The apeiron of arterial hypertension

Apeiron is the Greek term for „boundless or unlimited“.

In the 1920‘s, twin studies suggested that blood pressure is influenced by genetic variance

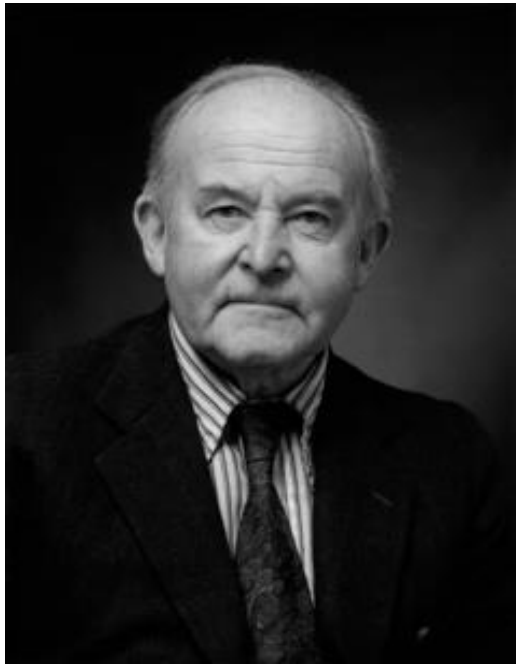
FCL working up patients 1970-present

In the 1970‘s, IUMC twin studies suggested that BP regulators were also influenced by genetic variance.

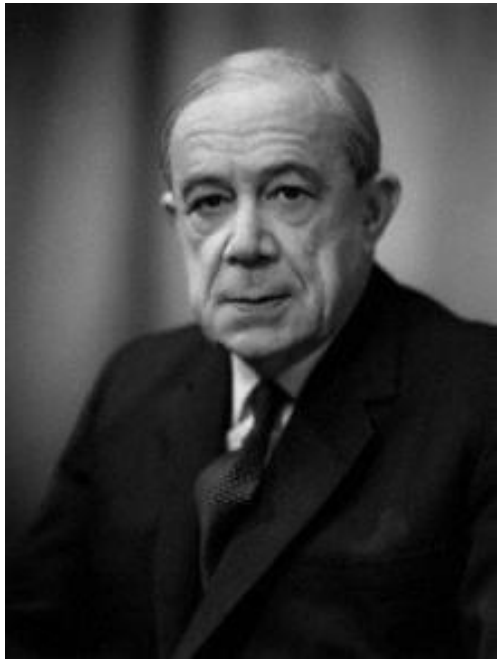
Association Between Push-up Exercise Capacity and Future Cardiovascular Events Among Active Adult Men JAMA 2019

In the late 1940's and 1950's, these two giants of cardiovascular medicine debated hypertension genetics

Platt presented evidence of its autosomal dominant inheritance and a bimodal distribution of blood pressures. In contrast, Pickering's viewpoint was that blood pressures varied continuously and unimodally, with hypertensives representing the upper end of the bell curve. Though Platt's view was favoured during his lifetime, Pickering's view ultimately dominated.



Sir George White Pickering, FRS (26 June 1904 – 3 September 1980) was a British medical doctor and academic.

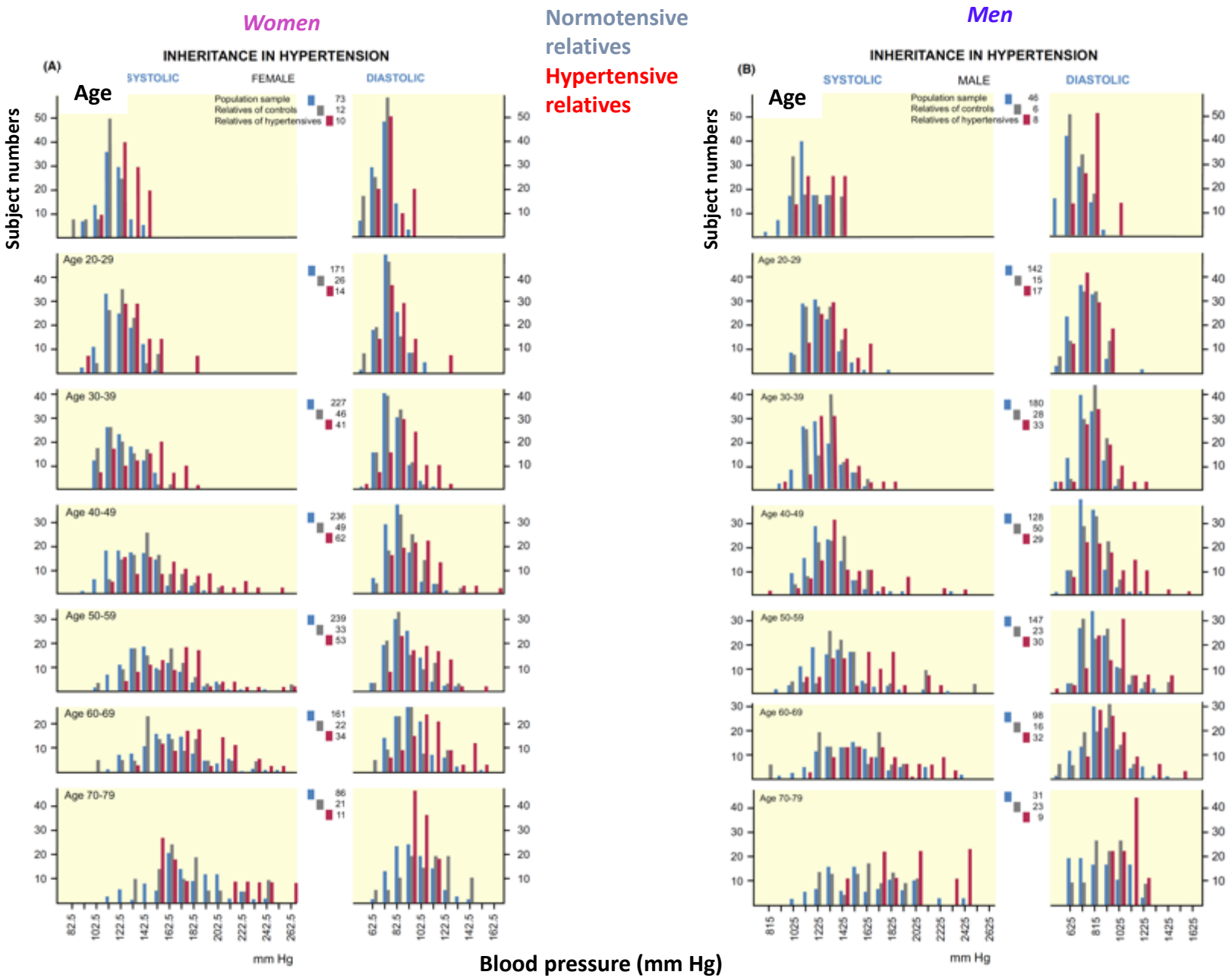


Sir Robert Platt, FRCP (16 April 1900 – 30 June 1978), 1st Baronet between 1959 and 1967, was a British physician.

