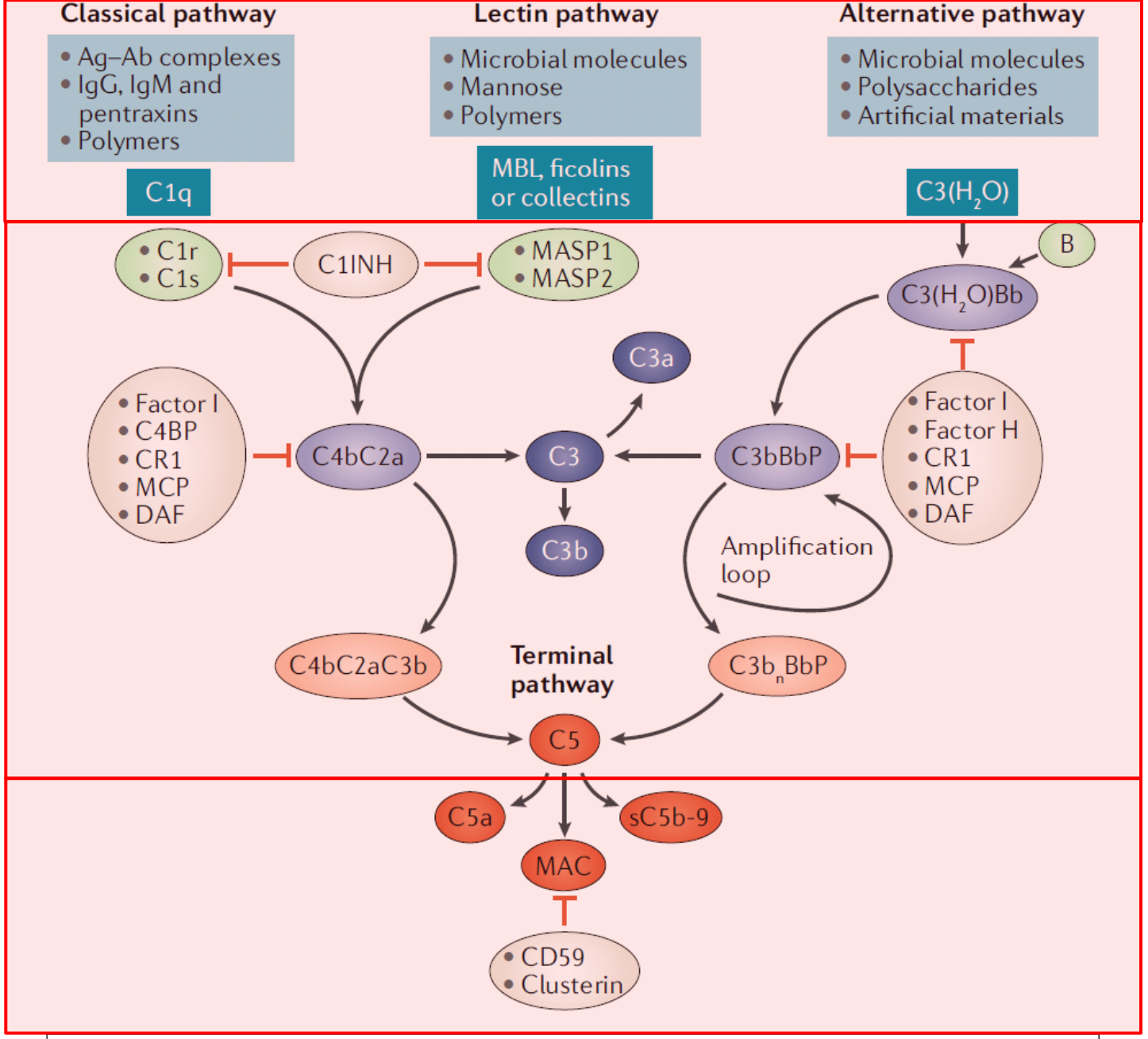
Cfh-/-Cfb-/-

Pickering et al, Nat Genet 2002

Activation pathways

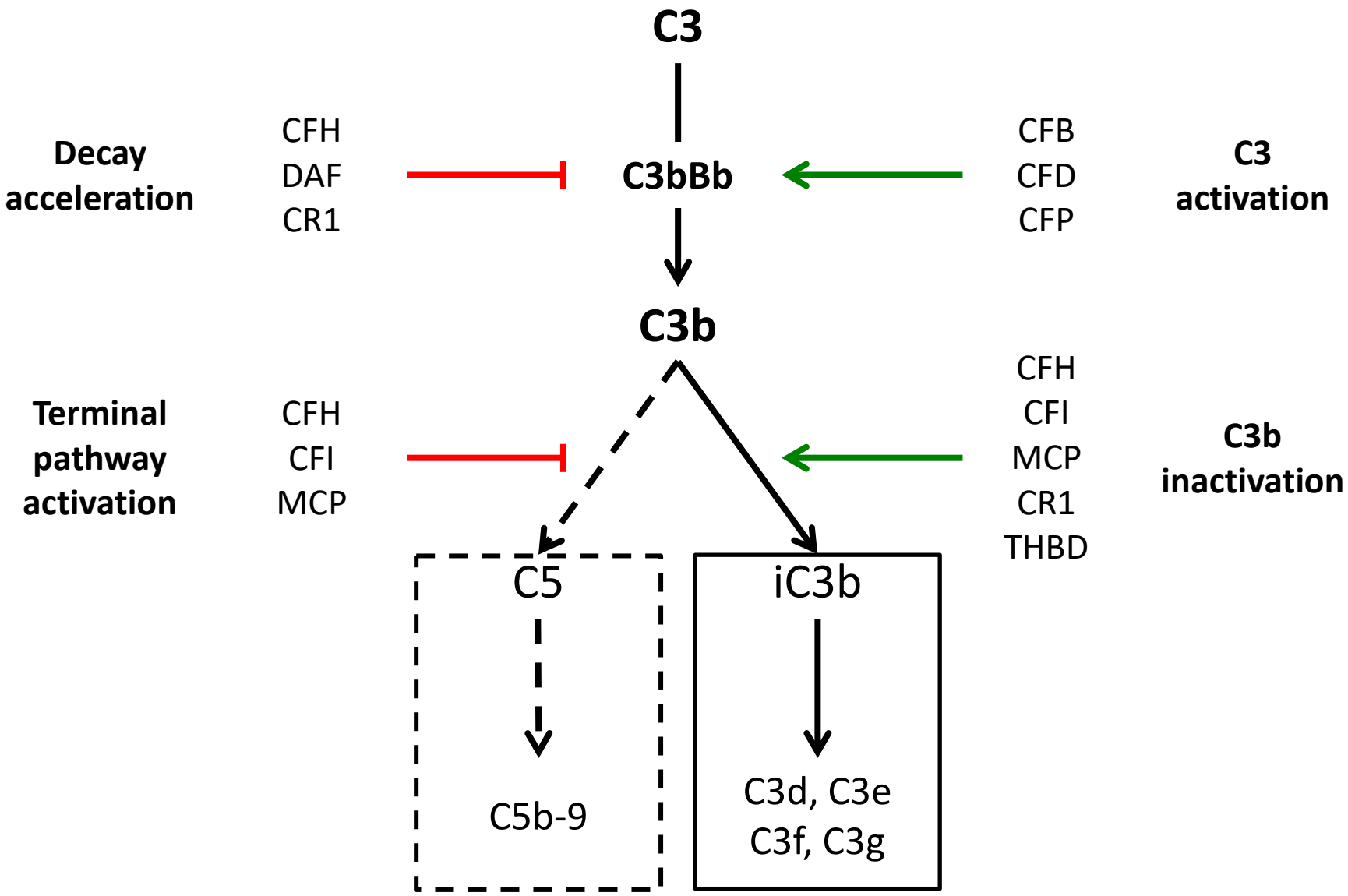
Amplification loop

Terminal pathway



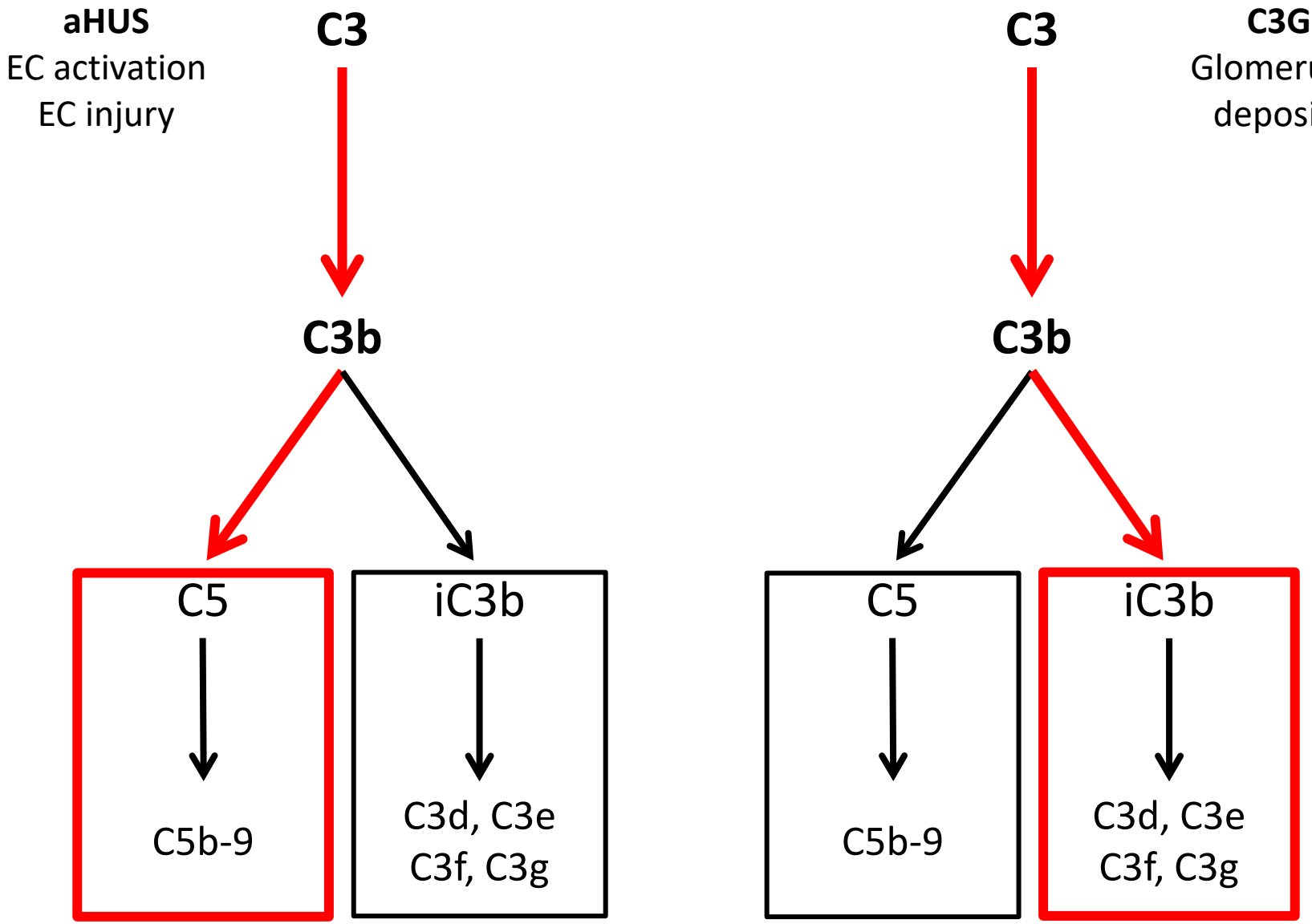
Biglarnia et al, Nat Rev Nephrol 2018

Alternative pathway activation and regulation



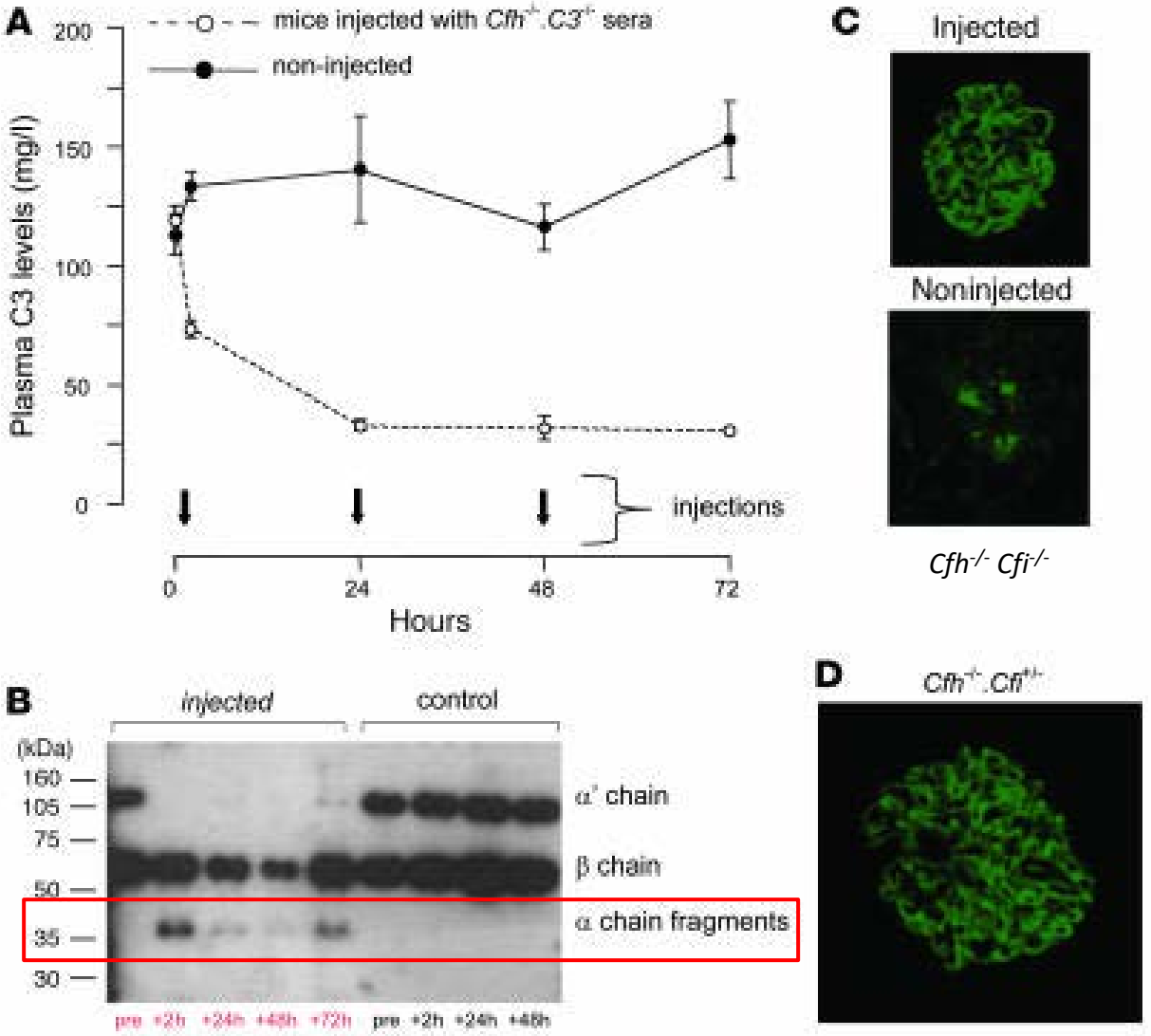
Adapted from Riedl / Licht, Pediatr Nephrol 2017

Alternative pathway dysregulation
aHUS vs. C3G



Adapted from Riedl / Licht, Pediatr Nephrol 2017

