I have my cysts with a bit of sugar, please

Detlef Bockenhauer

Great Ormond Street Hospital for Children NHS Trust

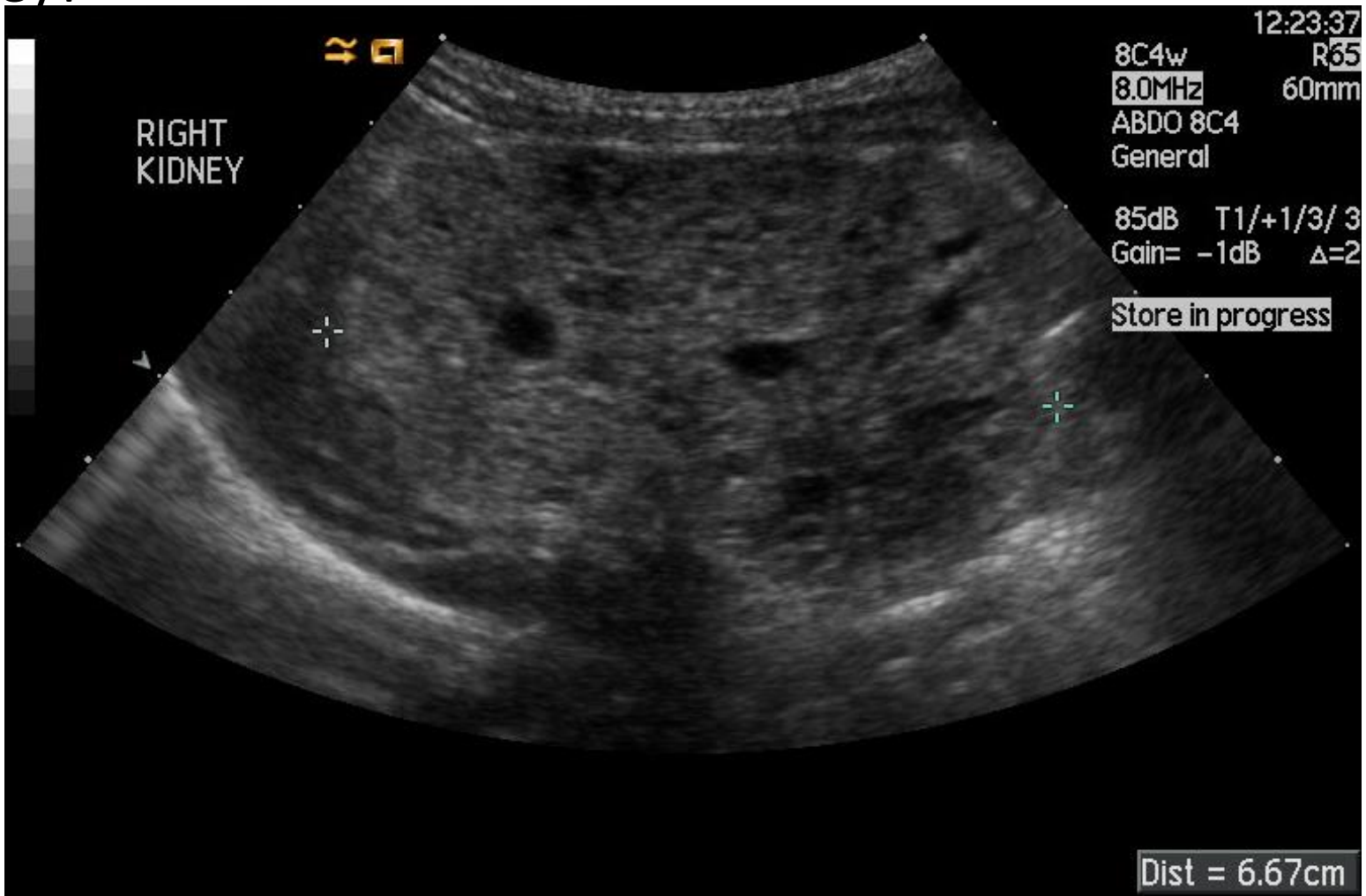
UCL

A patient

- A 1-day old neonate is admitted to the renal ward because of impaired kidney function and hypertension
- 1st child of unrelated parents, no family HX of kidney disease
- Pregnancy complicated by polyhydramnios with 33-wk scan showing cystic kidneys.
- Noted to be hypoglycaemic immediately after birth
- Unremarkable examination, kidneys not palpated
- Weight: 4200g, length: 51 cm, BP: up to 138 mmHg systolic

Investigations

- Blood: creatinine 109 mcmol/l, glucose: 1.7 mmol/l with insulin of 11.3 mU/l



Diagnosis?

- Suspected ARPKD
- Hyperinsulinaemic hypoglycaemia

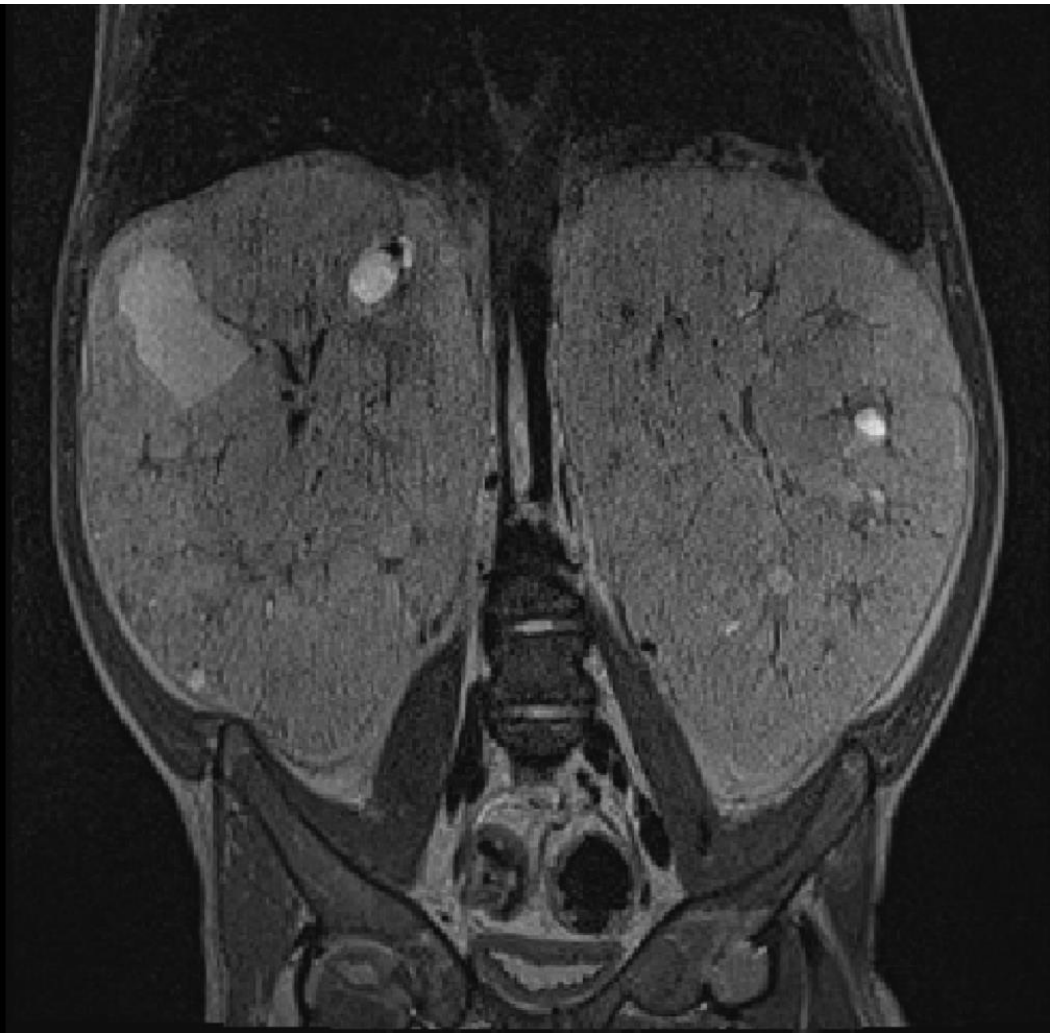
- Treatment:
- HI: Diazoxide, Chlorothiazide
- PKD: Amlodipine, Propranolol