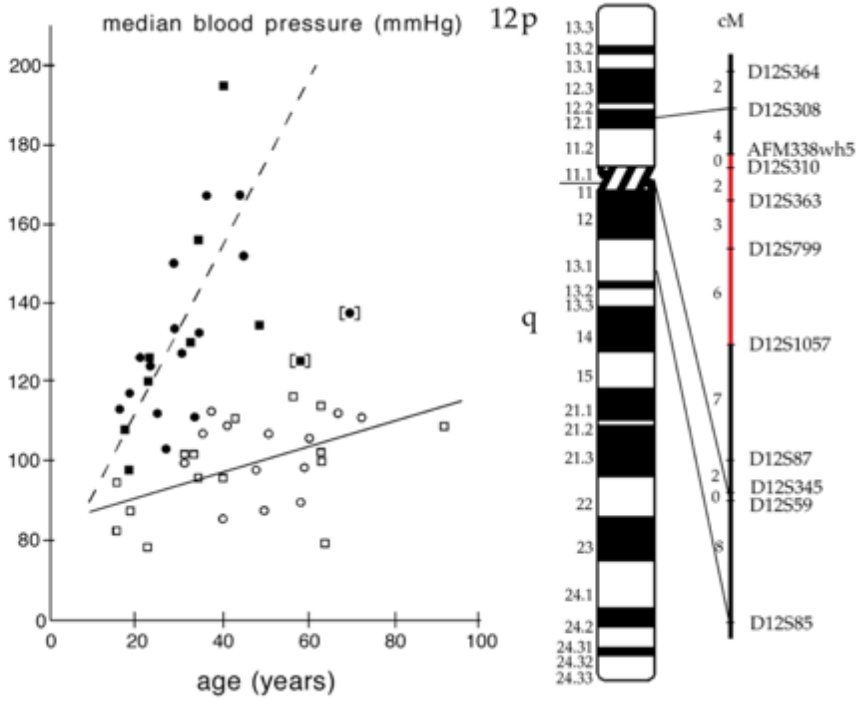
Sir Robert was then relegated to the House of Lords

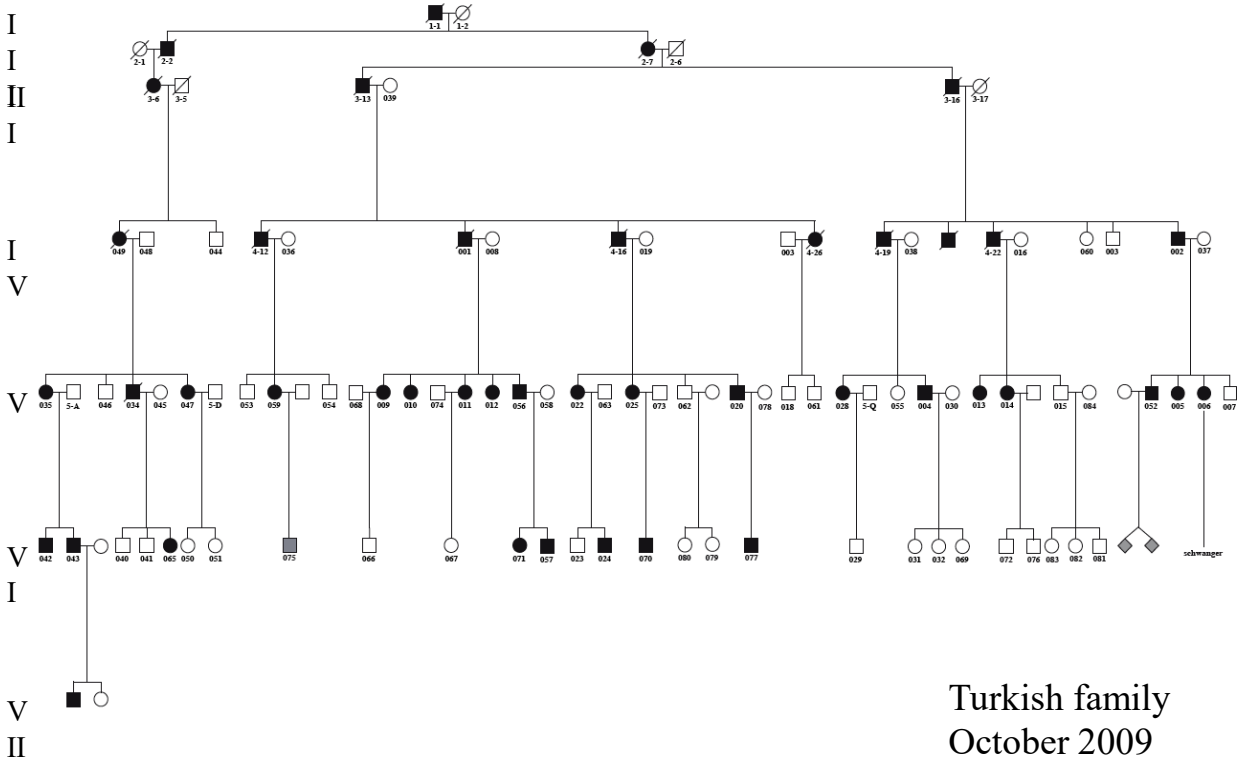
Pickering et al. published 4 papers in 1950's

All subjects
Normotensive relatives
Hypertensive relatives



Pickering flattened Platt, but really?





Turkish family
October 2009

Schuster et al. Nat Genet 1996

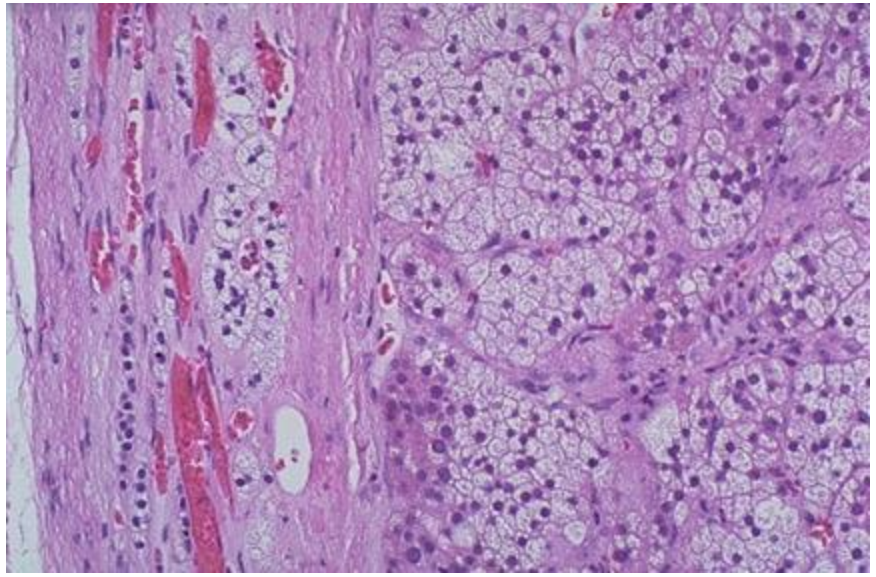
Thirty-seven year-old woman with rather severe hypertension

- Family history was negative.
- Receives HCTZ, amlodipine, valsartan.
- Physical examination was unremarkable; however, despite 3 drugs BP still poorly controlled.
- Urine status +1 Prot.
- Na 142, K 3.6, Cl 102, HCO_3 27 (mmol/l), $PaCO_2$ 41 mm Hg, pH 7.42.
- Creatinine 88 µmol/l.
- What to do?