Role of C3 in complement-mediated glomerular disease: *Cfh-/- Cfi-/-* mice



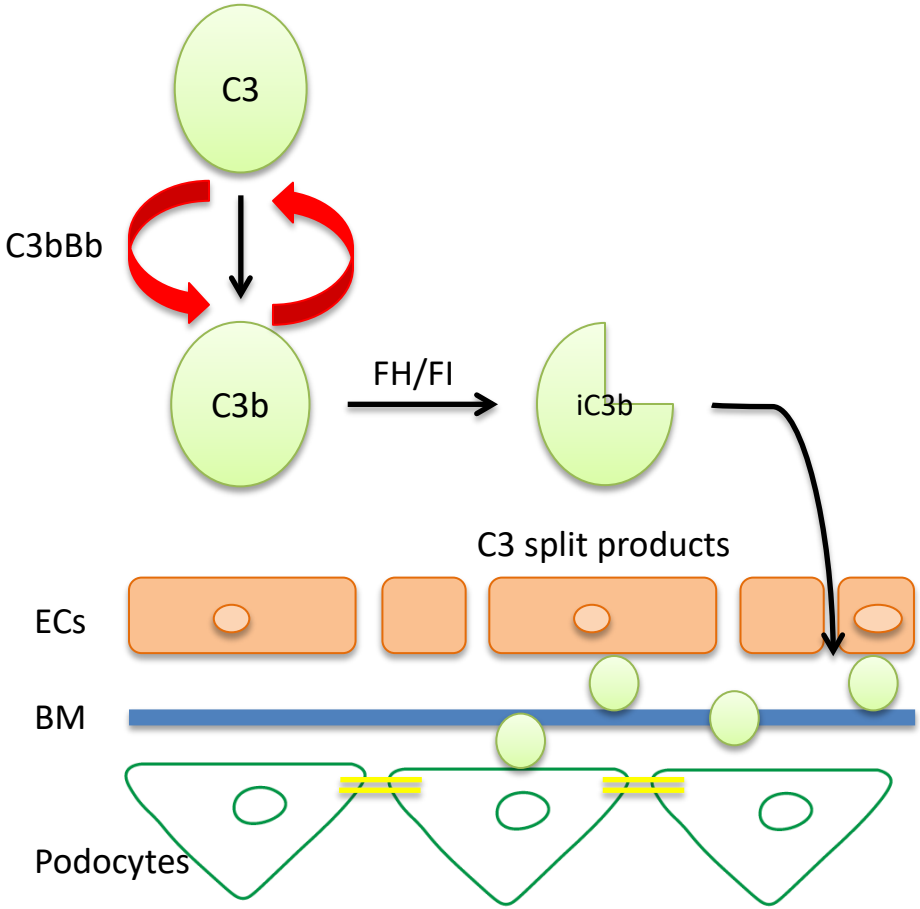
Rose et al, J Clin Invest 2008

Complement alternative pathway defects in C3G
Summary

Autoantibodies

<u>C3bBb components</u>
- C3NeF etc.
- FB
- C3b

<u>C3bBb regulators</u>
- FH

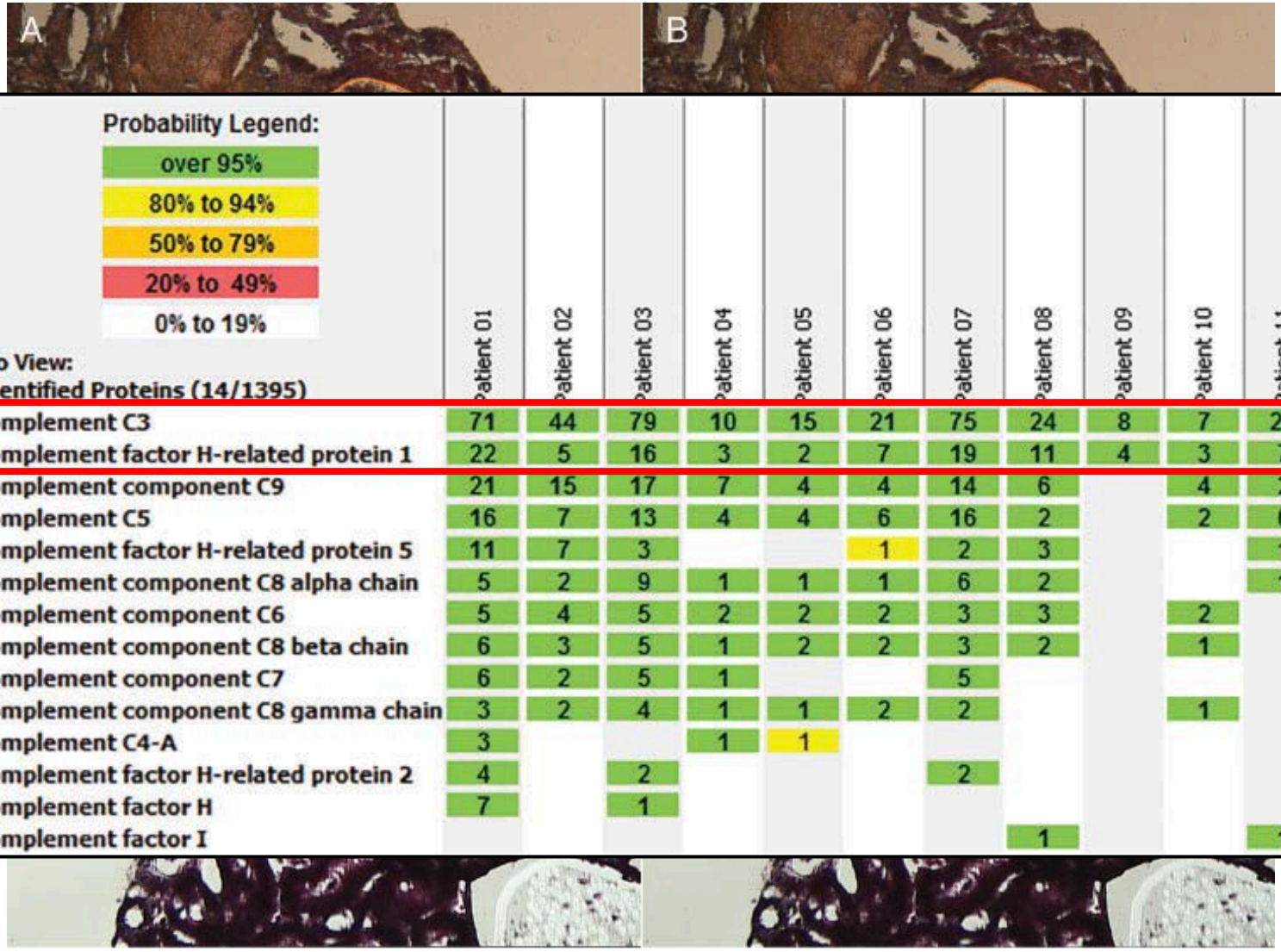


Mutations

<u>C3bBb components</u>
- C3
- FB

<u>C3bBb regulators</u>
- FH
- FI
- MCP/CD46
- THBD/CD141

Glomerular complement profile in C3G



Probability Legend:

Probability
over 95%
80% to 94%
50% to 79%
20% to 49%
0% to 19%

#	Starred	Bio View: Identified Proteins (14/1395)	Patient 01	Patient 02	Patient 03	Patient 04	Patient 05	Patient 06	Patient 07	Patient 08	Patient 09	Patient 10	Patient 11	Patient 12
1	★	Complement C3	71	44	79	10	15	21	75	24	8	7	25	22
2	★	Complement factor H-related protein 1	22	5	16	3	2	7	19	11	4	3	7	5
3	★	Complement component C9	21	15	17	7	4	4	14	6		4	2	3
4	★	Complement C5	16	7	13	4	4	6	16	2		2	6	7
5	★	Complement factor H-related protein 5	11	7	3			1	2	3			1	3
6	★	Complement component C8 alpha chain	5	2	9	1	1	1	6	2			1	2
7	★	Complement component C6	5	4	5	2	2	2	3	3		2		2
8	★	Complement component C8 beta chain	6	3	5	1	2	2	3	2		1		1
9	★	Complement component C7	6	2	5	1			5					3
10	★	Complement component C8 gamma chain	3	2	4	1	1	2	2			1		
11	★	Complement C4-A	3			1	1							2
12	★	Complement factor H-related protein 2	4		2				2					
13	★	Complement factor H	7		1									1
14	★	Complement factor I								1			1	

Sethi et al, Kidney Int 2009; Sethi et al, Nephrol Dial Transplant 2017

Autoimmune causes for C3G
Autoantibodies

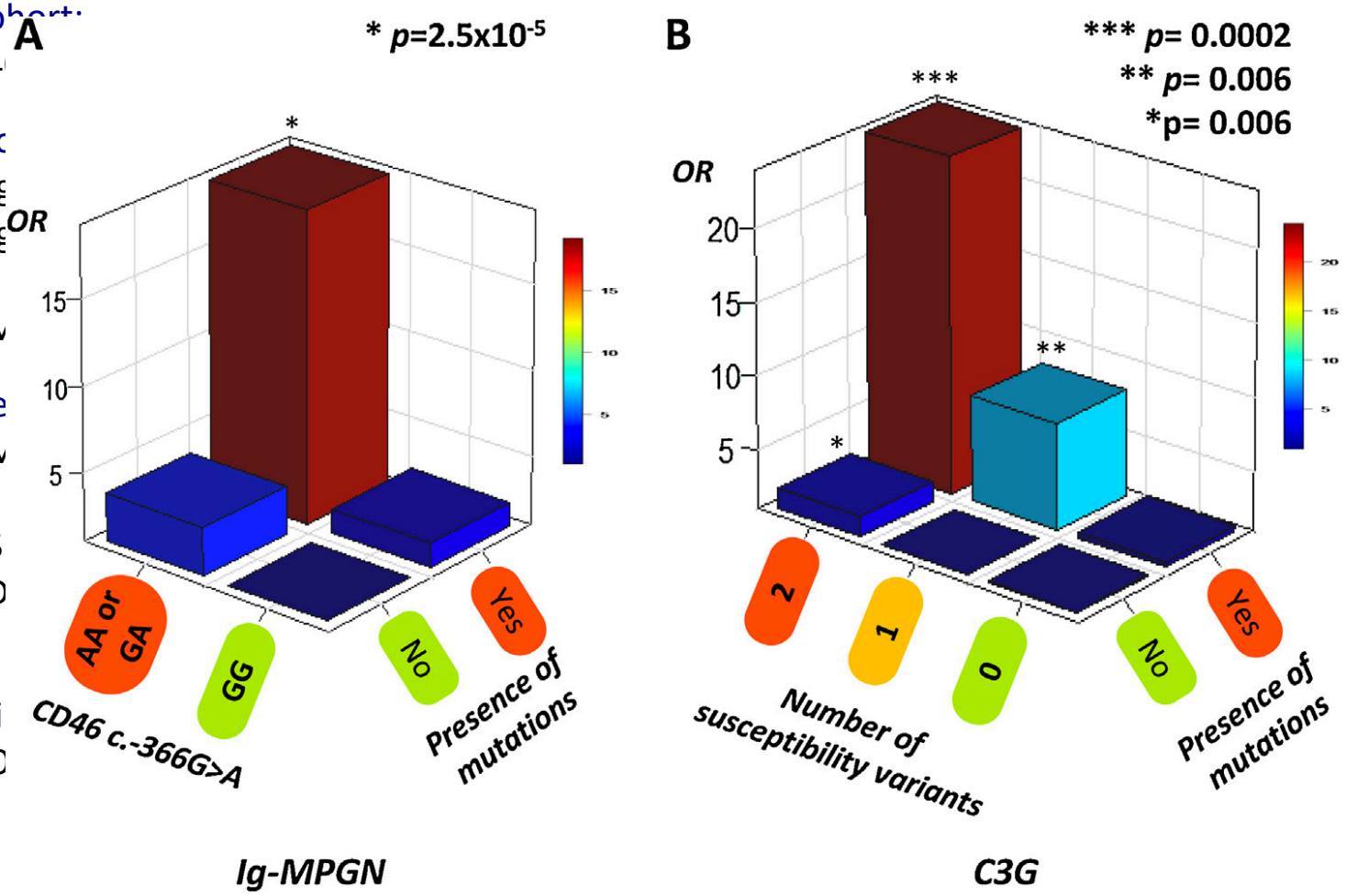
	Incidence	Co-existing with C3Nef?	Effect on complement	Routine testing?
C3NeF	Common	-	Stabilizes AP C3 convertase	Yes
C4NeF	Rare	Yes	Stabilizes CP C3 & C5 convertases	No
C5NeF	Rare	Yes	Stabilizes AP C5 convertase	No
Anti-FB Ab	Rare	No	Stabilizes AP C3 convertase	No
Anti-C3b Ab	Rare	No	Stabilizes AP C3 convertase	No
Anti-FH Ab	Rare	Yes	Fluid phase regulation	Yes

Genetic causes for C3G

Gene/Protein	Mutation/SNP	Function	Phenotype
FH	Homo-/compound heterozygous SCRs 1–4 (regulatory domain)	Intact surface binding Reduced C3b binding Loss of FH cofactor and decay-accelerating activity	C3G IC-MPGN
FI	Homozygous Heterozygous	Decreased FI mediated C3b degradation	C3G IC-MPGN
C3	Heterozygous	C3mut – resistant to cleavage by C3bBb C3mut convertase – resistant to FH inactivation C3 binding with FI or FH	C3GN IC-MPGN
FB	Heterozygous/ homozygous	Alters C3-FB interaction	C3G IC-MPGN
THBD	Homozygous	Not tested	DDD
DGKE	Homozygous Heterozygous – unclear impact	Not complement mediated	MPGN