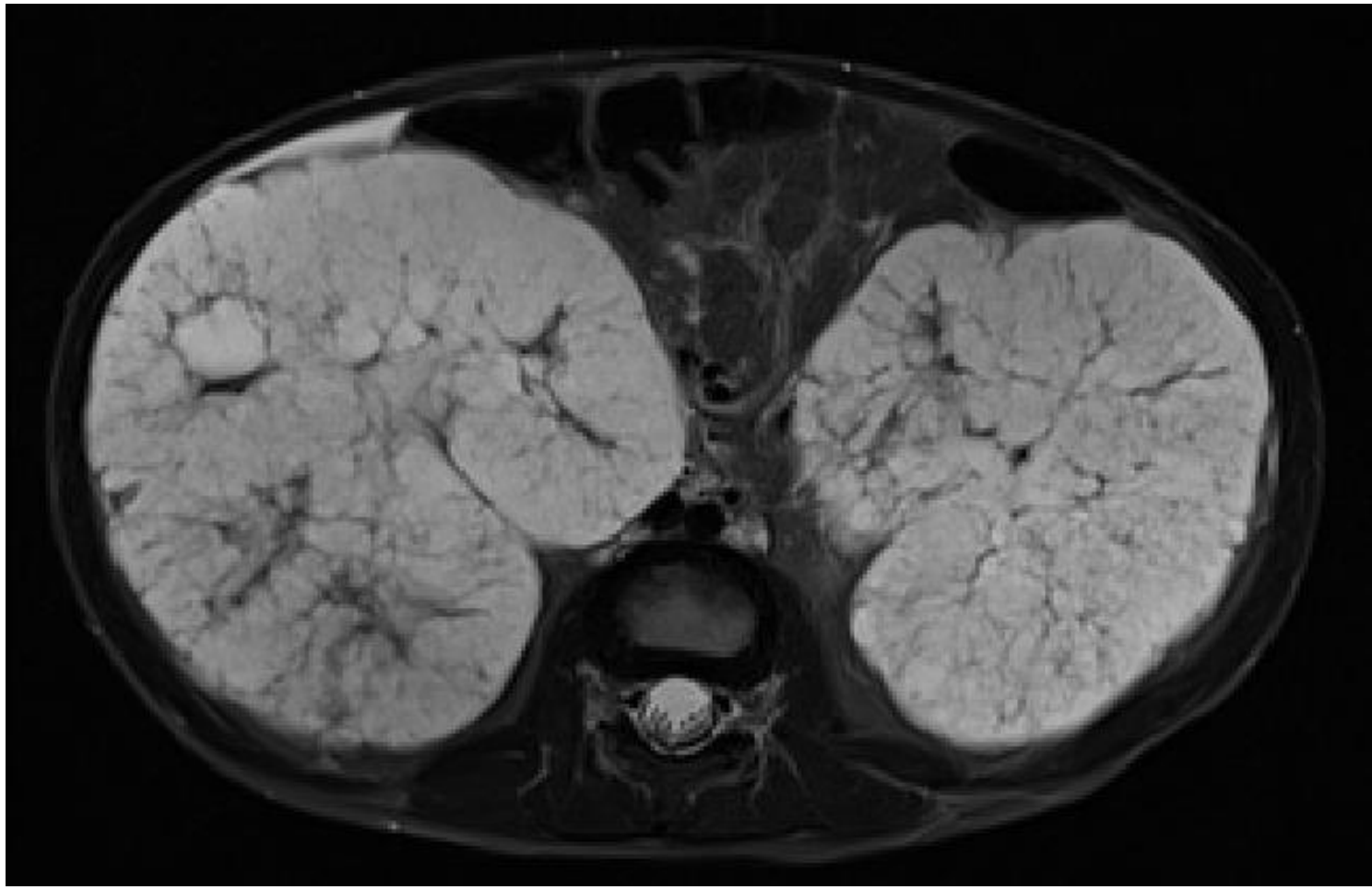
- ?unlucky coincidence of 2 rare diseases in one patient

Further course

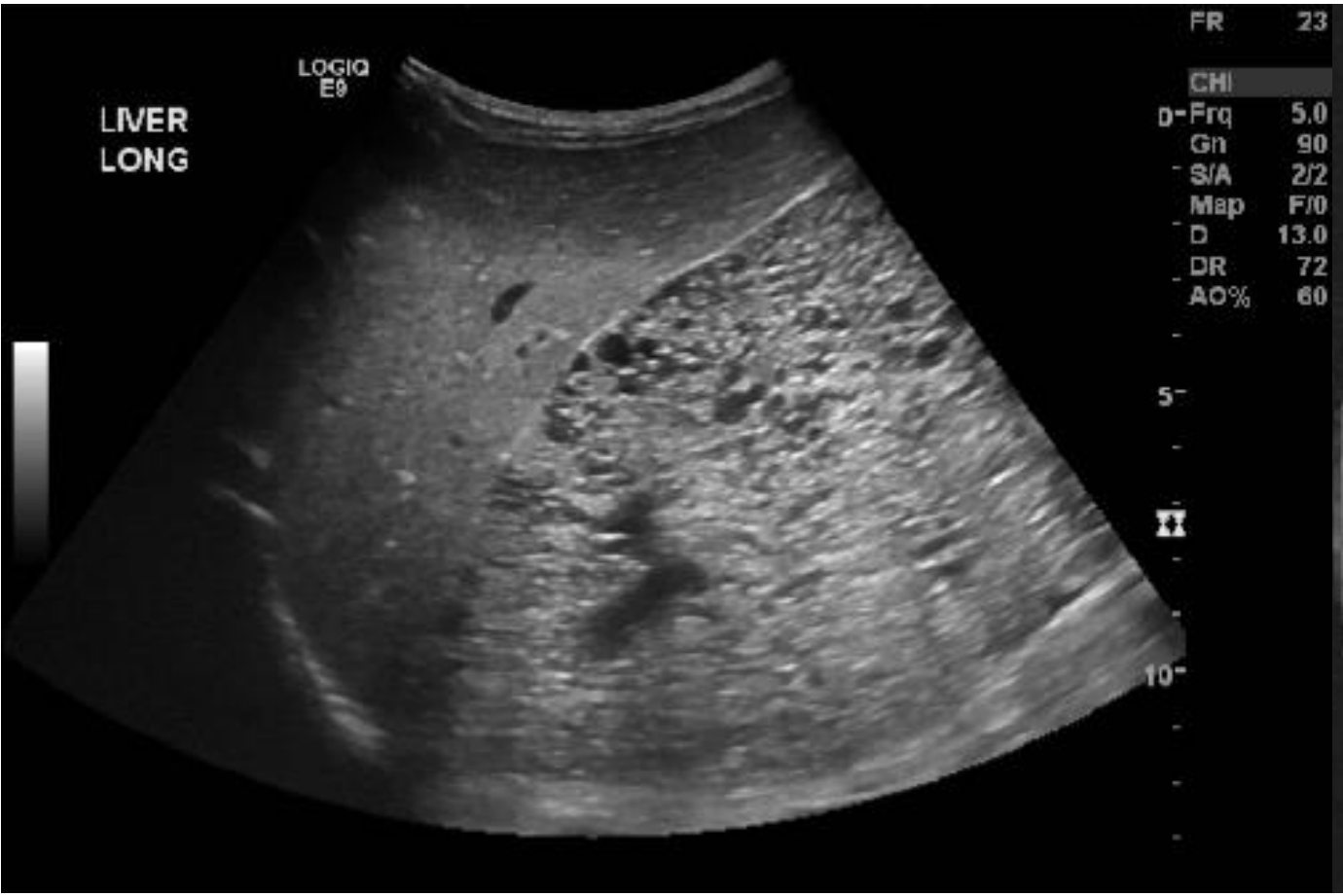
- Evidence of portal hypertension on endoscopy
- Progressive CKD age with nephromegaly