Work-up for secondary causes

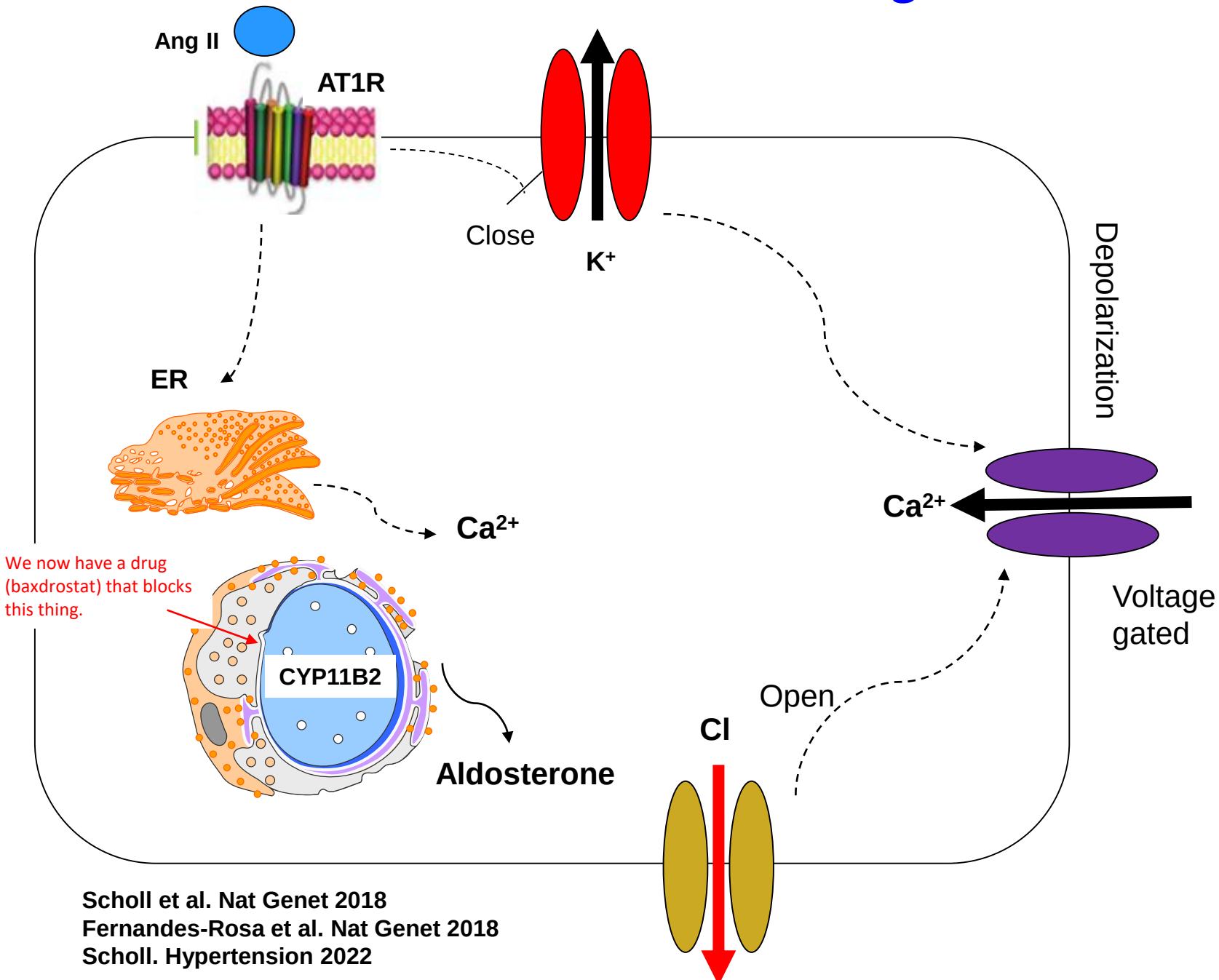
Aldo 25 ng/dl, PRA 0.6 ng/ml/h, ratio >30;
after salty diet, Urine Aldo 19 µg/24 h

Yellow fatty tumor



Capsule is left; Tumor resembles Zona fasciculata.
Tumor size not correlated with Aldo.
Some are very small.

Aldosteronism can be due to somatic or germ-line mutations

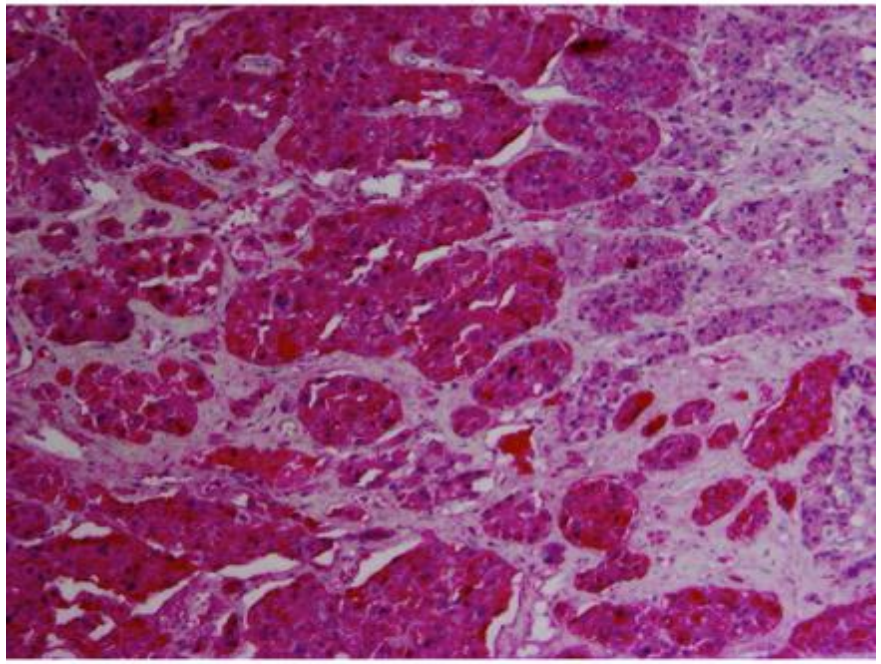


Scholl et al. Nat Genet 2018
Fernandes-Rosa et al. Nat Genet 2018
Scholl. Hypertension 2022

A 51 year-old sculptor from Czech Republic, BP 240/120 mm Hg. He did not look "Cushingoid".