Genetic causes for C3G
Susceptibility variants

- Co
  14
- Bic
  - Ig
  - Ig
  - M
- Ge
  - M
  - S
  - D
- Cli
  - C



Iatropoulos et al, Mol Immunol 2016

The role of FHRs in C3G

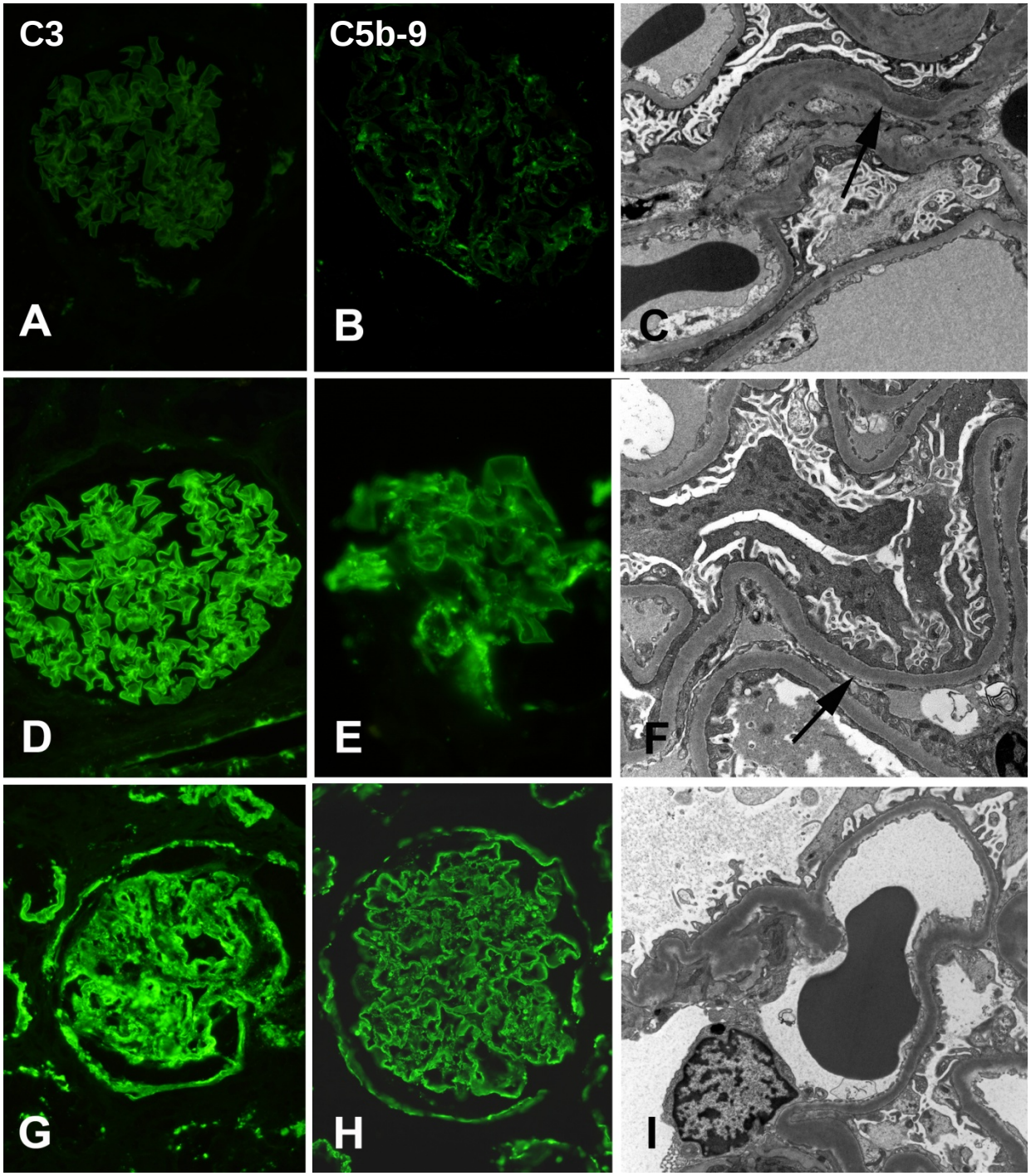
- 11 y/o male
- Macroscopic hematuria, nephrotic range proteinuria
  3 days post chicken pox immunization
- Kidney biopsy: DDD
- Treatment with prednisone and MMF
- Nevertheless, progression to ESKD within 3 months (PD)
- Kidney transplant (deceased donor) after 4 years
- Proteinuria recurs immediately post TX
- Complement system:
  - Low C3; normal C4
  - C3NeF positive
  - No mutations in FH, FHR5, FI, MCP/CD46, THBD/CD141, FB, C3

Serial biopsies of renal graft

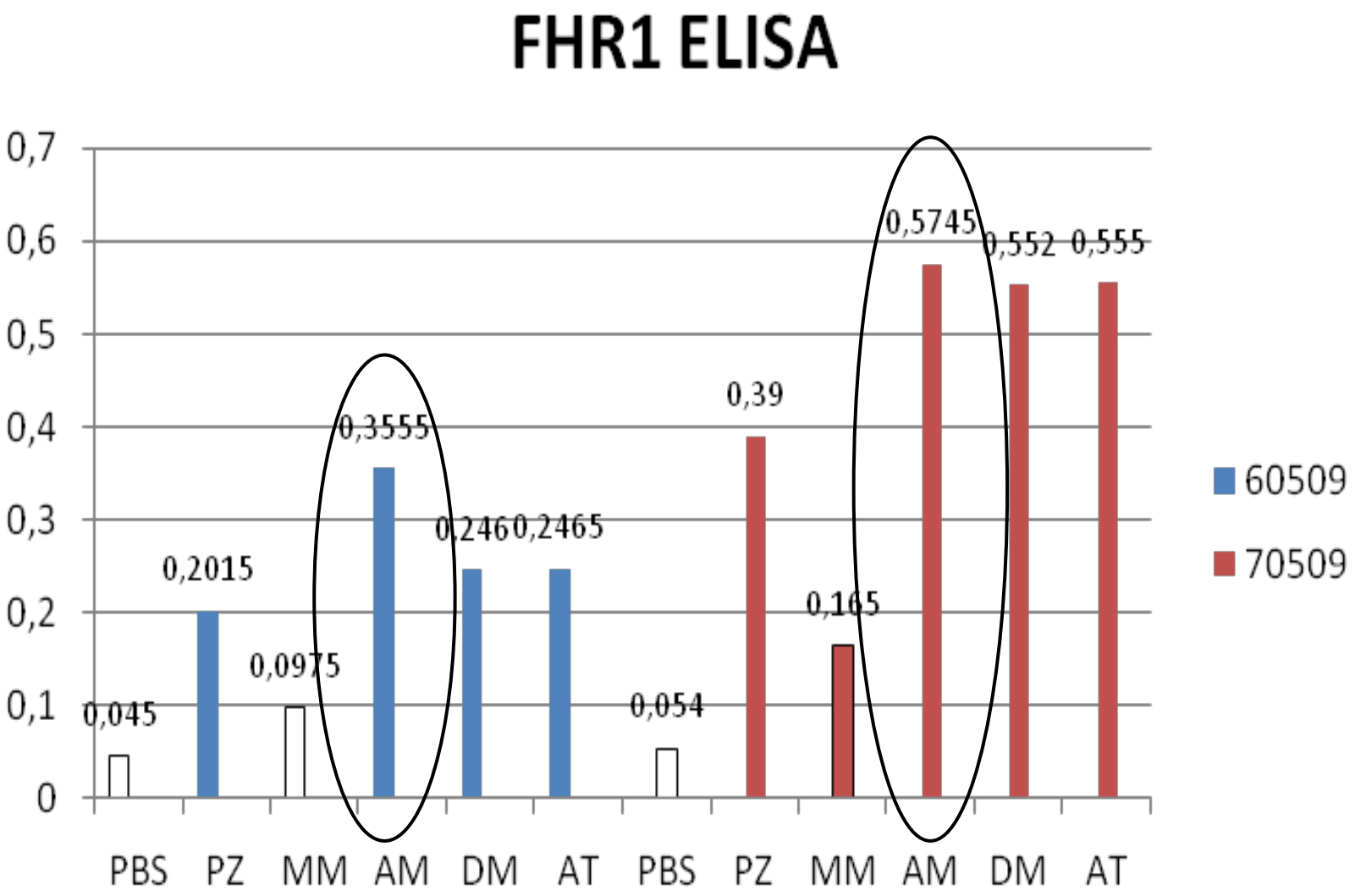
Post-TX day 6

Post-TX day 17

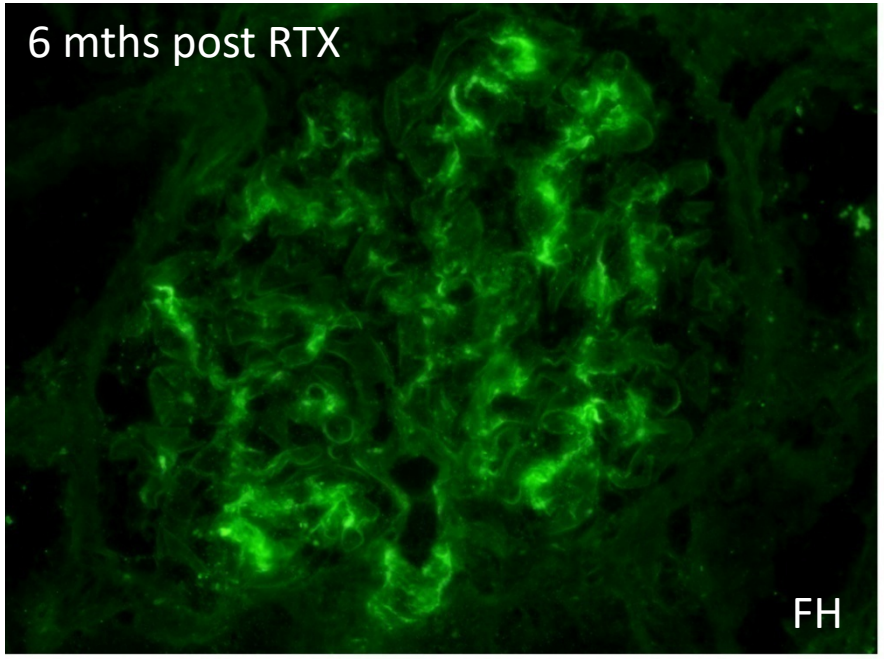
Post-TX day 180

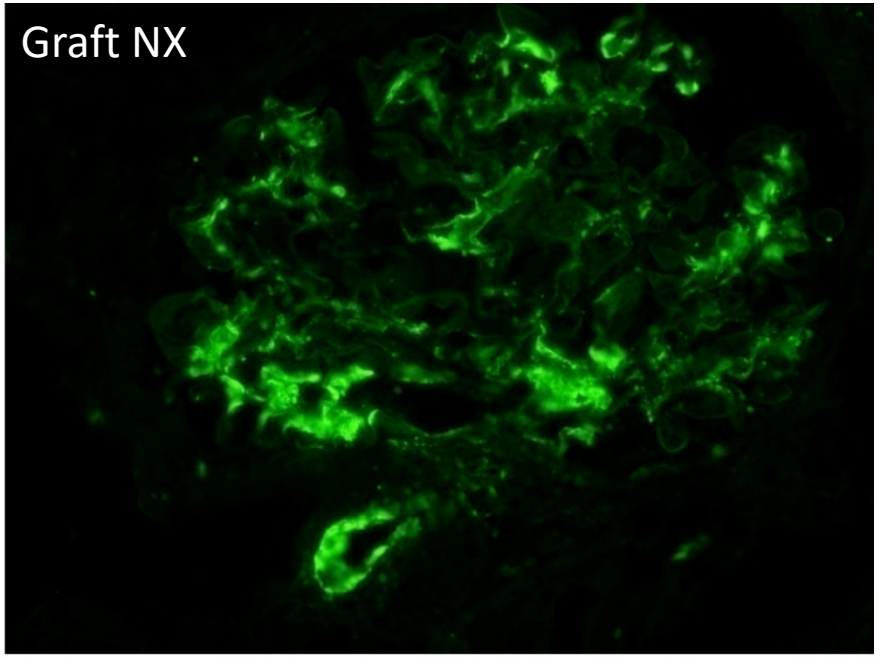


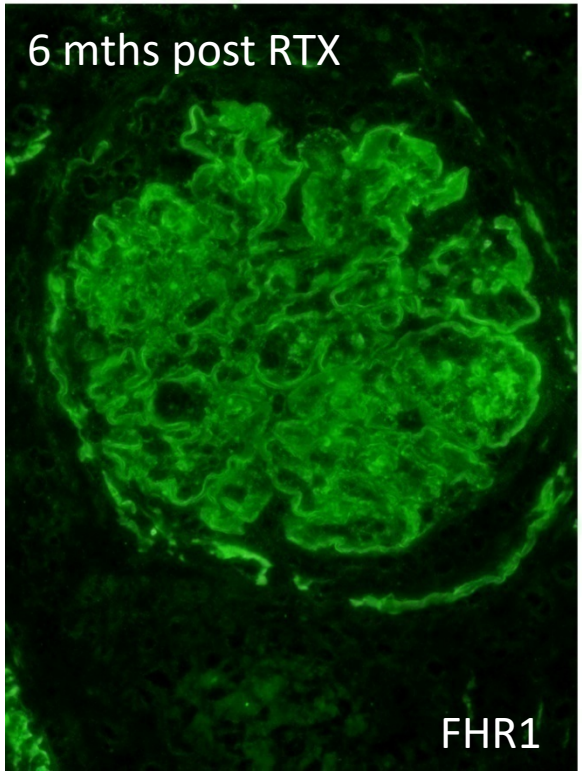
Three copies of *FHR1* - 150% plasma FHR1