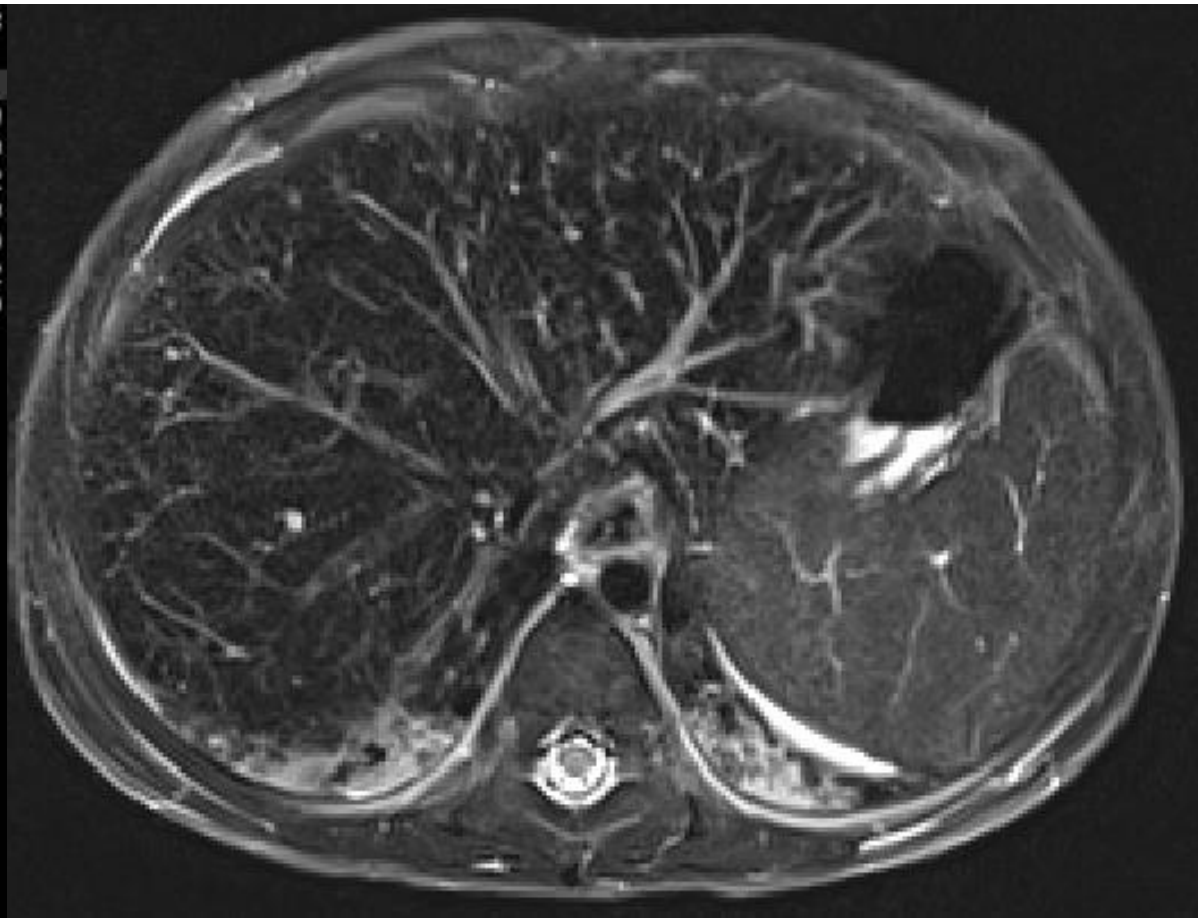
MRI kidneys



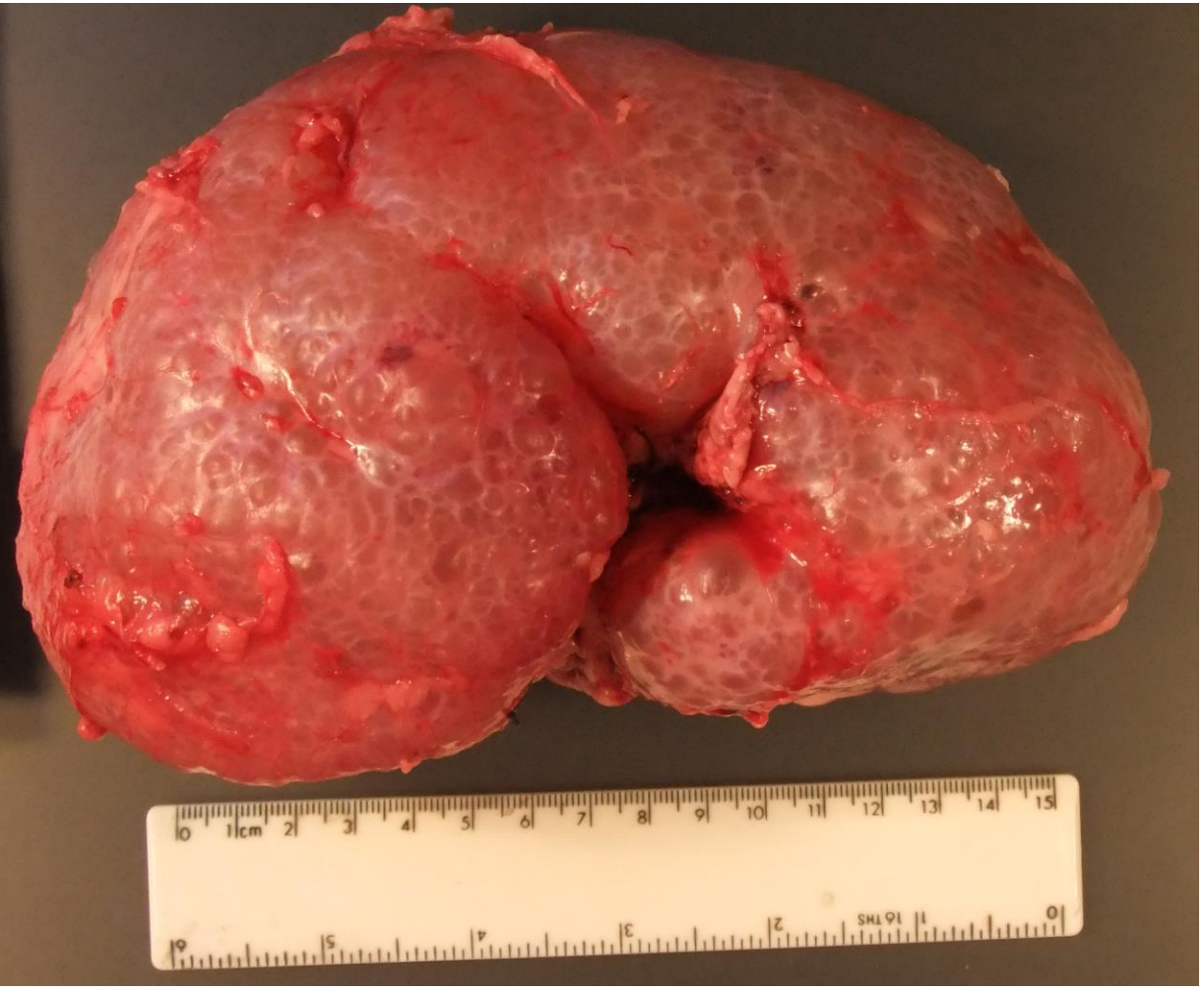


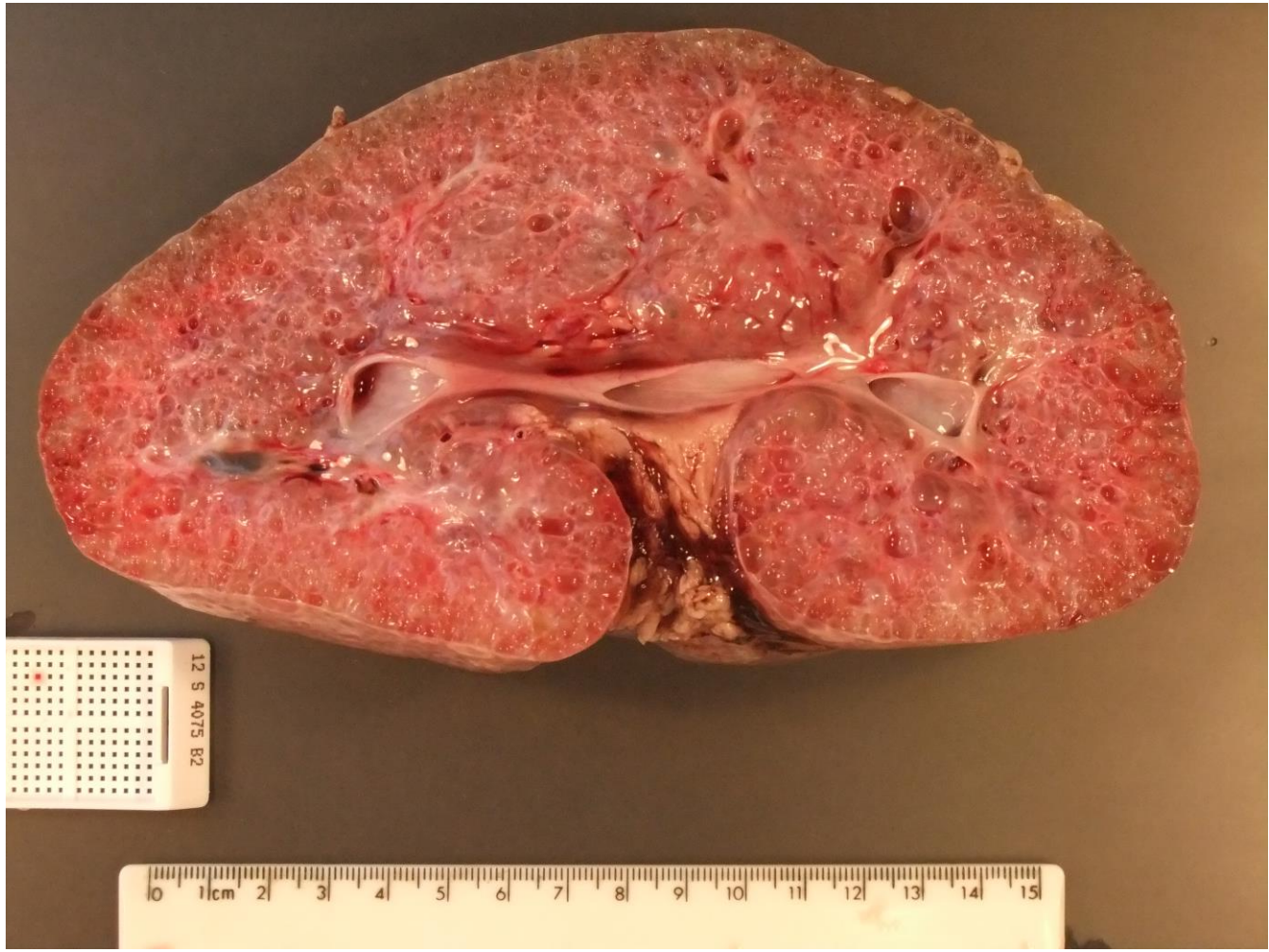
Liver imaging



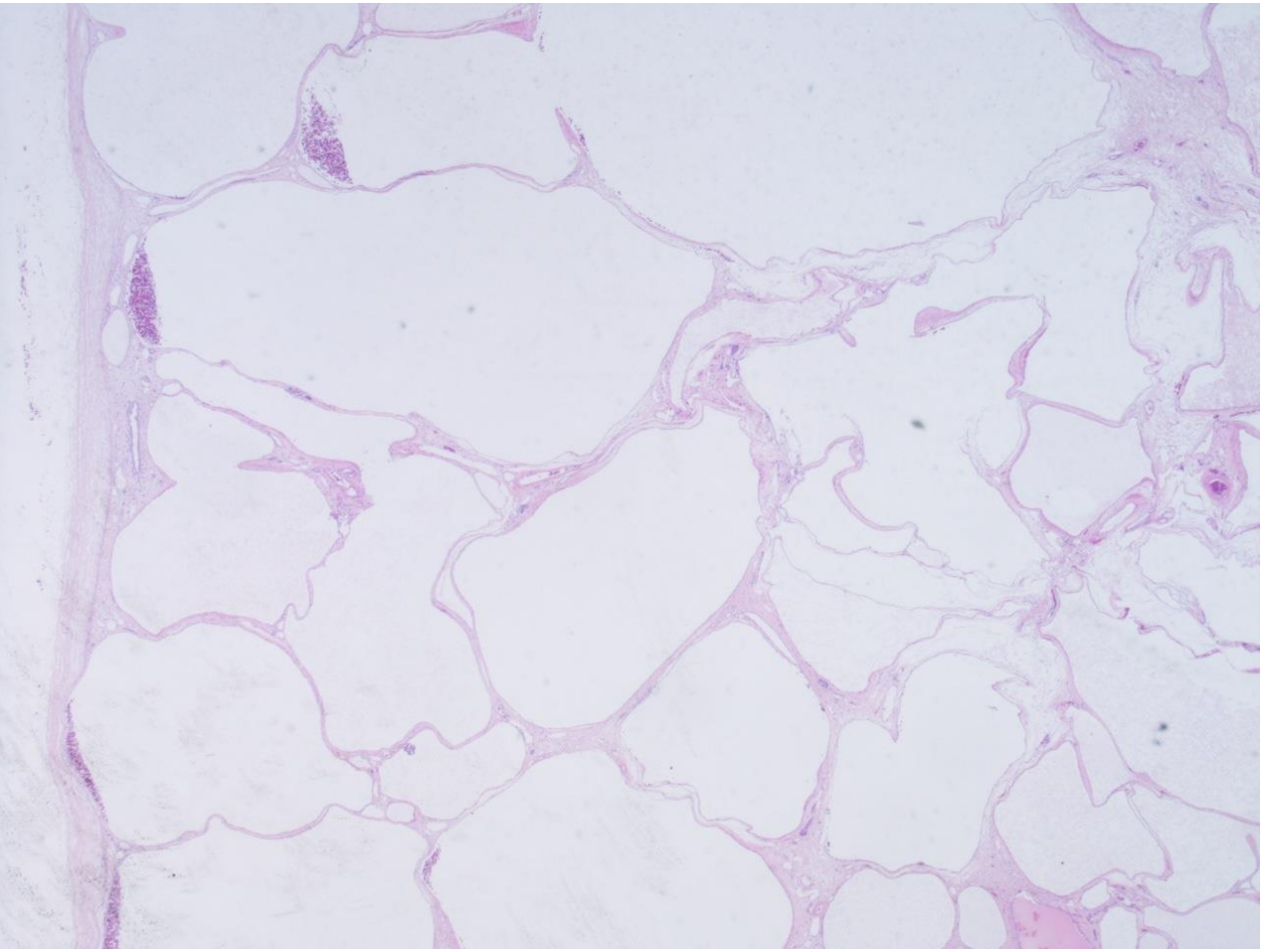


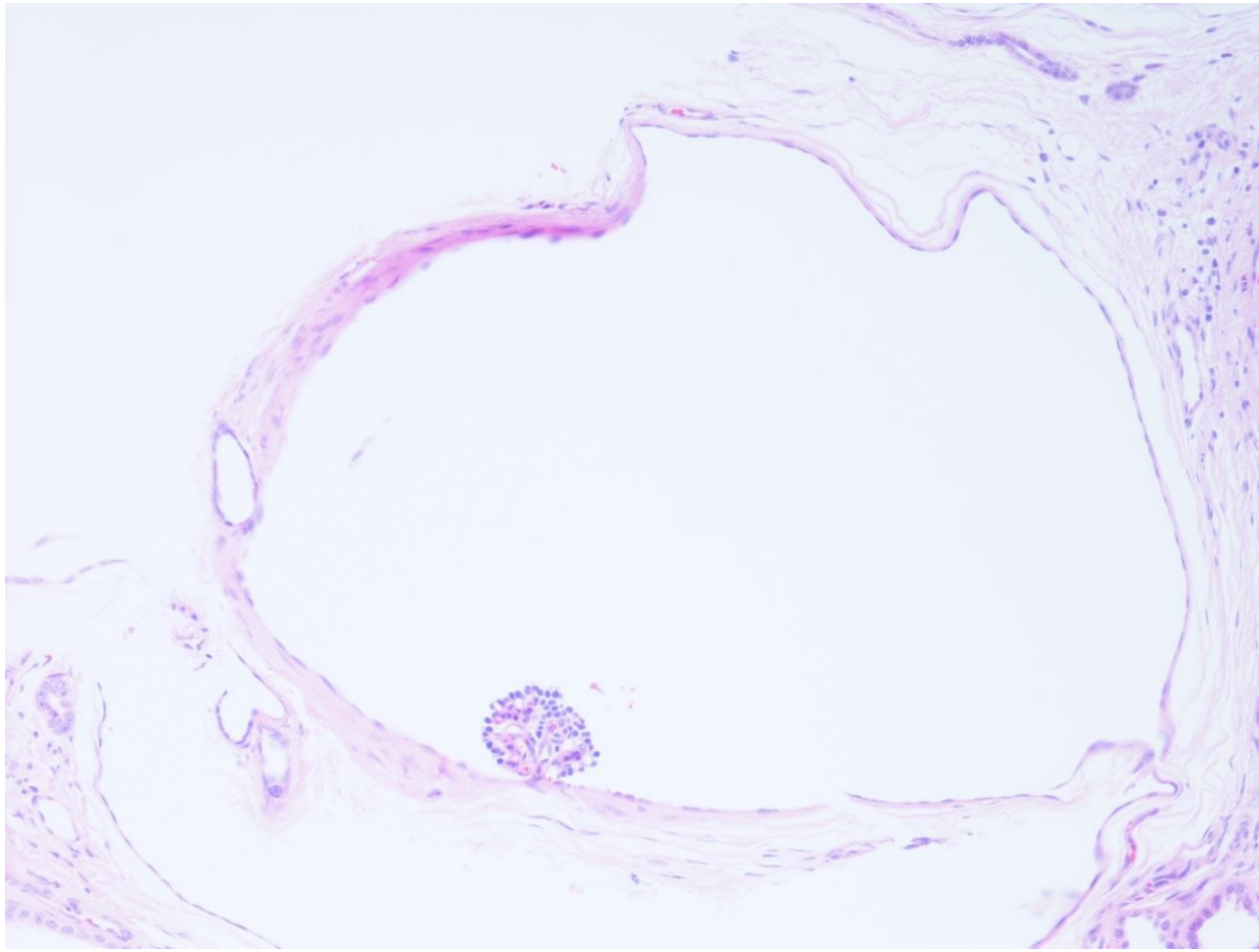
Nephrectomies at time of transplant





Histology

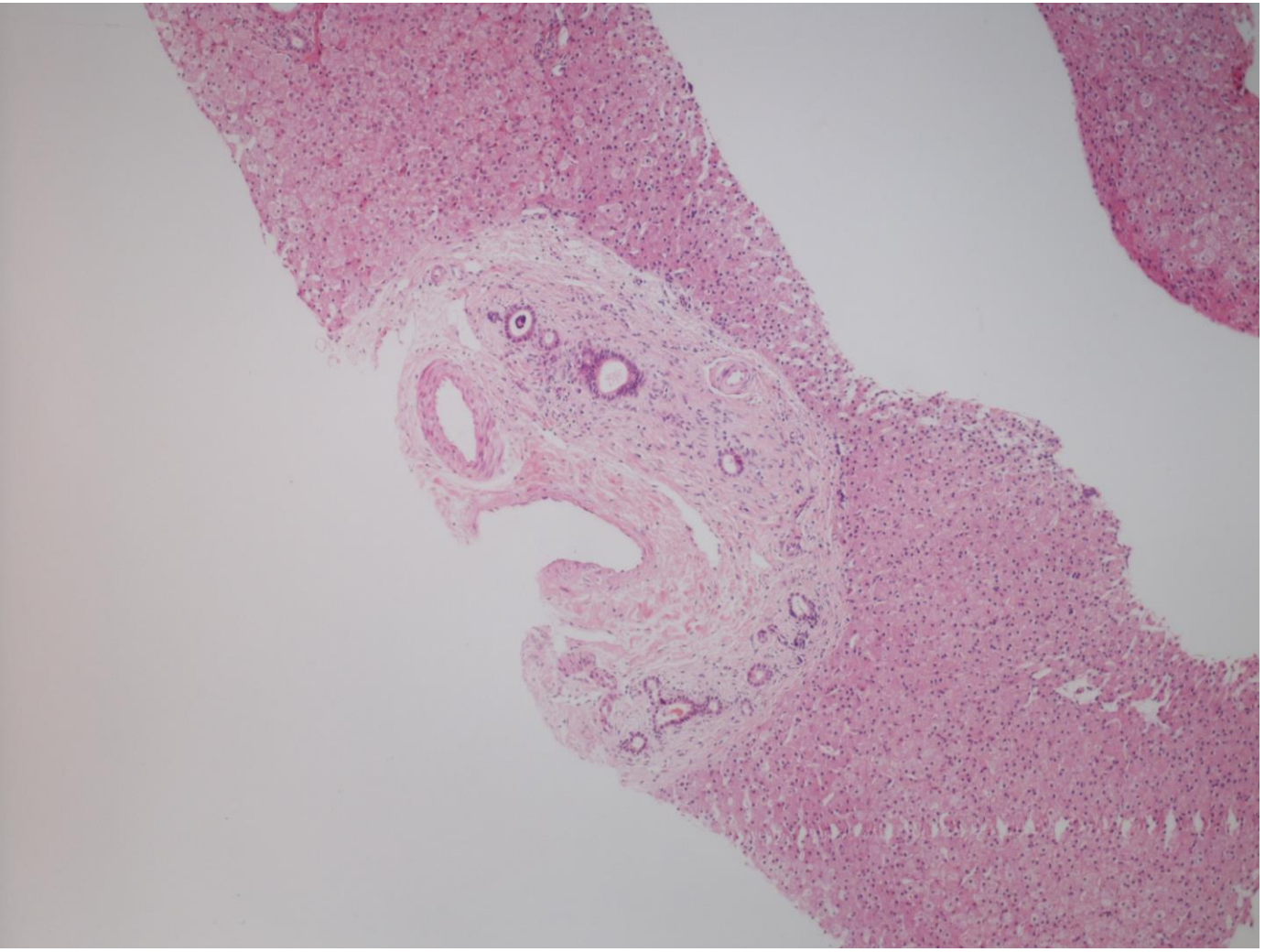


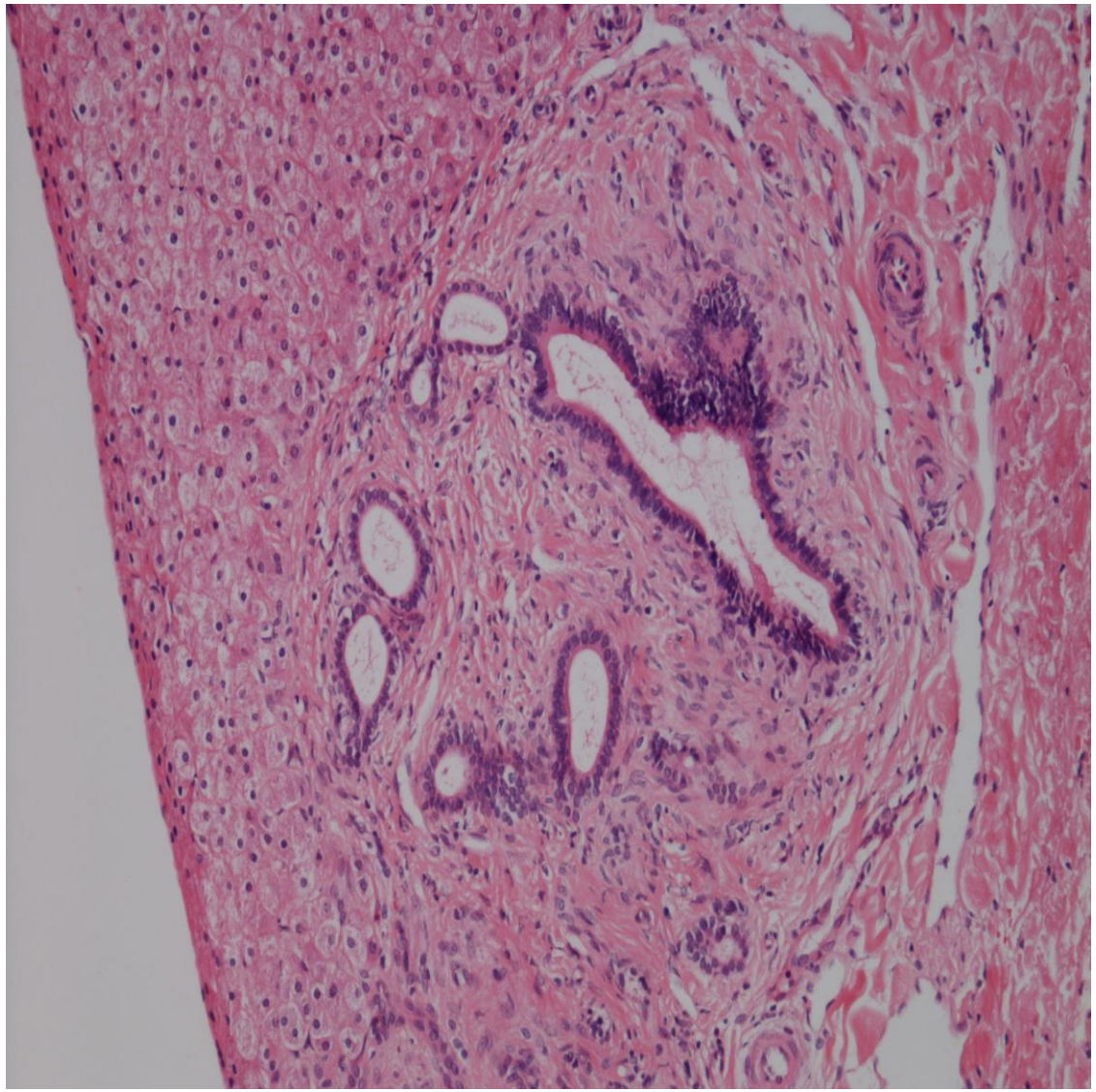