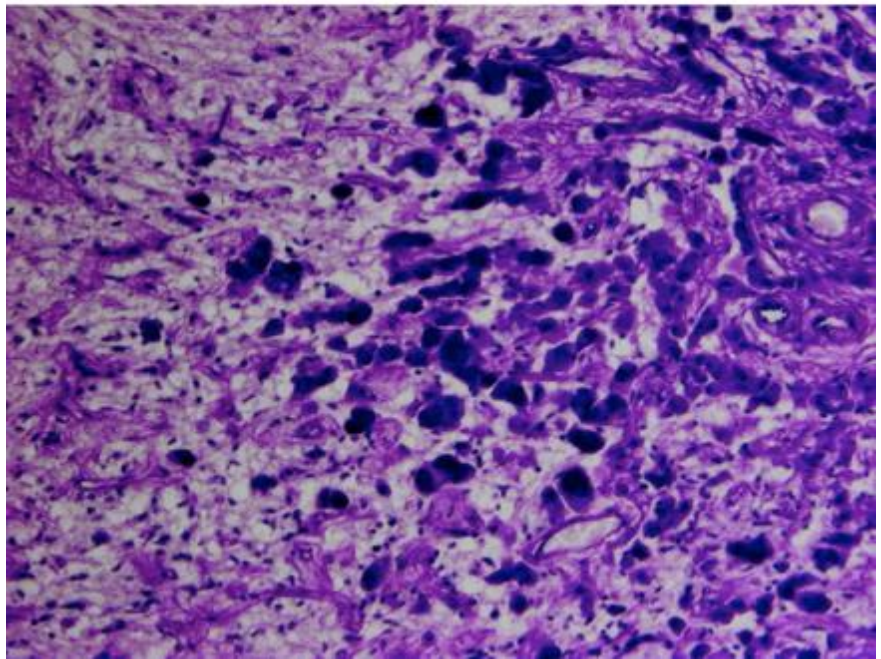
- Na 143, K 2.7, Cl 99, HCO_3 31 (mmol/L).
- pH 7.53, $PaCO_2$ 38 mm Hg, Hb 16.7 g/dl, glucose 8 mmol/L. Aldo/PRA ratio 8.
- Cortisol 1750 nmol/L, ACTH 22 pmol/L.
- CT und MRI negative.
- Torasemide, amoldipine, spiro, candesartan, clonidine and BP 140/90.
- Petrosal-Sinus sampling was scheduled.
- On the night before, he jumped out of a 3rd story window and was killed.
- We missed the psychiatric symptoms of Cushing‘s disease.

A



A. ACTH Immunohistochemistry for basophilic cells in the anterior pituitary.

B



B. Invasion of the basophilic cells into the posterior pituitary

Singer et al. Hypertension 2008

Gain-of-function somatic mutations in the ubiquitin specific protease 8 (*USP8*) gene have recently been reported as a cause of pituitary adenomas in Cushing disease.

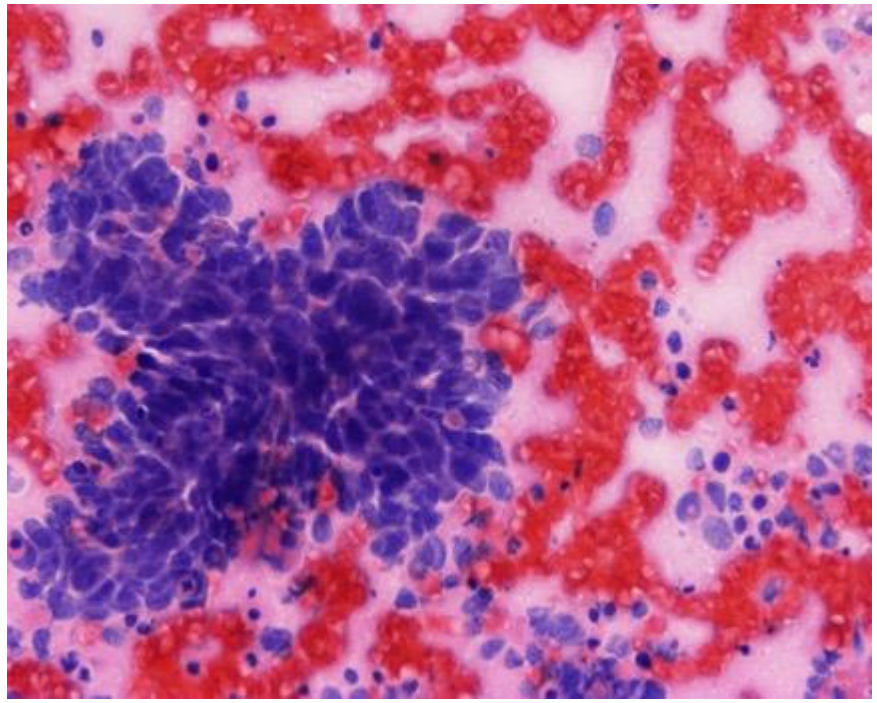
Ballmann – Scholl. USP8 mutations. J Endocr Soc 2018

68 year-old woman with acute coronary syndrome; she received abciximab and an LAD stent.