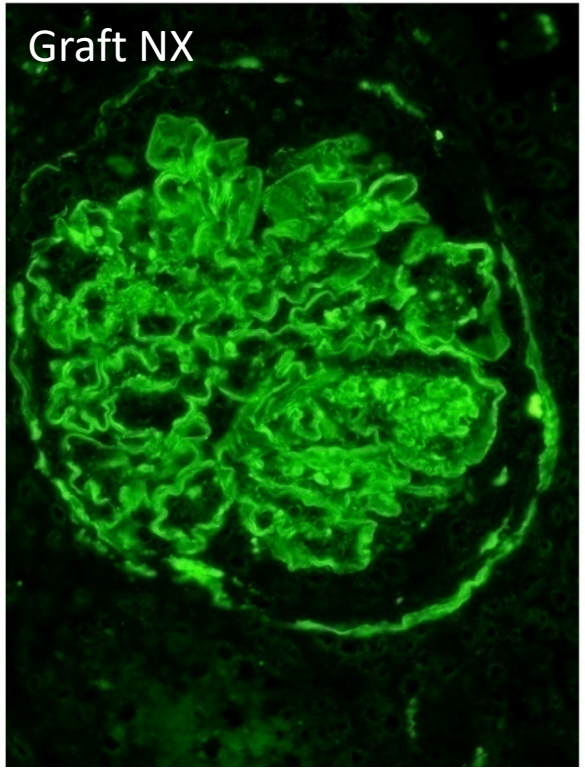


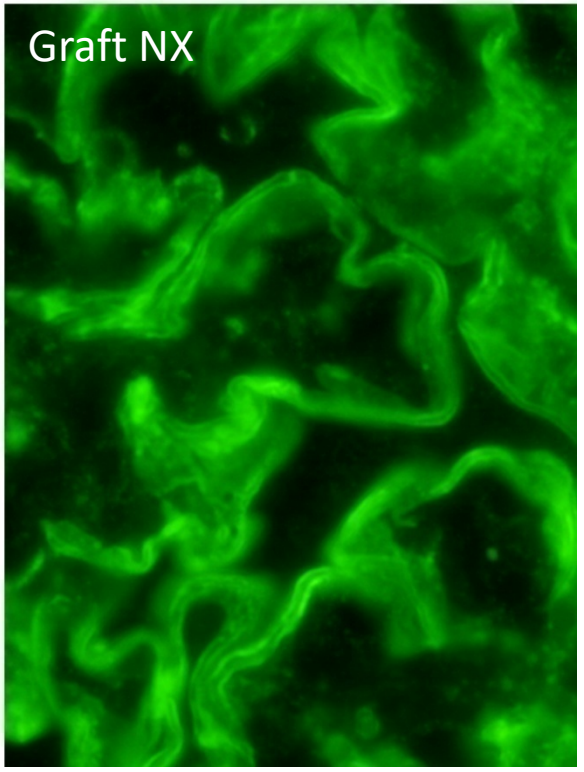
Localization of renal FH and CFHR1



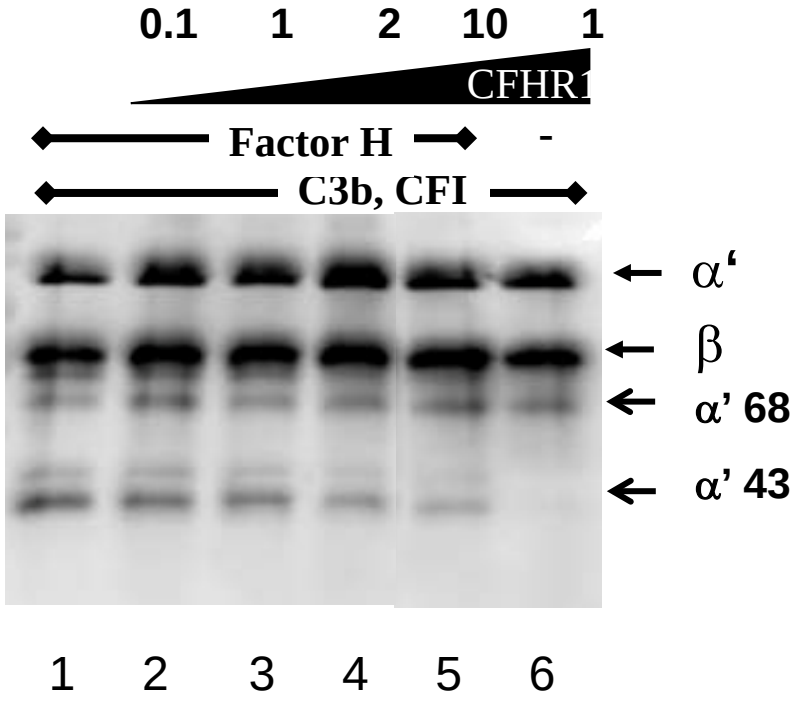


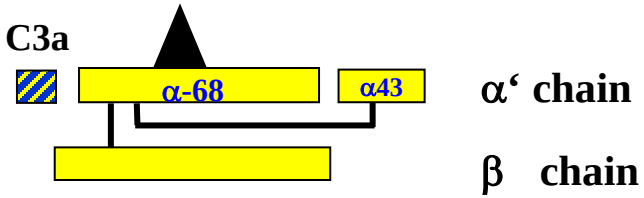


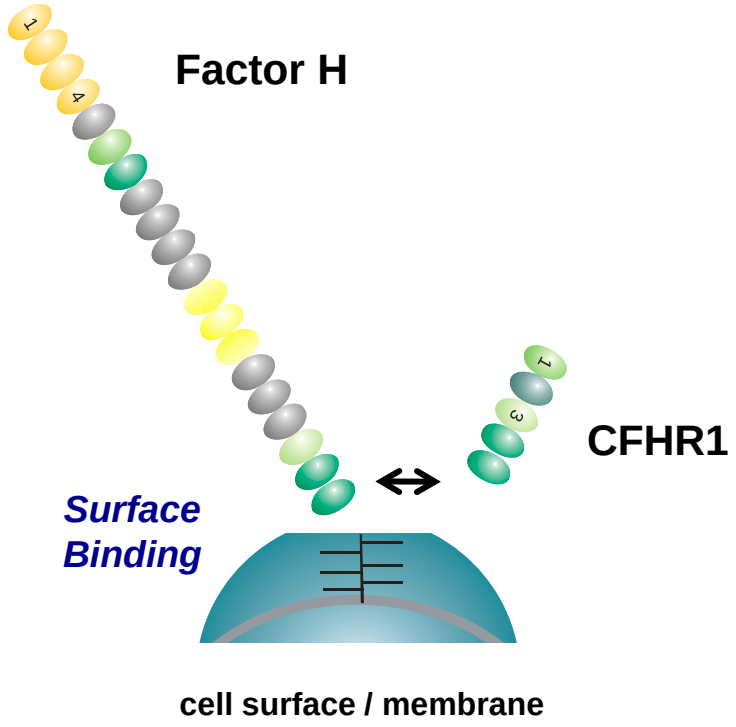




Competition between FHR1 and FH

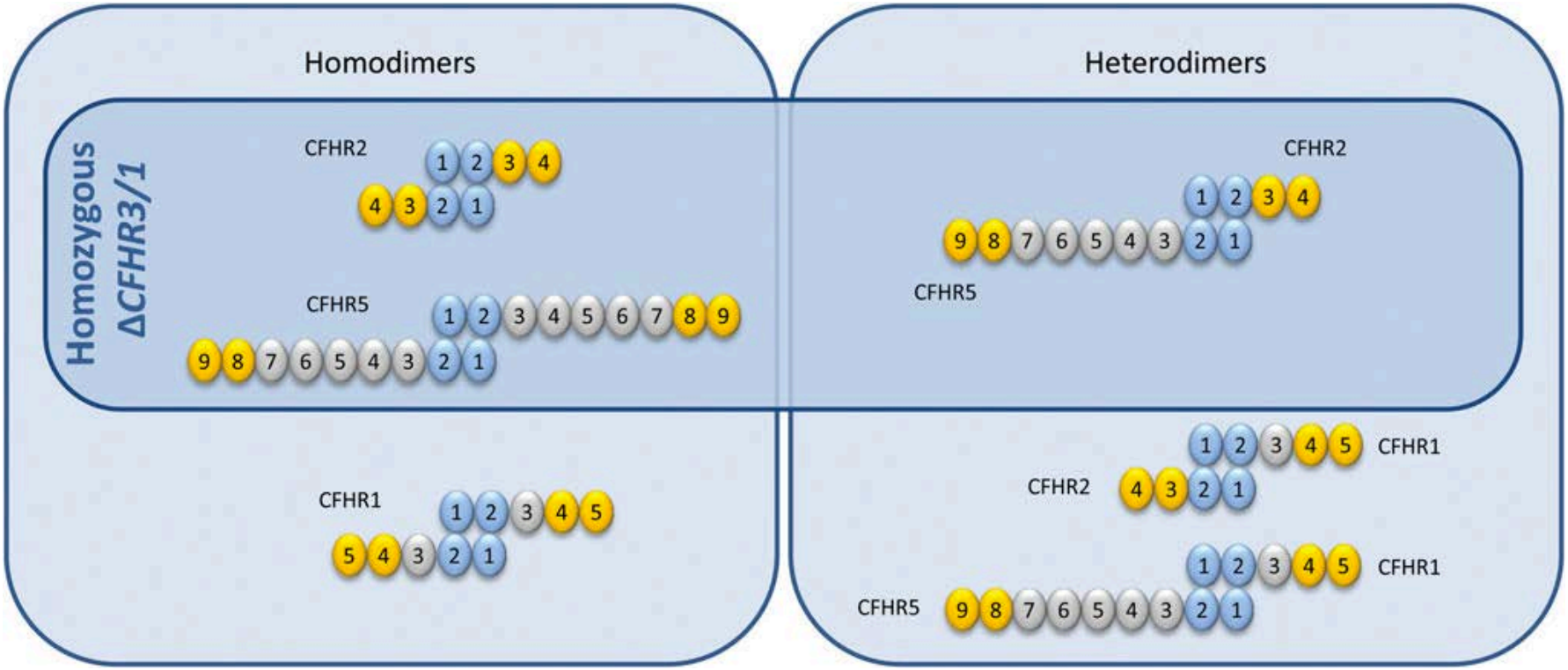






- FHR1 competes with FH for surface and C3b binding
- FHR1 lacks intrinsic cofactor activity
- FHR1 surplus impairs complement regulatory activity of FH on surfaces