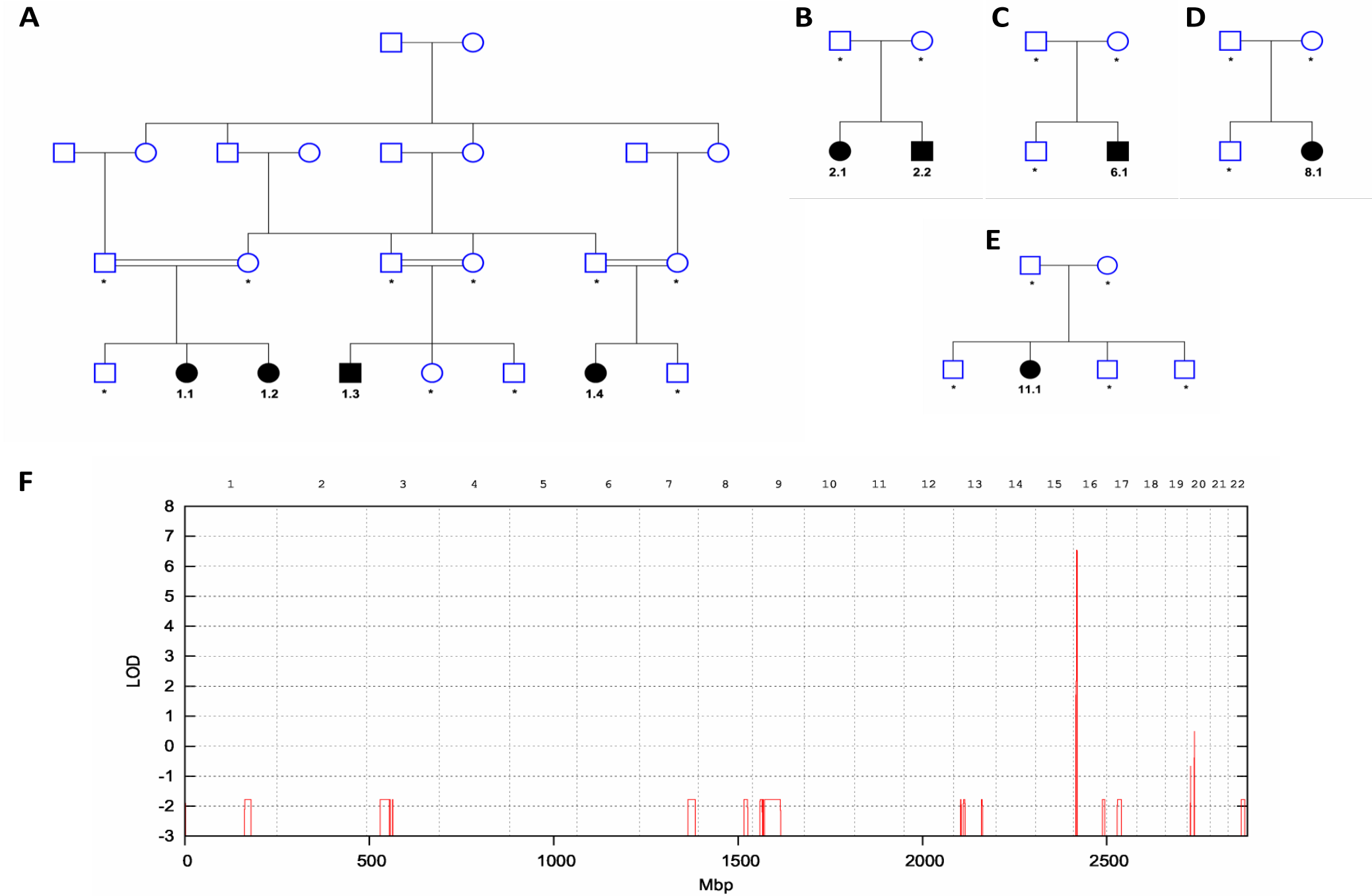
More patients with “HIPKD”?

- Parents identify through social media another patient with HI and PKD in the US
- A RaDaR ARPKD patient day is attended by a family with 2 siblings, both affected by HI and PKD
- A review of clinical features of patients followed at the GOSH HI service identifies another 9 patients with associated renal cysts
- A Spanish doctor presents a poster at an endocrine meeting describing a consanguineous family with 4 affected siblings

Liver biopsy in a 2-year old girl






A
1.1
1.2
1.3
1.4
B
2.1
2.2
C
6.1
D
8.1
E
11.1
F
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22
LOD
8 7 6 5 4 3 2 1 0 -1 -2 -3
0 500 1000 1500 2000 2500
Mbp

Where's the problem?