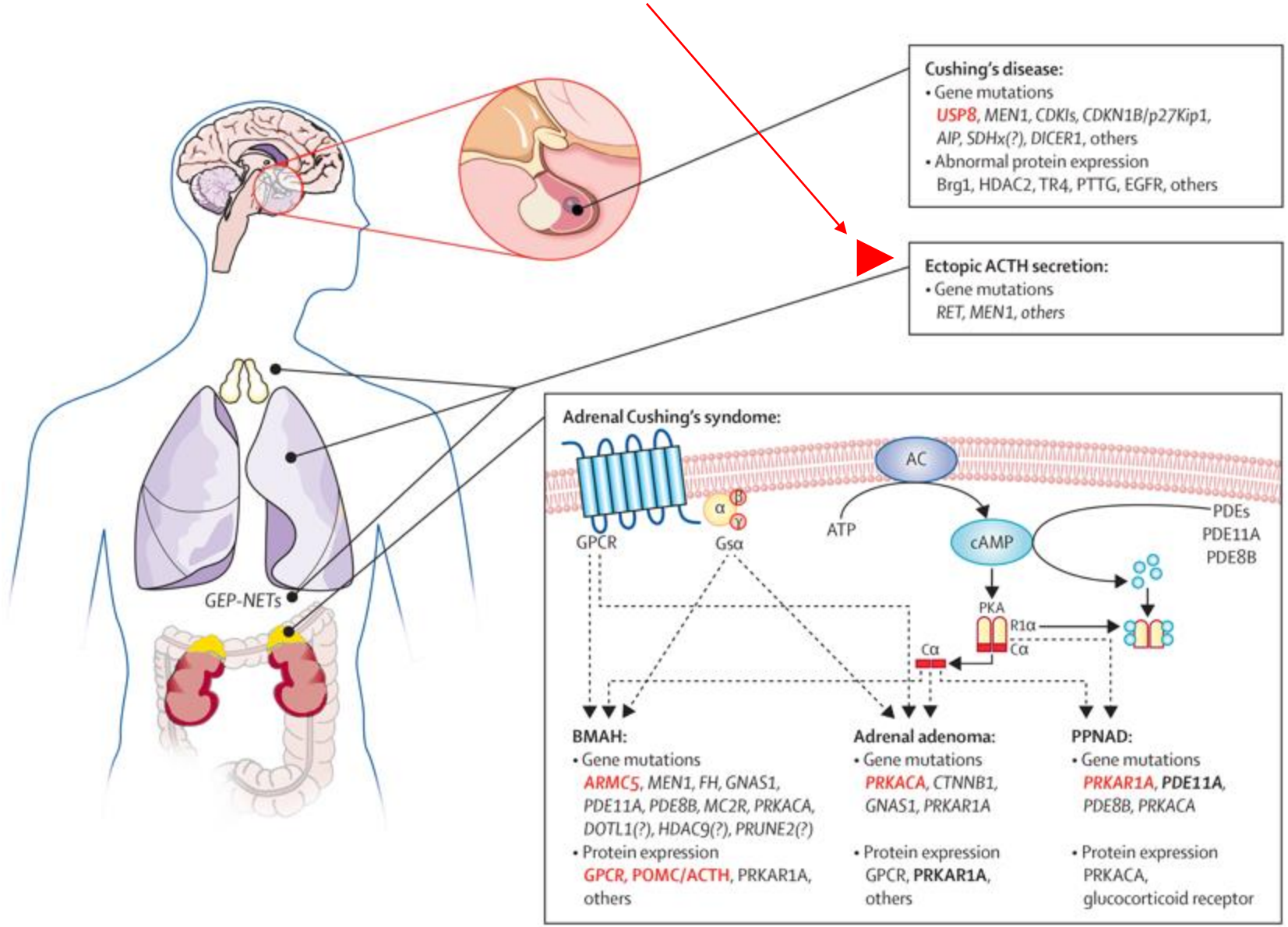
- pH 7.52, $PaCO_2$ 43 mm Hg, PaO_2 82 mm Hg, Na 141, K 2.7, Cl 96, HCO_3 36 mmol/l.
- Acid-base disturbance?
- UCl, UK, UNa, UOsm, TTKG?
- TTKG = (U/P K) ÷ (U/P Osm).
- UK was 36 mmol/l UOsm was 270.
- TTKG was 14-fold; thus, the kidneys are responsible for the potassium loss
- Urine K is far too high for serum K. That alone is to declare kidneys guilty.

Chain smoker, but again – no Cushingoid habitus

- PRA was low.
- PA was normal.
- Cortisol 2000 nmol/l.
- ACTH 90 pmol/l.
- No fall with Dex.
- Bronchoscopy and Bx.