FHRs 1, 2, 5 are prone to form dimers / oligomers



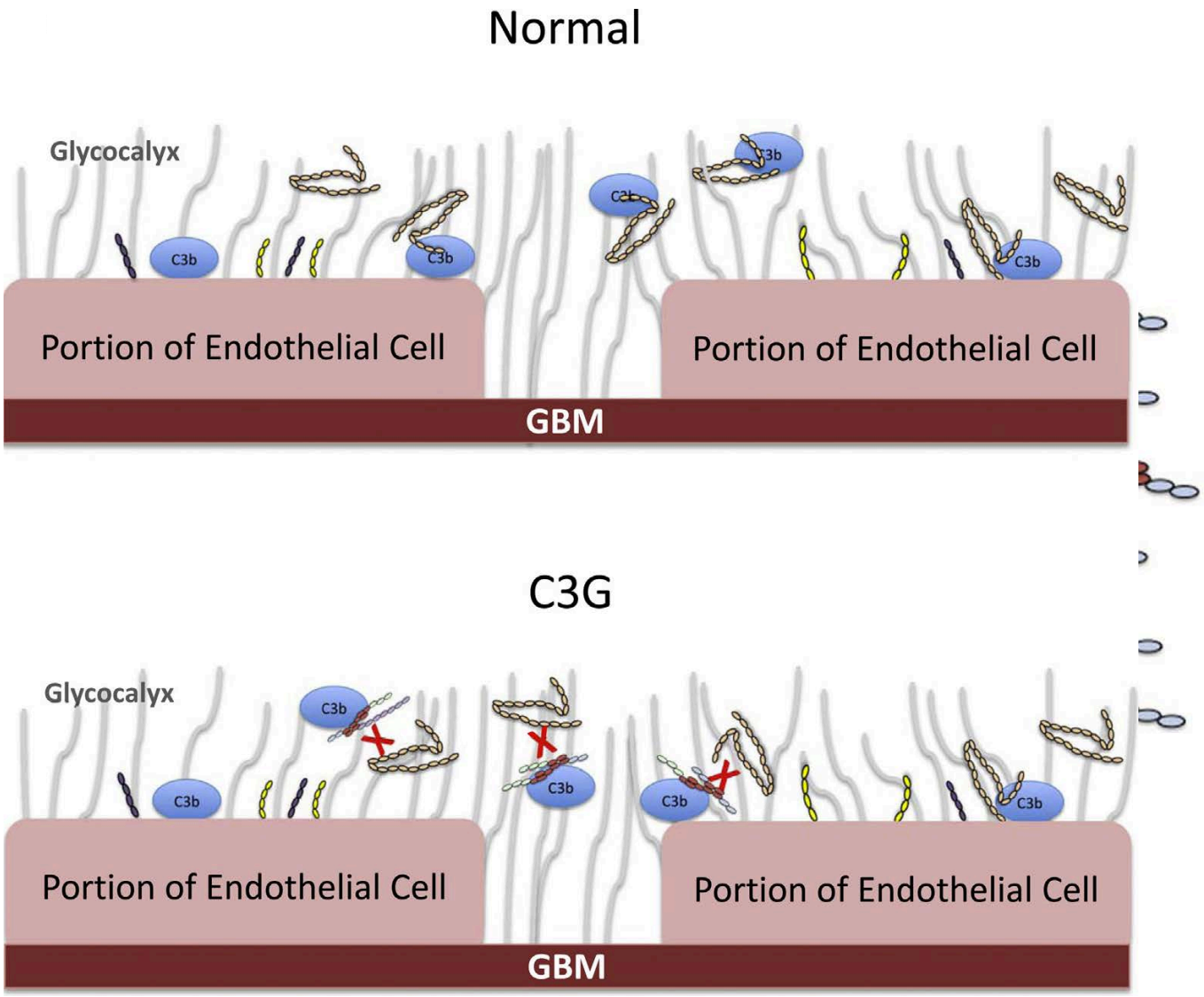
Goicoechea de Jorge et al, PNAS 2013; Barbour et al, Nephrol Dial Transplant 2016

Genetic causes for C3G – *FH deregulation*

Protein name	Protein structure	Phenotype	Comments
Normal proteins			
FHR1		NA	Homodimerizes and heterodimerizes with FHR2; competitive antagonist of factor H; C5 convertase inhibitor and terminal complement cascade blocker
FHR2		NA	Homodimerizes and heterodimerizes with FHR1; competitive antagonist of factor H; C3 convertase inhibitor
FHR3		NA	Exact function unknown
FHR5		NA	Homodimerizes; competitive antagonist of factor H; binds to extracellular matrix; complement amplifier and surface anchor for properdin
Fusion proteins			
$FHR2_{1,2}$–$FHR5_{1-9}$		DDD	Normal gene copies present in variant allele: *CFHR3*, *CFHR1* and *CFHR4*
$FHR5_{1,2}$–$FHR5_{1-9}$		C3GN	Normal gene copies present in variant allele: *CFHR3*, *CFHR1*, *CFHR4*, *CFHR2* and *CFHR5*
$FHR3_{1-2}$–$FHR1_{1-5}$		C3GN	Normal gene copies present in variant allele: *CFHR3*, *CFHR1*, *CFHR4*, *CFHR2* and *CFHR5*
$FHR1_{1,2}$–$FHR5_{1-9}$		C3GN and/or DDD	Normal gene copies present in variant allele: *CFHR3* and *CFHR5*
$FHR1_{1-4}$–$FHR1_{1-5}$		C3GN	Normal gene copies present in variant allele: *CFHR3*, *CFHR4*, *CFHR2* and *CFHR5*
$FHR5_{1,2}$–$FHR2_{1-4}$		C3GN	Normal gene copies present in variant allele: *CFHR3*, *CFHR1*, *CFHR4*, *CFHR2* and *CFHR5*

Smith et al, Nat Rev Nephrol 2019

FH deregulation in C3G via FHR multimers



Xiao et al, Mol Immunol 2016

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Diagnosis, treatment and outcome

Diagnostic workup for C3G patients

Global complement function	CH50, APH50
Complement activation	C3, C4, C3d
Terminal pathway activation	SC5b-9
Complement protein levels	CFH, CFI, CFB
Autoimmune forms	C3 Nephritic factor (C3NeF) CFH/CFB/C3b autoantibodies
Genetic forms	Mutations/CNVs in CFH, CFI, CFB, MCP/CD46, C3 CFHR–5 (MLPA)

Other
Nephritic factors

Genetic variants in
FHR locus

Riedl / Licht, Pediatr Nephrol 2017

How to treat C3G patients?

A. There is no treatment for C3G.

B. Transplant is a definitive treatment for C3G.

C. Current treatment recommendations for C3G mainly rely on case reports / small case series.

D. Current treatment recommendations for C3G build on RTCs.

Treatment
Immunosuppression

Steroids:

- Benefit of long-term (6-12 months) low-dose steroids in patients with MPGN and C3G with respect to proteinuria and renal function.

Mycophenolate mofetil (MMF):

- Benefit of use alone or in combination with steroids in patients with primary MPGN with respect to proteinuria and renal function.
- 5/9 children with MPGN treated with steroids + MMF for 40 months had CR/PR (of note, all patients with low C3 failed).
- 42/97 C3G patients (81 C3GN; 16 DDD) treated with steroids + MMF had remission of proteinuria and prevention of renal failure compared to other immunosuppression and eculizumab regardless of presence of complement mutations and/or autoantibodies.
- 22/30 C3G patients treated with steroids + MMF and f/u of 22 months had CR/PR. 50% of patients with MMF taper relapsed.

KDIGO expert opinion:

- MMF is beneficial in patients with IC-MPGN and C3G.
- Following steroid taper over 6-12 months, MMF monotherapy is continued for 12-18 months.

Goodship et al, Kindey Int 2017

Treatment - *Plasma*

Reference	Diagnosis	Patients	Treatment	Outcome
Pérez-Sáez et al, Transplant Proc 2011	MPGN I	1 adult	Rituximab + PE	**Partial response** Deterioration of renal function Proteinuria
Kmar et al, Clin Nephrol 2011	DDD	1 adolescent	Steroids + Cyclo + PE	**Complete remission** Normal kidney function Normal AP actvity
Reference	**Diagnosis**	**Patients**	**Treatment**	**Outcome**
Haeffner et al, Pediatr Nephrol 2015	C3G (Mut. neg., C3Nef pos.)	4 children	PE + Steroids + MMF *Eculizumab*	**Partial response** Proteinuria improved (4/4) / normalized (2/4) eGFR normalized (4/4) C3 improved (2/4) TCC improved (2/4) (2 were always normal) C3NeF negative (4/4)
McGinley et al, Nephron 1985	MPGN I / III DDD	4 adults	PE	MPGN: Improved proteinuria Stabilization of kidney function DDD: Improved kidney function in 1 patient
Oberkircher et al, Transplant Proc 1988	DDD (TX)	1 child	PE	**Partial response** Improved renal outcome
Kurtz and Schlueter, J Clin Apher 2002	DDD (TX)	1 adolescent	PE	Stabilized kidney function Treatment discontinued – graft loss

Treatment
Eculizumab

	Patients	Complete response	Partial response	No response	Treatment response correlated with
Vivarelli, 2014	13 patients C3G / DDD Native / TX kidneys C3Nef / genetic	10 (77%)	1 (8%)	2 (15%)	- Elevated sC5b-9 - Shorter disease duration
LeQuintrec, 2018	26 patients 13/26 pediatric 14 months	6 (23%)	6 (23%)	14 (54%)	- Rapidly progressive disease & intense extracapillary proliferation - Complement AP mutations / autoantibodies made no difference
Ruggenenti, 2019 (EAGLE study: 2 x 48 weeks ecu with 12 weeks washout; aimimg at improvement in proteinuria @ 24 / 48 weeks)	10 patients 6 MPGN, 4 C3G All normal renal function, high proteinuria and high sC5b-9	0	3 (30%)	7 (70%) – improvement during first period, but benefit lost during washout	

Eculizumab in IC-MPGN/C3G currently recommended as rescue therapy, only.
Best response in patients with recent onset of disease, intense inflammation, and high C5b-9 levels.