- No bi-allelic coding mutations in the linked region
- However: all patients share a non-coding mutation c.-167G>T in the promoter of *PMM2*
- Promoter mutation is either homozygous (consanguineous family) or *in trans* with *PMM2* coding mutation

Who's PMM2

- Phosphomannomutase 2
- Key enzyme on protein glycosylation ("post translational modification")
- Recessive coding mutations cause CDG1A, which occurs in 2 forms:
- Mild form with neurological involvement only (ataxia, cerebellar hypoplasia)
- Severe, multivisceral form with dysmorphic features (abnormal fat pads, inverted nipples) and severe neurological problems. 20% die in infancy
- Essentially any organ system can be involved, including: renal cysts and HI

CDG1A vs HIPKD

CDG1A

- Renal cysts and HI occasionally seen
- only in conjunction with severe neurology and dysmorphic features
- Abnormal transferrin mobility

HIPKD

- HI, Renal cysts +/- liver involvement only
- No apparent neurological problems
- Normal transferrin mobility

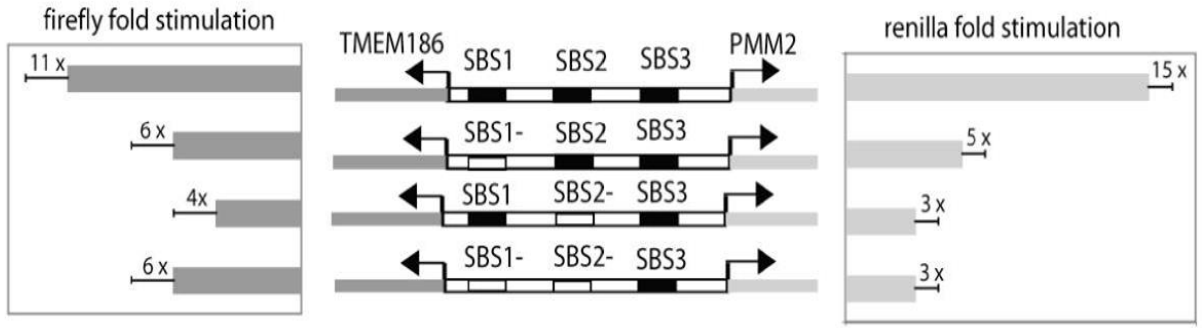
PMM2 pleiotropy

- **HIPKD = organ specific PMM2 dysfunction**
- **CDG1A = generalised PMM2 dysfunction**

?organ specific effect of the promoter mutation?

Trying to make sense

Bidirectional *PMM2* promoter



Genome-wide evidence for an essential role of the human Staf/ZNF143 transcription factor in bidirectional transcription

Yannick-Noël Anno[1,2], Evelyne Myslinski[2], Richard Patryk Ngondo-Mbongo[2], Alain Krol[2], Olivier Poch[1], Odile Lecompte[1] and Philippe Carbon[2,*]

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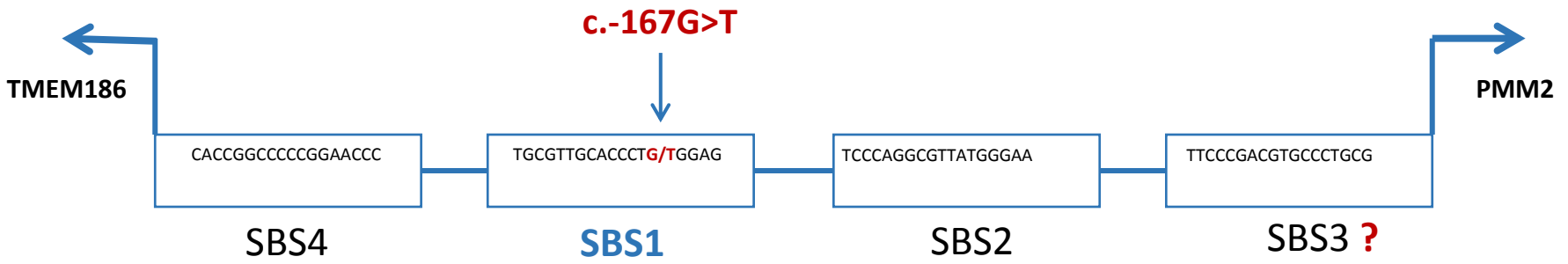
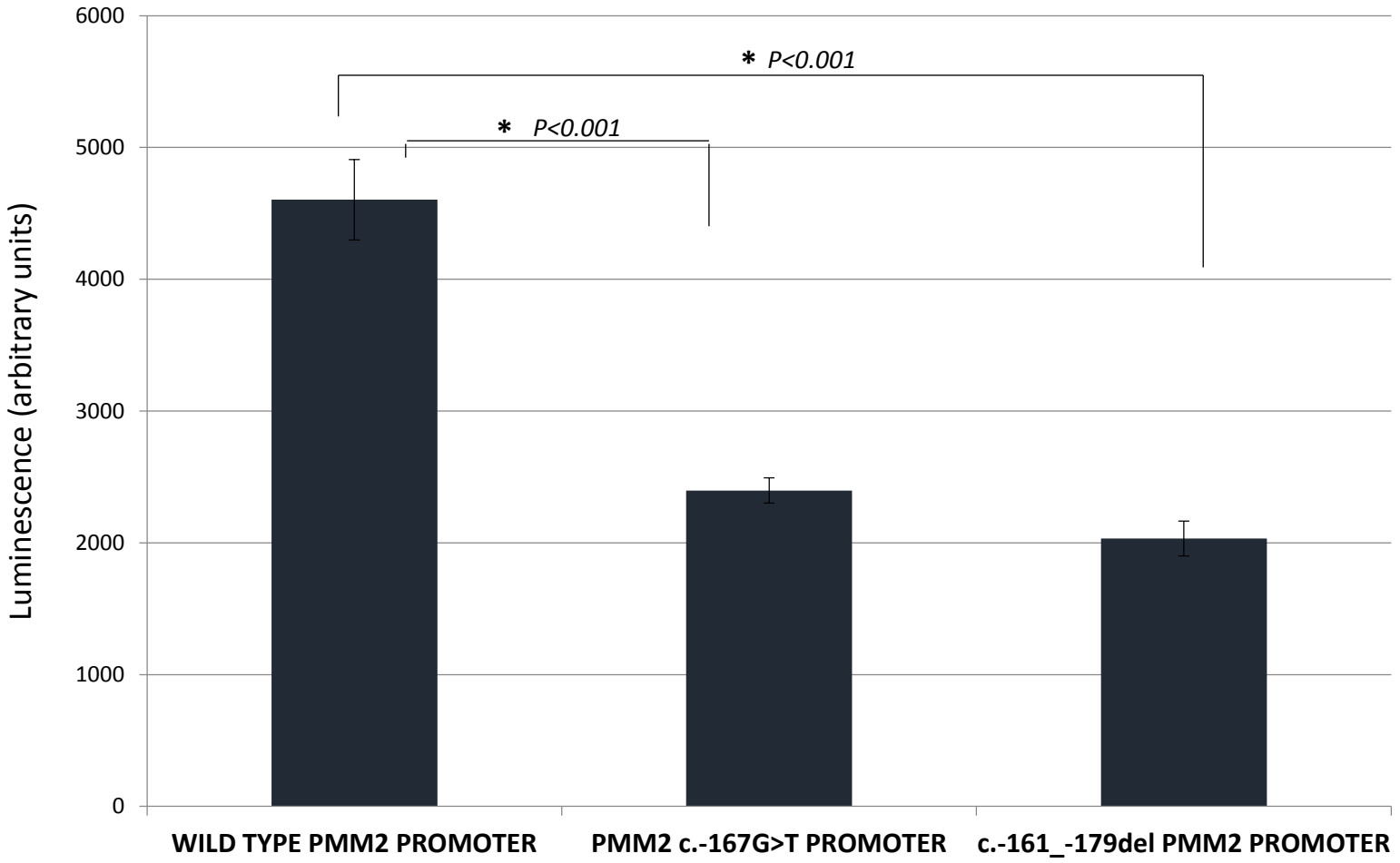
Mutation in consensus sequence of bidirectional *PMM2* promoter



modified from Anno et al. 2010, Nucleic Acids Res.

PMM2 PROMOTER
LUCIFERASE ASSAY

→ Human Epithelial Kidney Cells



n=3 (in triplicates)

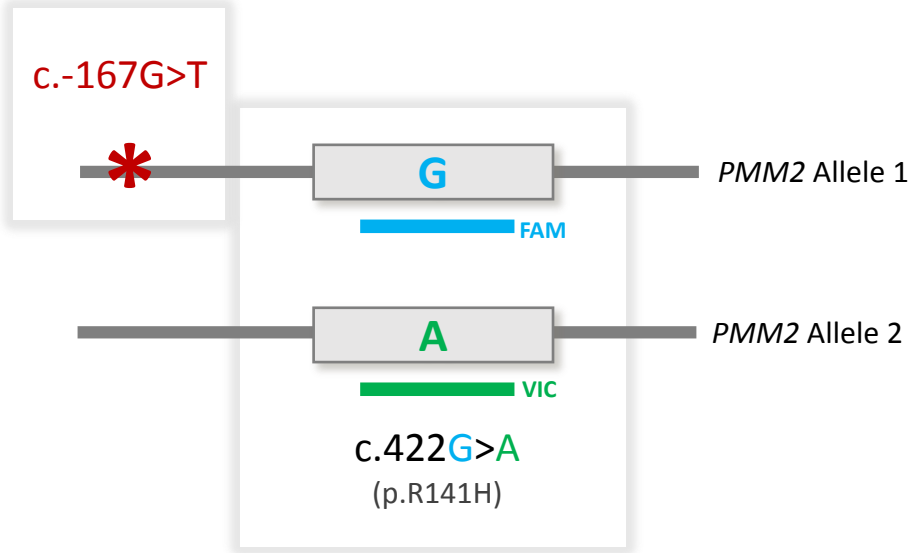
+/- SEM

30,000 cells/well

GENE EXPRESSION STUDIES - dPCR

PMM2 ALLELIC DISCRIMINATION

→ Compound heterozygous patient cells (**c.-167G>T**/c.422G>A)