Ectopic ACTH Syndrome



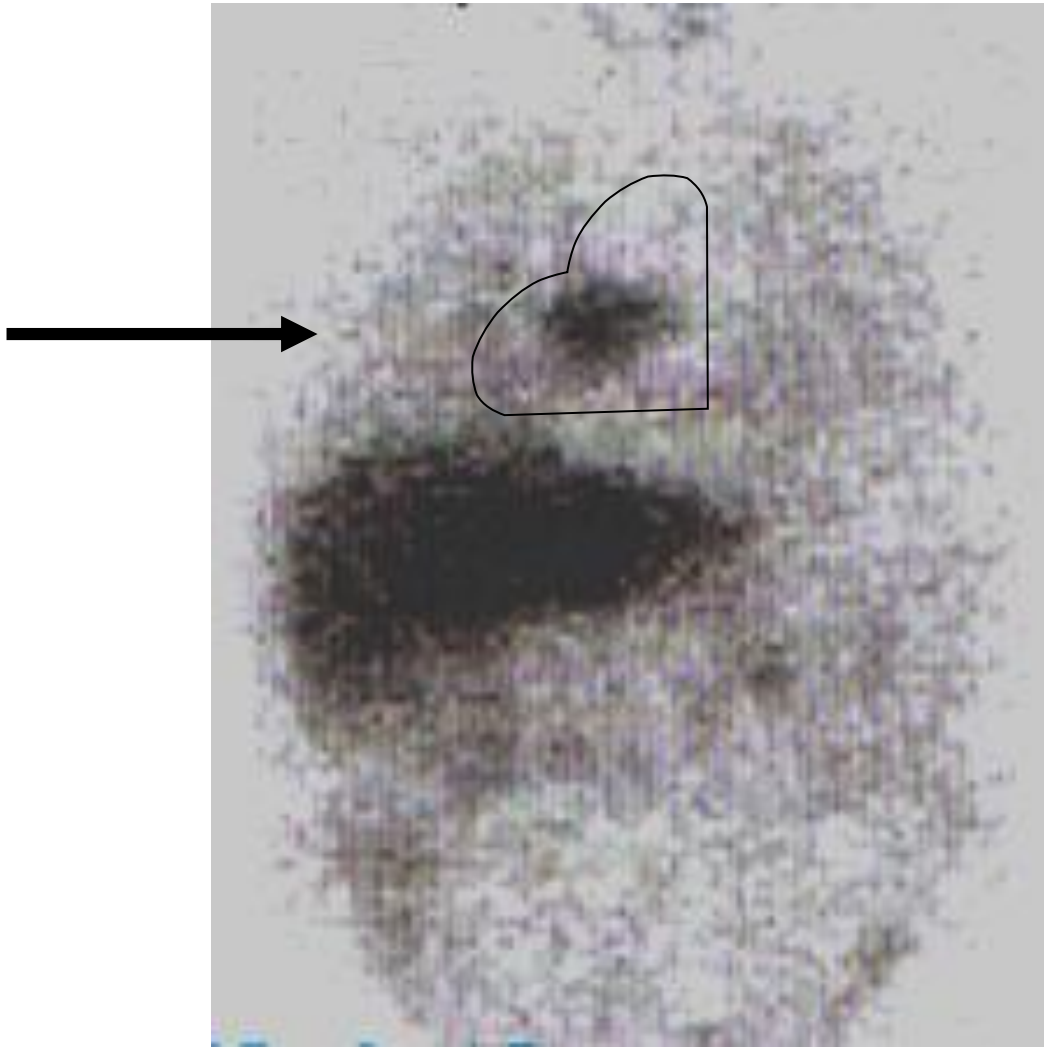
Lacroix et al. Lancet 2015

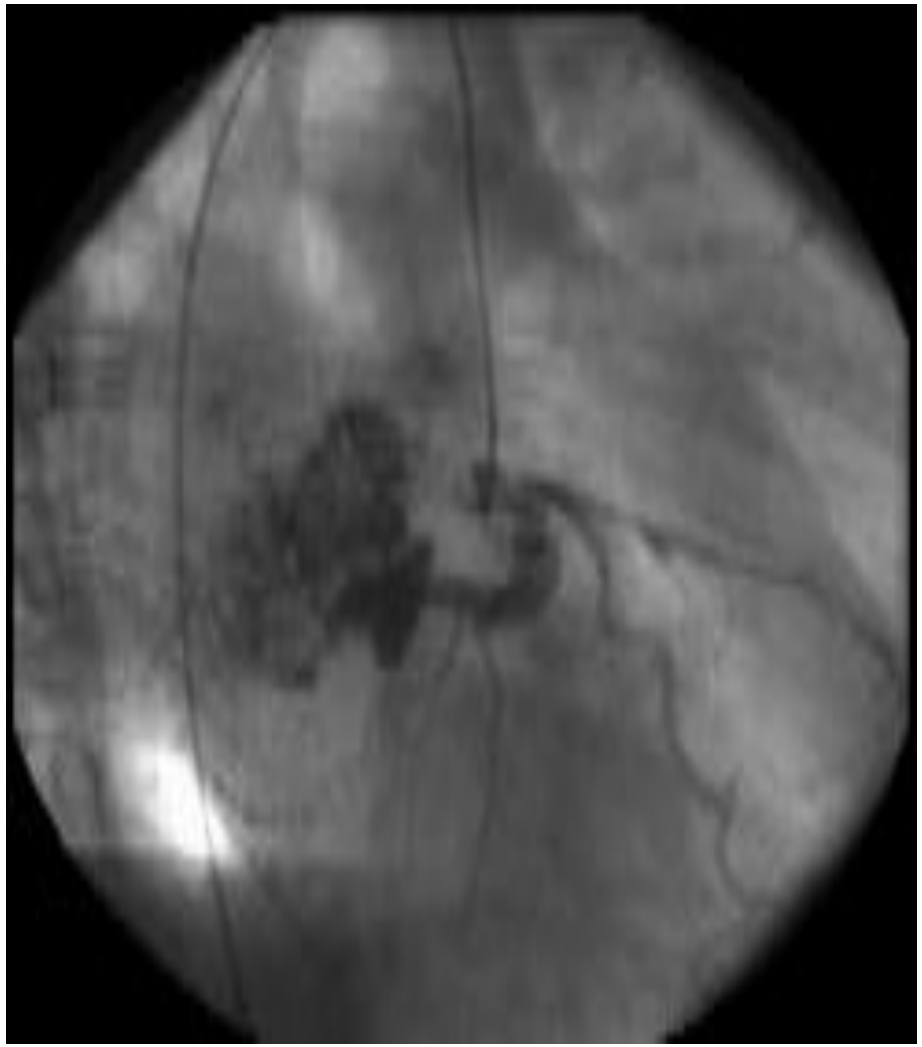
52 year-old patient with multiple previous workups for pheochromocytoma

- Complaints have been there for 5 years.
- BP 140/90 but values up to 212/114 mm Hg are recorded.
- Earlier pheochromocytoma workups were negative.
- Abdominal MRI was negative.
- We put him on a tilt table and he passed out.

Birkenfeld et al. Am J Med 2004

^{123}I-Meta-iodobenzylguanidine (MIBG)
"Unclear" medicine finally came through





Hartmut Neumann and N Engl J Med 2021

Clinical Scenario	Initial Studies	Follow-up Studies
Patient has disease-causing mutation		
RET mutation	Obtain levels of metanephrines and serum calcitonin and calcium; perform MRI of the abdomen; if the thyroid gland has not been resected, seek consultation for endocrine surgery	Obtain levels of metanephrines and serum calcitonin and calcium annually
VHL mutation	Obtain levels of metanephrines; perform MRI of the head, spinal cord, and abdomen; perform ophthalmoscopy	Obtain levels of metanephrines annually; perform MRI of the head, spinal cord, and abdomen; perform ophthalmoscopy; if no tumor is found, monitor every 2 or 3 yr
SDHA, *SDHB*, or *SDHD* mutation	Obtain levels of metanephrines; perform MRI of the skull base, neck, thorax, retroperitoneum, and pelvis or perform PET-CT with ^{68}Ga-labeled DOTATATE	Obtain levels of metanephrines annually; if a pheochromocytoma or paraganglioma has been resected, perform MRI of the surgical region annually for 3 yr; perform MRI of the neck, chest, abdomen, and pelvis every 3 yr
SDHC or *SDHAF2* mutation	Obtain levels of metanephrines; perform MRI of the skull base and neck or perform PET-CT with ^{68}Ga-labeled DOTATATE	If a paraganglioma or pheochromocytoma has been resected, perform MRI of the surgical region annually for 3 yr; perform MRI of nonsurgical regions every 3 to 5 yr
MAX or *TMEM127* mutation	Obtain levels of metanephrines; perform MRI of the abdomen or perform PET-CT with ^{68}Ga-labeled DOTATATE	Obtain levels of metanephrines annually; if a pheochromocytoma or paraganglioma has been resected, perform MRI of the surgical region annually for 3 yr; perform MRI of the abdomen every 3 yr
Neurofibromatosis type 1	Obtain levels of metanephrines	If hypertension or clinical symptoms develop, obtain levels of metanephrines

TMEM127 is a tumor suppressor gene, inactivating germline mutations in which causes hereditary pheochromocytoma and paraganglioma.
Diseases associated with MAX include Hereditary Paraganglioma-Pheochromocytoma Syndromes and Pheochromocytoma.