Outcome

Diagnosis	Patients	Follow Up	Outcome	Reference
MPGN	*50 children*	*11 years (median)*	*50% ESKD in 10-15 years*	*Schwertz et al, Pediatr Allergy Immunol 2001*
C3GN	12	26.4 months	Stable kidney function	Sethi et al, Kidney Int 2012
DDD vs C3GN	80	28 months	47% DDD and 23% C3GN progress to ESKD	Medjeral-Thomas et al, Clin J Am Soc Nephrol 2014
MPGN / C3GN	134	10 years	63.5% renal survival (MPGN, DDD, C3GN show no difference)	Servais et al, Kidney Int 2012
IC-MPGN / C3G	165	4 years	100% show preserved kidney function	Kirpalani et al, Kidney Int Reports 2020
		10 years	80% do not meet composite outcome (eGFR <30; 50% eGFR reduction; initiation of RRT)	

Negative outcome predictors:

- Age at presentation
- eGFR
- Proteinuria
- Hypertension
- Kidney biopsy: glomerular crescents

Outcome
Renal transplant

- 32.4% risk of graft loss from recurrence at 5 years post TX in children with MPGN.
  (Van Stralen et al, Nephrol Dial Transplant 2013 – ESPN/ERA-EDTA registry)
- Greater risk of graft loss from recurrence in children with DDD.
  (Braun et al, J Am Soc Nephrol 2005 – NAPRTCS registry)
- 66.7% risk of graft loss from recurrence in C3G patients
  with median time to graft failure of 6.4 years.
  (Zand et al, J Am Soc Nephrol 2014)
- Cohort of n=35 C3G patients:
  Recurrence risk of 43% in MPGN, 55% in DDD, and 60% in C3GN patients.
  (Servais et al, Kidney Int 2012 – French cohort)
- Cohort of n=13 C3G patients (6 DDD; 7 C3GN):
  - 69% overall graft survival at 5 years.
  - All 6 DDD recurred, and 3 (50%) failed due to recurrence.
  - 4/7 (57%) C3GN recurred, and 3/4 (75%) failed due to recurrence.
  (Medjeral-Thomas et al, J Am Soc Nephrol 2014 – English cohort)

High risk of disease recurrence and graft failure due to recurrence
in patients with MPGN < DDD < C3GN.

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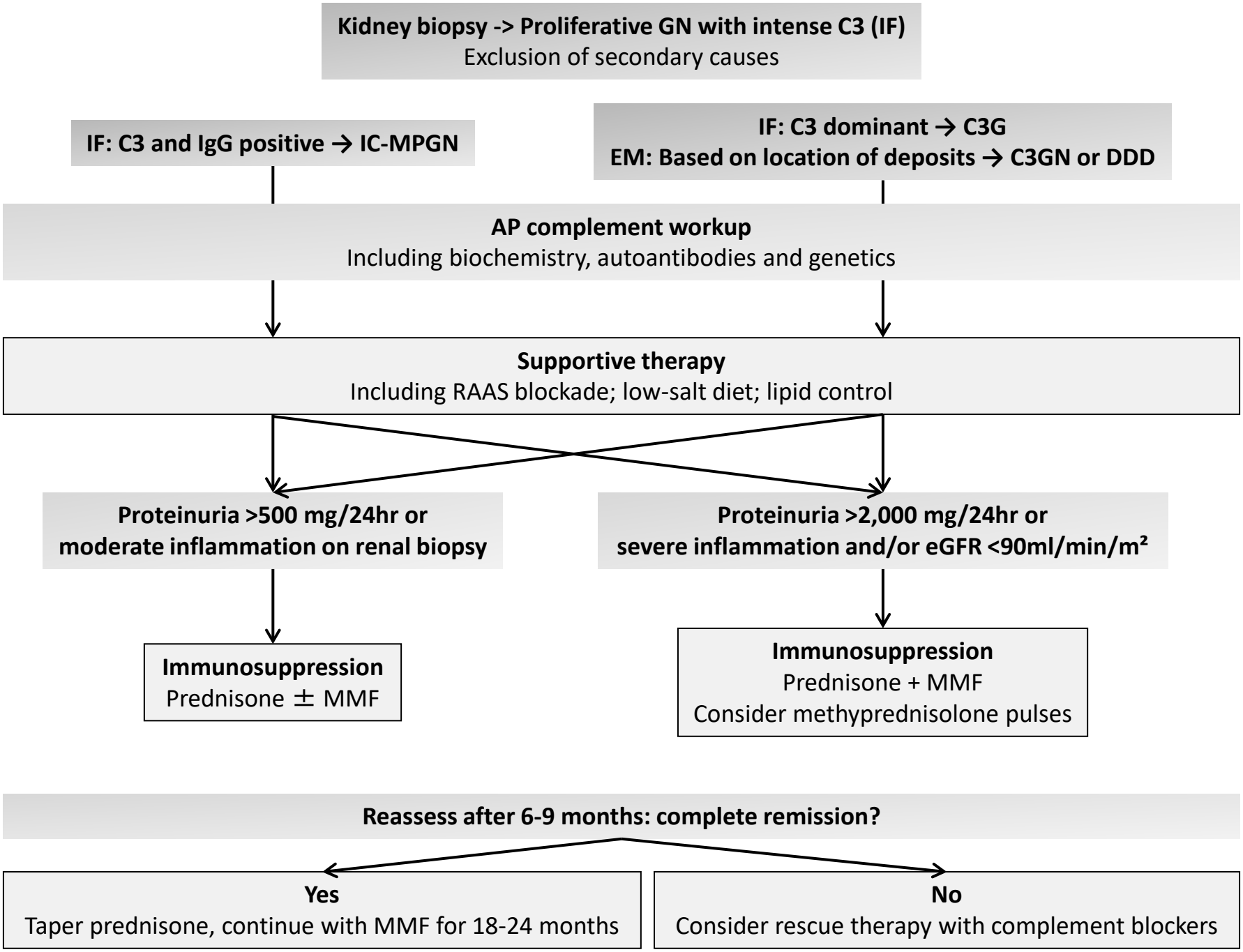
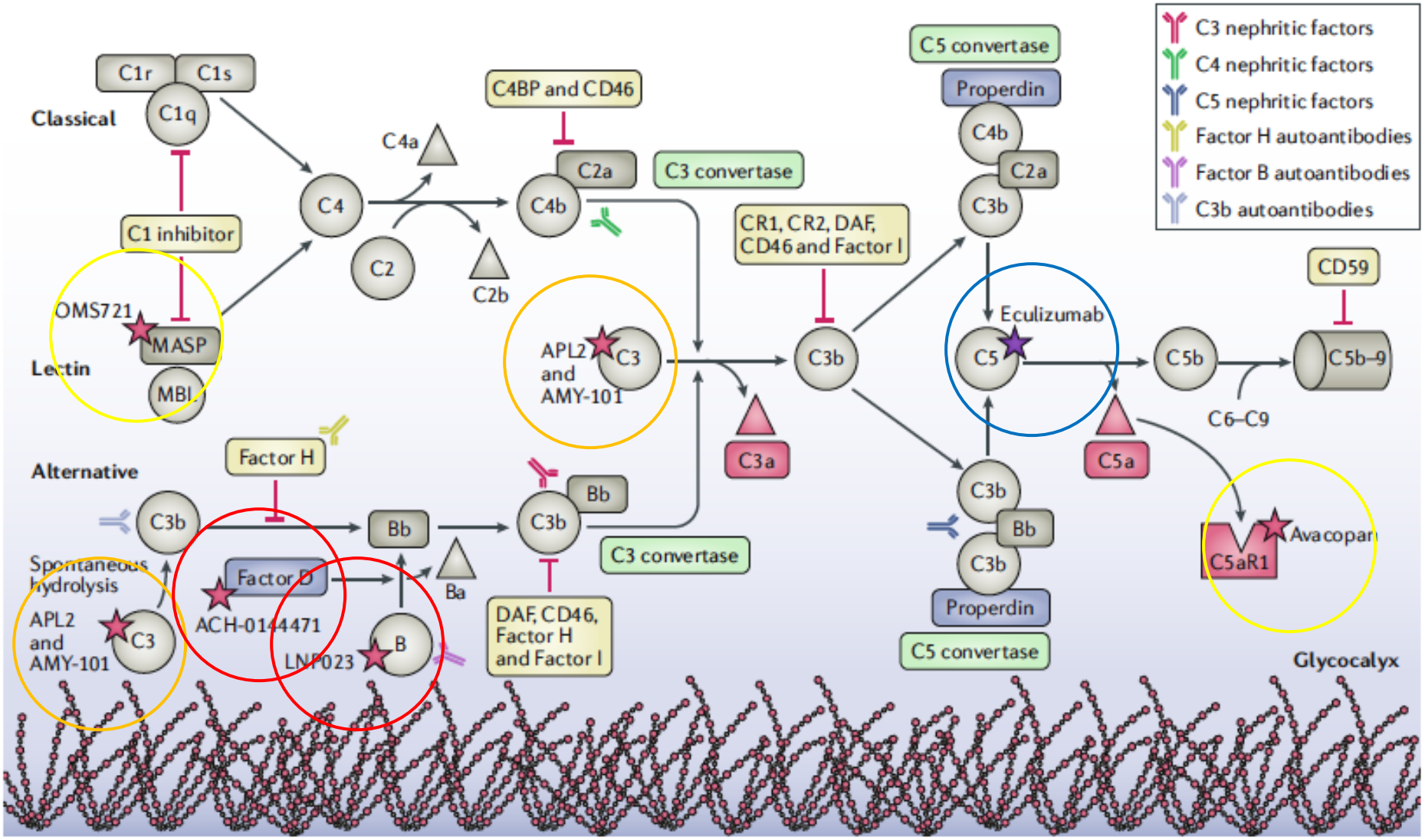
Summary and perspectives


Kidney biopsy -> Proliferative GN with intense C3 (IF)
Exclusion of secondary causes
IF: C3 and IgG positive → IC-MPGN
IF: C3 dominant → C3G
EM: Based on location of deposits → C3GN or DDD
AP complement workup
Including biochemistry, autoantibodies and genetics
Supportive therapy
Including RAAS blockade; low-salt diet; lipid control
Proteinuria >500 mg/24hr or
moderate inflammation on renal biopsy
Proteinuria >2,000 mg/24hr or
severe inflammation and/or eGFR <90ml/min/m²
Immunosuppression
Prednisone ± MMF
Immunosuppression
Prednisone + MMF
Consider methyprednisolone pulses
Reassess after 6-9 months: complete remission?
Yes
Taper prednisone, continue with MMF for 18-24 months
No
Consider rescue therapy with complement blockers