Labelling: FAM & VIC TaqMan® probes

GENE EXPRESSION STUDIES - dPCR
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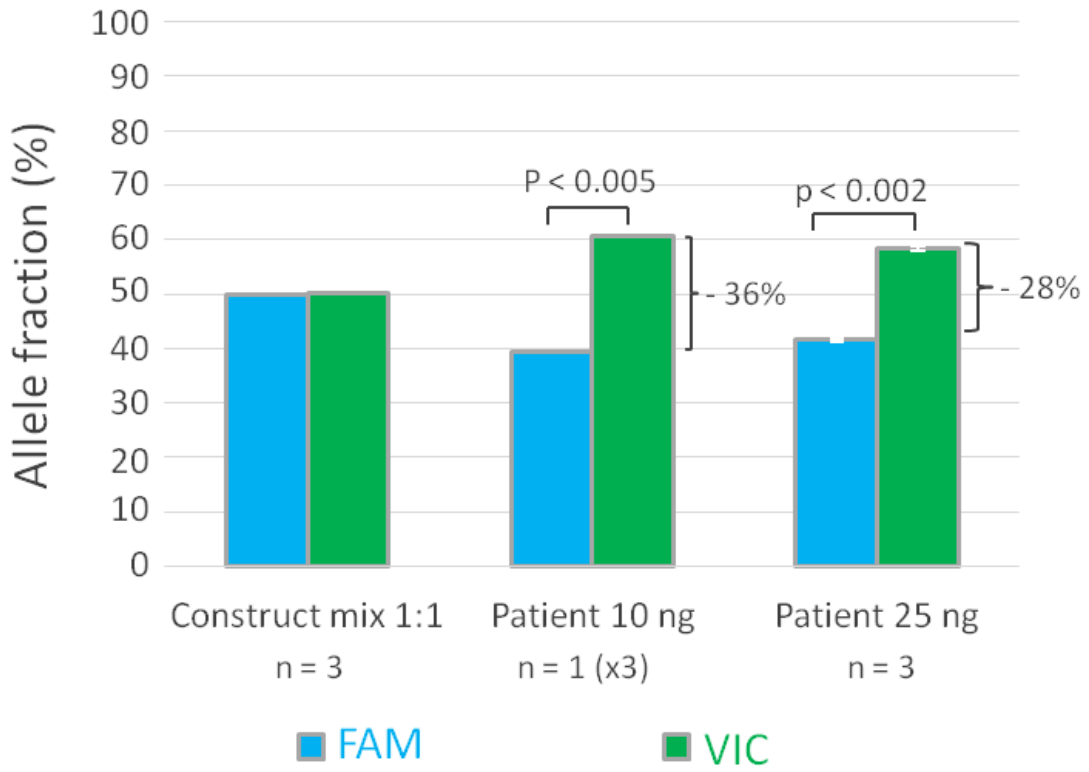
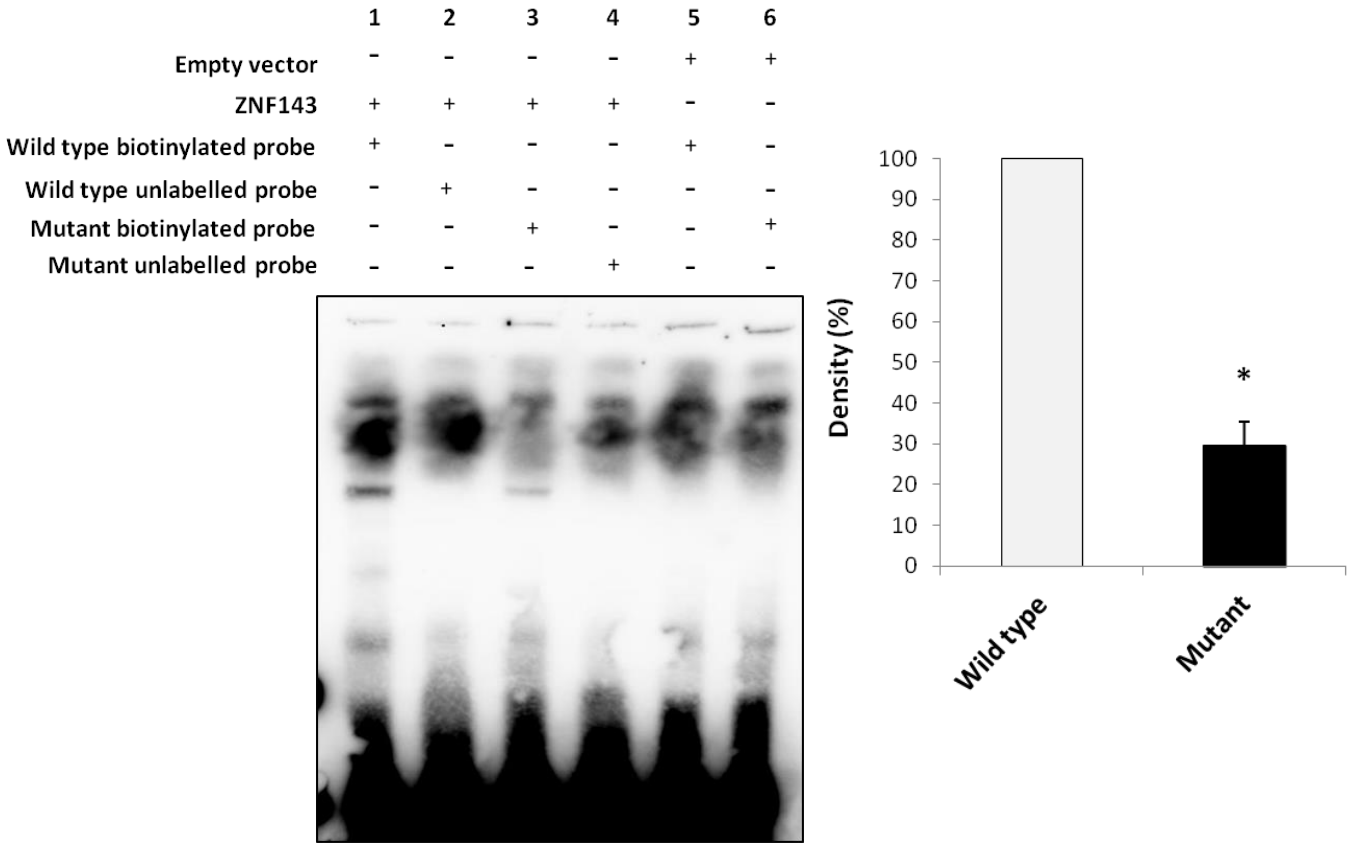


→ Expression level mutant allele 1/3 reduced in patient cells.

Protein-DNA interaction: EMSA



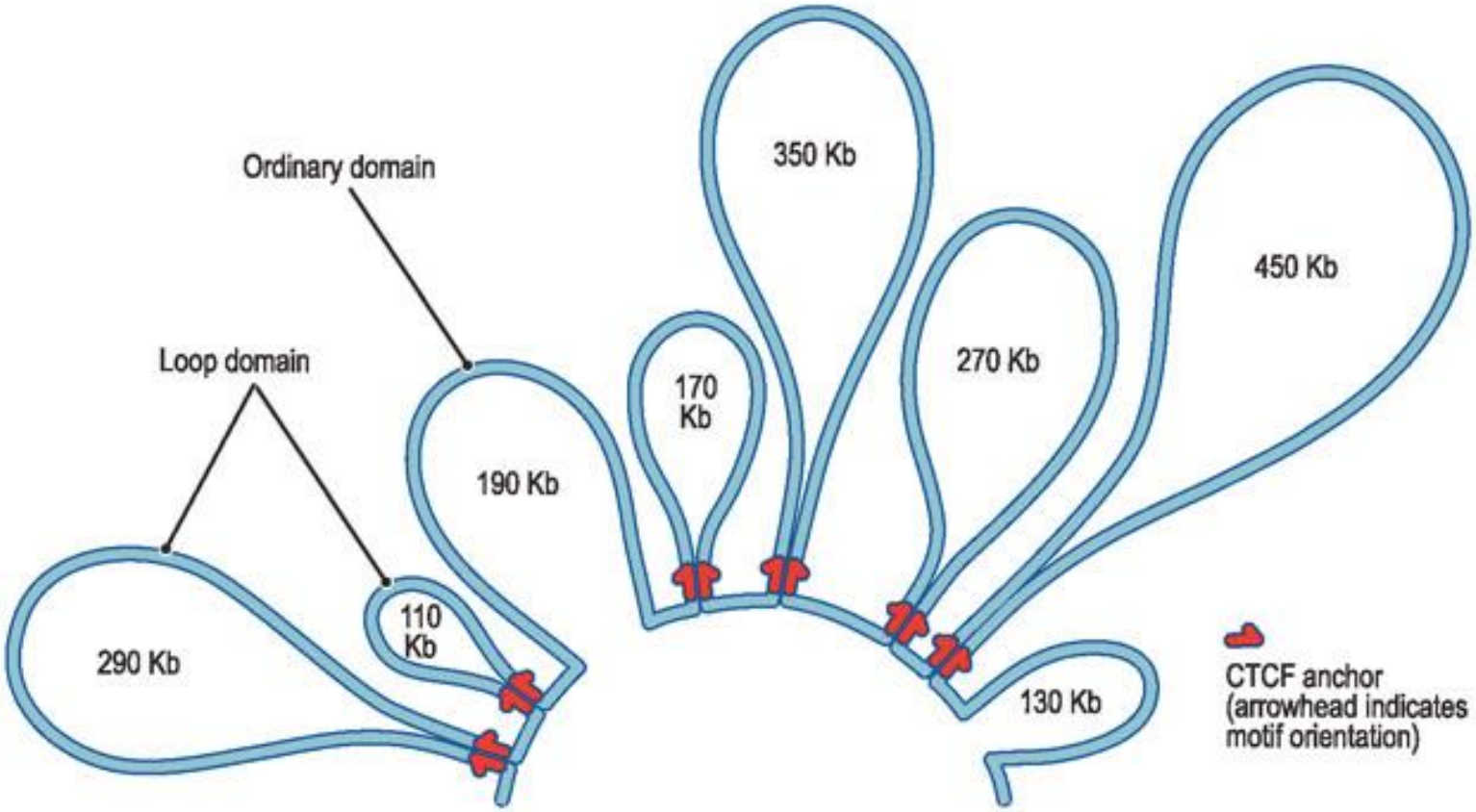
Yet: how does this cause organ-specificity?