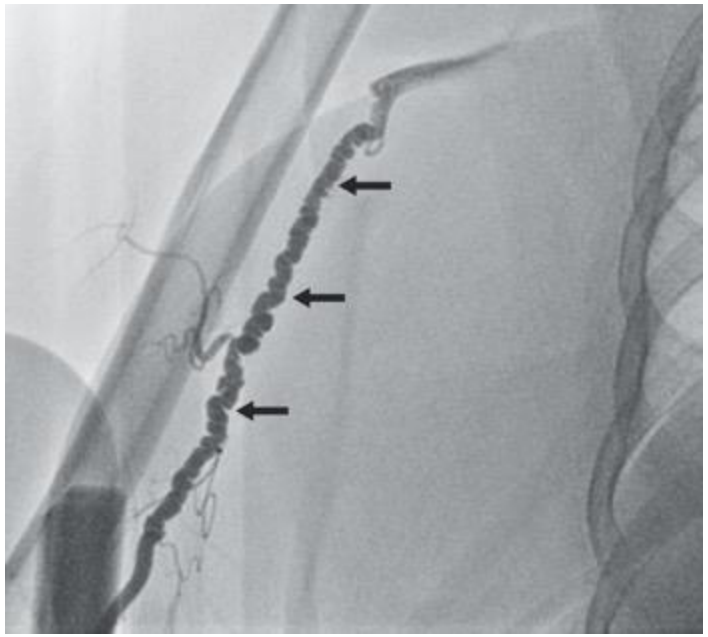
Pacak-Zhuang Syndrome and HIF2alph: EPAS1 somatic gain-of-function mutation
Belzutifan, a Potent HIF2α Inhibitor, in the Pacak-Zhuang Syndrome- N Engl J Med 2021

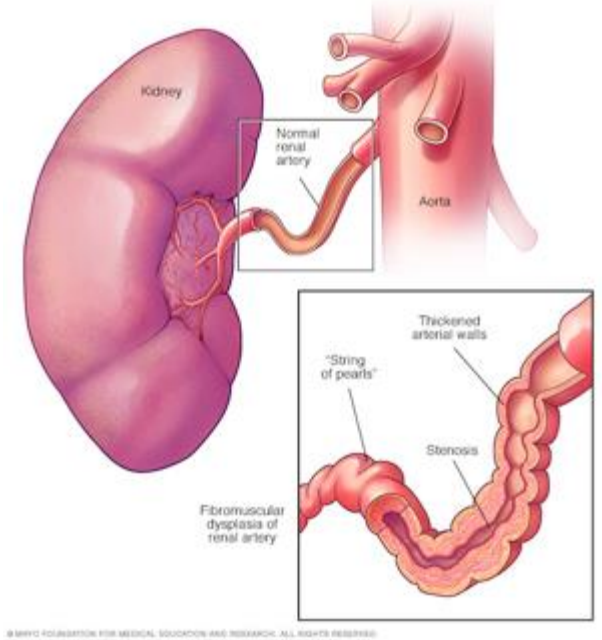
63 year-old woman with acute coronary syndrome. Hypertensive for 5 years. Sister had RVH from FMD.

- Troponin slightly increased.
- Exam otherwise OK.
- Na 139, Cl 100, K 4.5, HCO_3 24 (mmol/l).
- ECG consistent with acute coronary syndrome.
- Catheter could not be advanced along brachial artery.

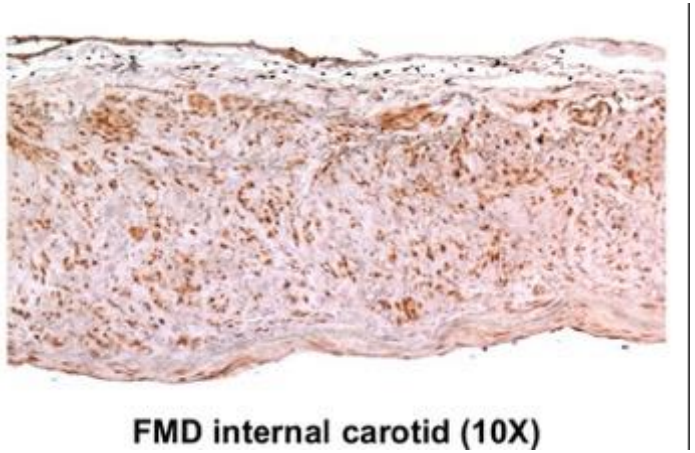
Miller et al. N Engl J Med 2017

Fibromuscular dysplasia of the brachial artery





Phosphatase and actin regulator-1 gene (*PHACTR1*).



FMD internal carotid (10X)

Kiando et al. PLoS Genet 2016

INSERM, UMR970 Paris

This protein has been associated with coronary artery disease and migraines through genome-wide association studies. The PHACTR1 gene also contains one of 27 SNPs associated with increased risk of coronary artery disease.

38 year-old woman referred because BP 250/110 mm Hg

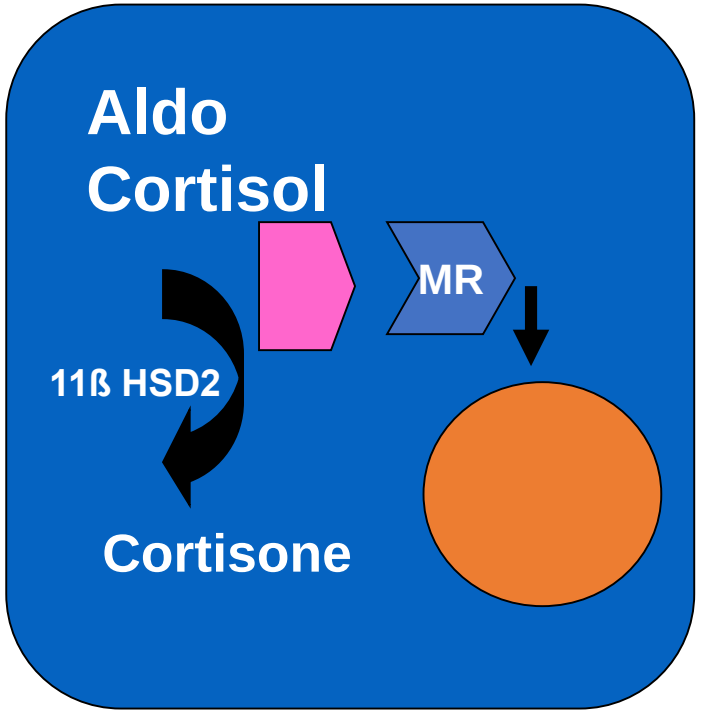
- Family history was negative.
- Physical examination normal.
- Na 143, Cl 105, K 3.5, HCO_3 26 (mmol/l).
- PRA <1 ng/ml/h; PA low.
- Three-drug therapy was unsuccessful.
- We were clueless.