Smith et al, Nat Rev Nephrol 2019

Next Webinars

ERKNet/ERA-EDTA Advanced Webinars on Rare Kidney Disorders

Date: **16 Feb 2021**

Speaker: **Frank Bridoux**

Topic**: Paraprotein associated disease**

ESPN/ERKNet & ERA-EDTA Webinars on Rare Kidney Disorders

Date: **02 March 2021**

Speaker: **Hans Joachim Anders & Steven Marks**

Topic: **Lupus nephritis in children & adults**

ERKNet/ERA-EDTA Advanced Webinars on Rare Kidney Disorders

Date: **16 March 2021**

Speaker: **Olivier Devuyst**

Topic: **Uric acid disorders**

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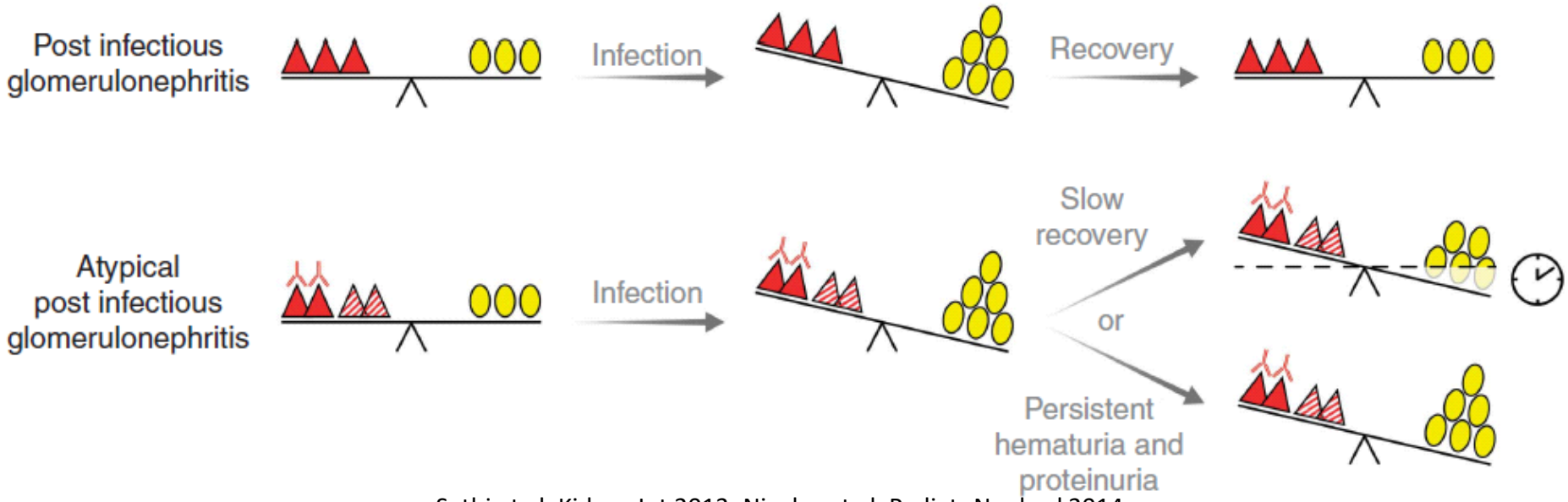
Atypical PIGN - a role for complement defects?

	Postinfectious glomerulonephritis (PIGN)	'Atypical' postinfectious glomerulonephritis (aPIGN)	C3 glomerulonephritis (C3GN)
LM	Diffuse proliferative, less commonly mesangial proliferative, or crescentic	Diffuse proliferative, less commonly mesangial proliferative, or crescentic	Membranoproliferative and less commonly mesangial proliferative
IF	Bright mesangial and capillary wall C3, usually with Igs (garland pattern)	Bright mesangial and capillary wall C3, usually without Igs. If present IgG (trace to 1+)	Bright mesangial and capillary wall C3, usually without Igs
EM	Numerous subepithelial humps, few mesangial, and subendothelial deposits	Numerous subepithelial humps, many mesangial and subendothelial deposits, and ±intramembranous deposits	Many mesangial and subendothelial deposits, ±few intramembranous, and subepithelial humps

Abbreviations: EM, electron microscopy; IF, immunofluorescence; Ig, immunoglobulin; LM, light microscopy.

9	No mutations	No mutations	Negative	9% Abnormal	77%	Positive (both assays)	ND
10	c.1699A>G, p.Arg567Gly	No mutations	Negative	0%, Normal	0% Abnormal	Positive (both assays)	2.03 mg/l
11	No mutations	No mutations	Negative	0%, Normal	130%	Positive (C3CSAP)	0.21 mg/l



Sethi et al, Kidney Int 2013; Nicolas et al, Pediatr Nephrol 2014

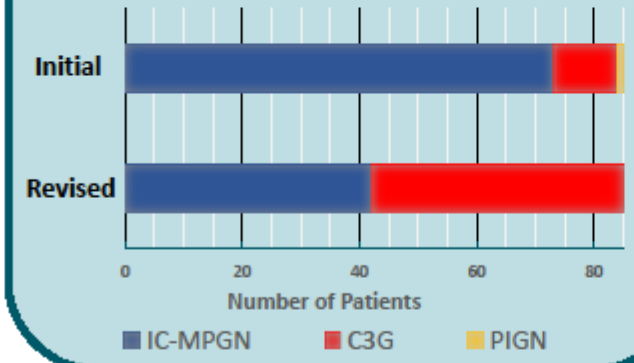
Long-Term Outcomes of C3 Glomerulopathy and Immune-Complex MPGN in Children

IC-MPGN & C3G cohort

- **165 patients**
- 17 hospitals
- 3 countries
- Largest pediatric cohort

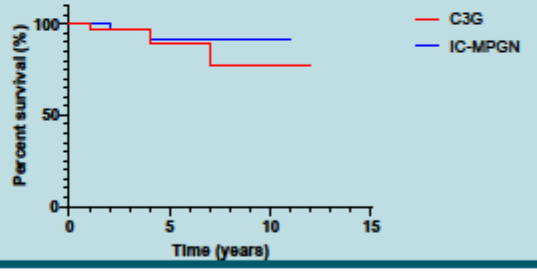
Reclassification

- 85 biopsy reports available
- 42% initially diagnosed as 'MPGN' reclassified as C3G



Clinical outcomes in IC-MPGN vs. C3G

Survival without eGFR <30 mL/min/1.73m^2
50% reduction in eGFR
or kidney replacement therapy

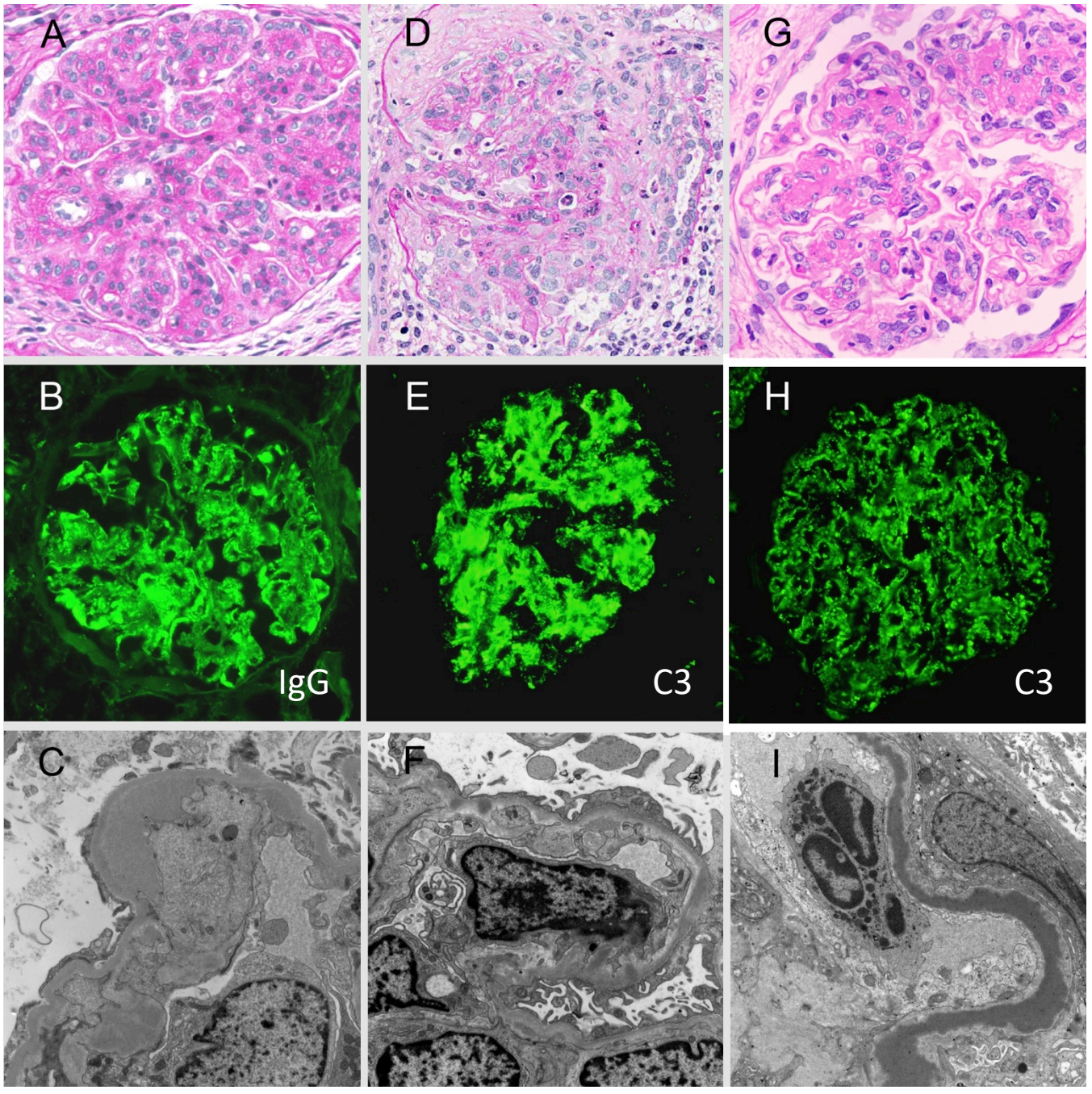


CONCLUSION:

Many patients initially diagnosed as MPGN would meet criteria for C3G.
Longer follow-up may reveal a worse kidney prognosis in C3G vs. IC-MPGN.

KI REPORTS
KIReports.org

Kirpalani et al, 2020



Riedl / Licht, Pediatr Nephrol 2017