- PMM2 is ubiquitously expressed
- why does the promoter mutation only affect pancreas, kidney and liver?

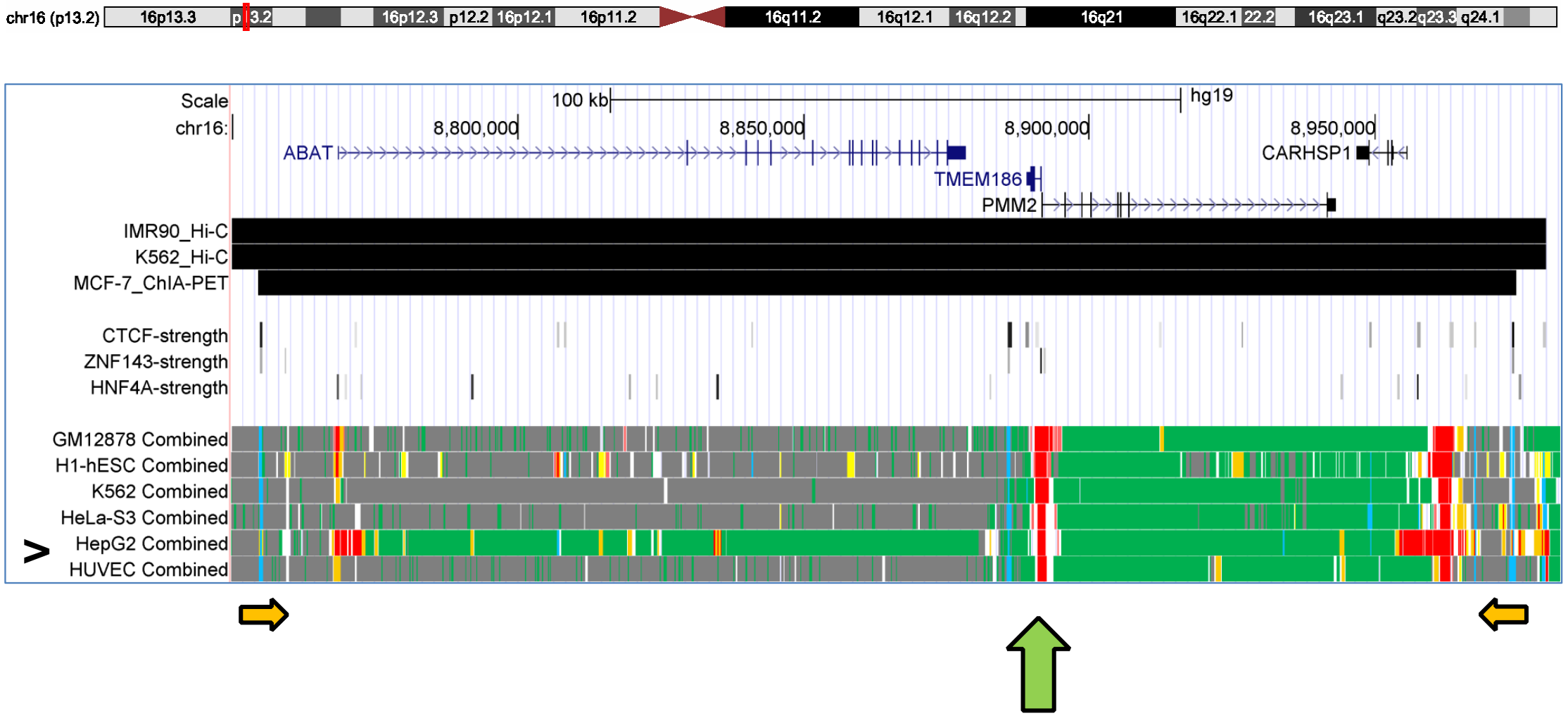
Between a chicken and a grape plant

- Chicken: 17.000 genes
- Humans: 22.000 genes
- Grape plant: 30.000 genes

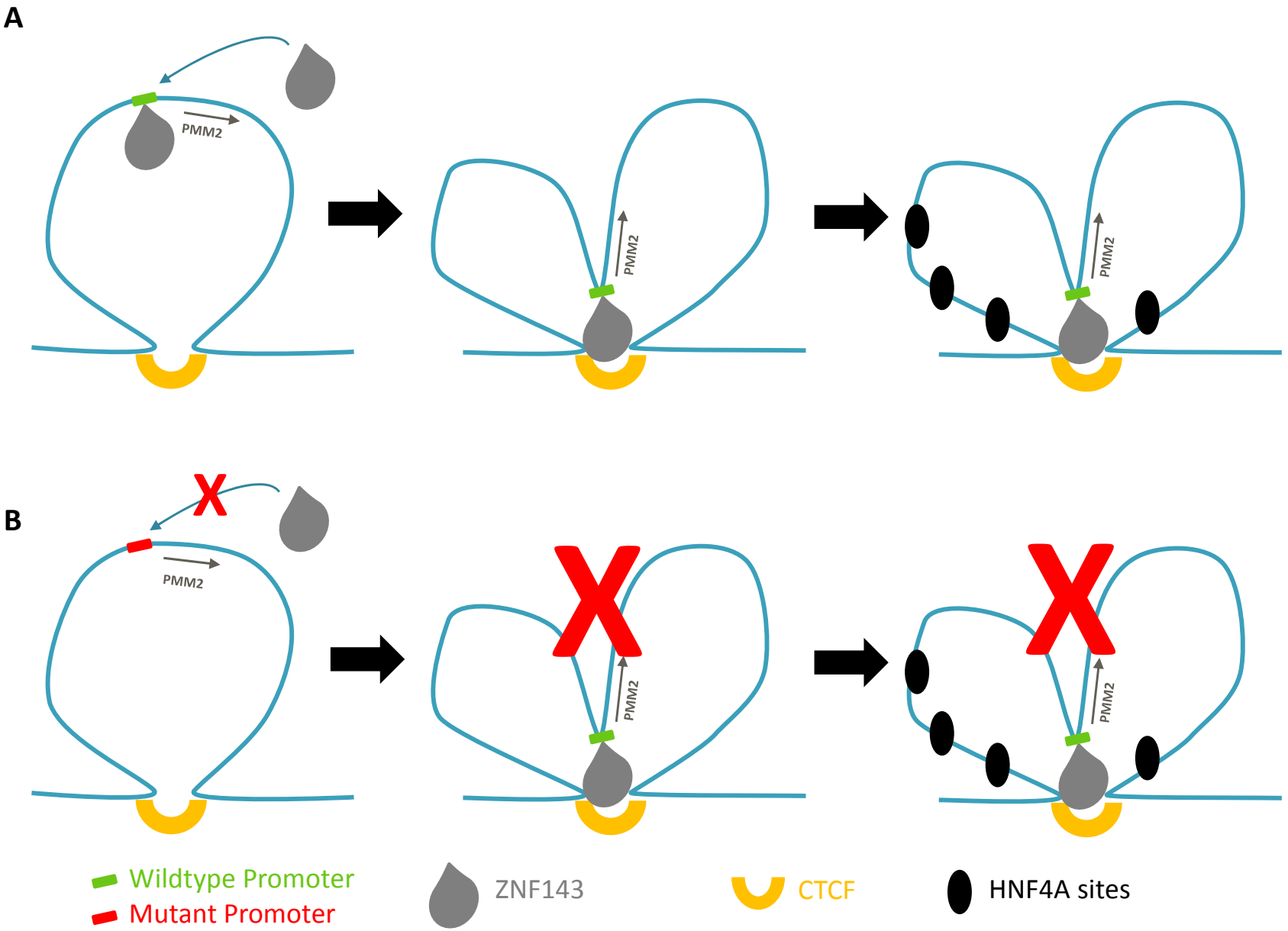
I’s not the size that matters…



What does ZNF143 do?



A hypothesis



Conclusions

- HIPKD: a newly recognised disorder consisting of HI and PKD +/- liver involvement
- Currently 18 patients identified
- Spectrum of severity (3 reached ESKD in childhood)
- Promoter mutation affects PMM2 transcription in an organ-specific manner
- Provides insights into gene regulation

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Transferrin isoelectric focusing