Found in her belongings

Woywodt et al. Postgrad Med J 2000

11-Beta-hydroxysteroid dehydrogenase-2

Mutation or licorice with glycyrrhetinic acid



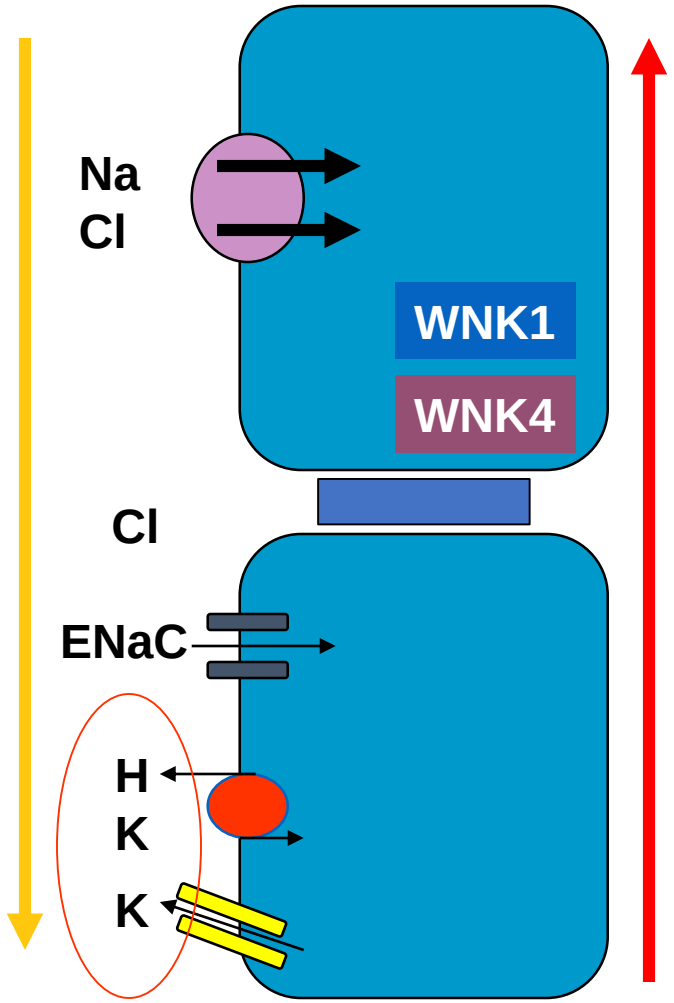
Licorice, „smokeless“ tobacco,
Huangin-Tang,
other herbal products
etc.

„Apparent mineralocorticoid excess“

24 year-old woman (patient of a friend from Paris) spends time in Berlin. She has a Mendelian form of hypertension

- pH 7,41, $PaCO_2$ 35, PaO_2 100 (mm Hg), Na 140, K 6.3, Cl 109 HCO_3 21 (mmol/l).
- Creatinine 80 µmol/l, UpH 5.3, UNa 53, UK 34, UOsm 450.
- TTKG 3.6 (inappropriately low).
- PRA <1 ng/ml/h; PA normal upper limit.
- Mild metabolic acidosis, no AG with Hyperkalemia + high BP.
- What is going on here?

With-no-lysine kinases in pseudohypoaldosteronism type II

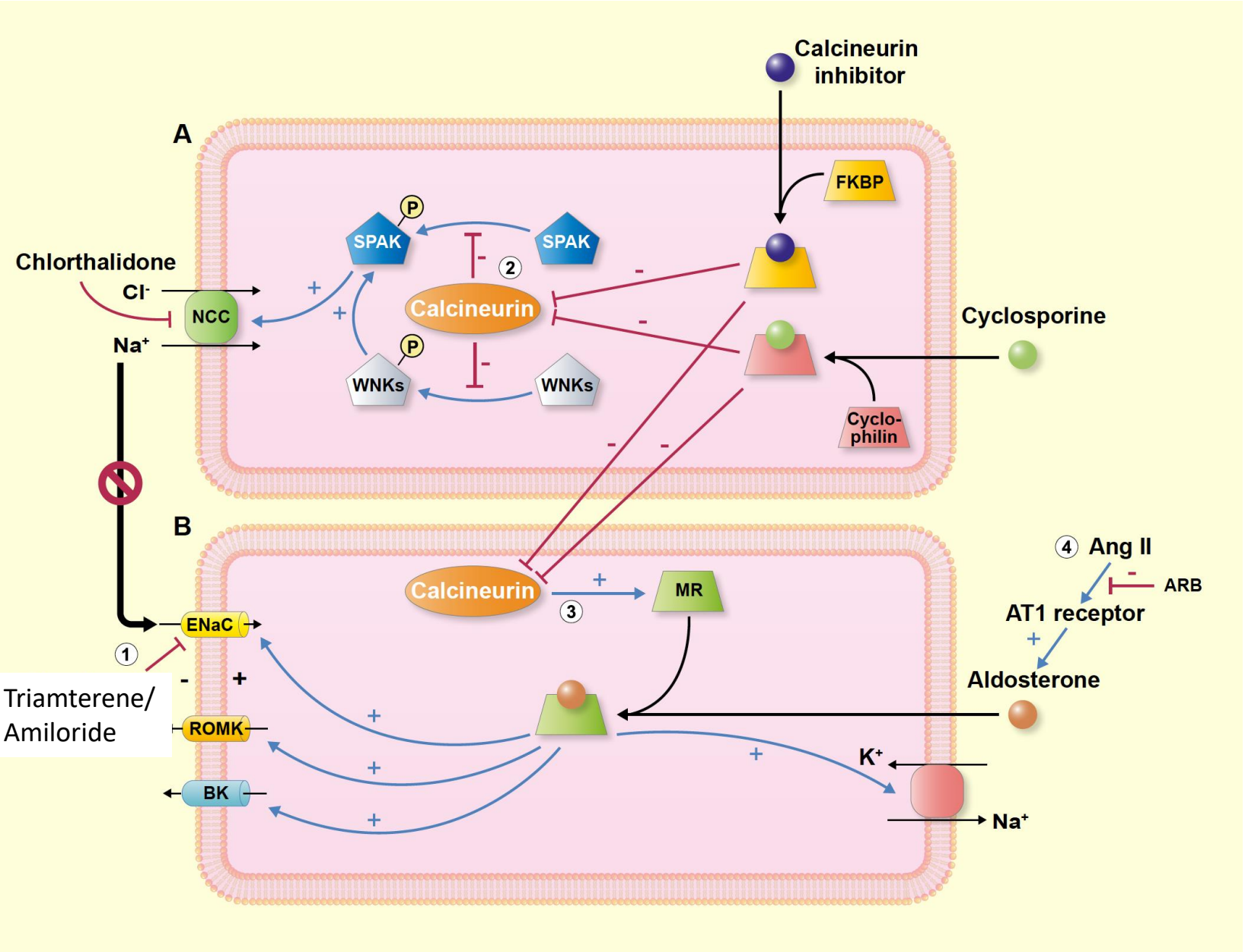


- The WNKs regulate sodium. chloride cotransporter (NCC) in DT.
- NCC is the HCTZ binding site.
- NCC is tightly regulated.
- Salt-sensitive hypertension.
- Less sodium is delivered to ENaC.
- Electrochemical gradient to eliminate potassium and protons is reduced.
- HCTZ cures this process.
- The condition is also called „Gordon“ Syndrome.

Wilson et al. Science 2001

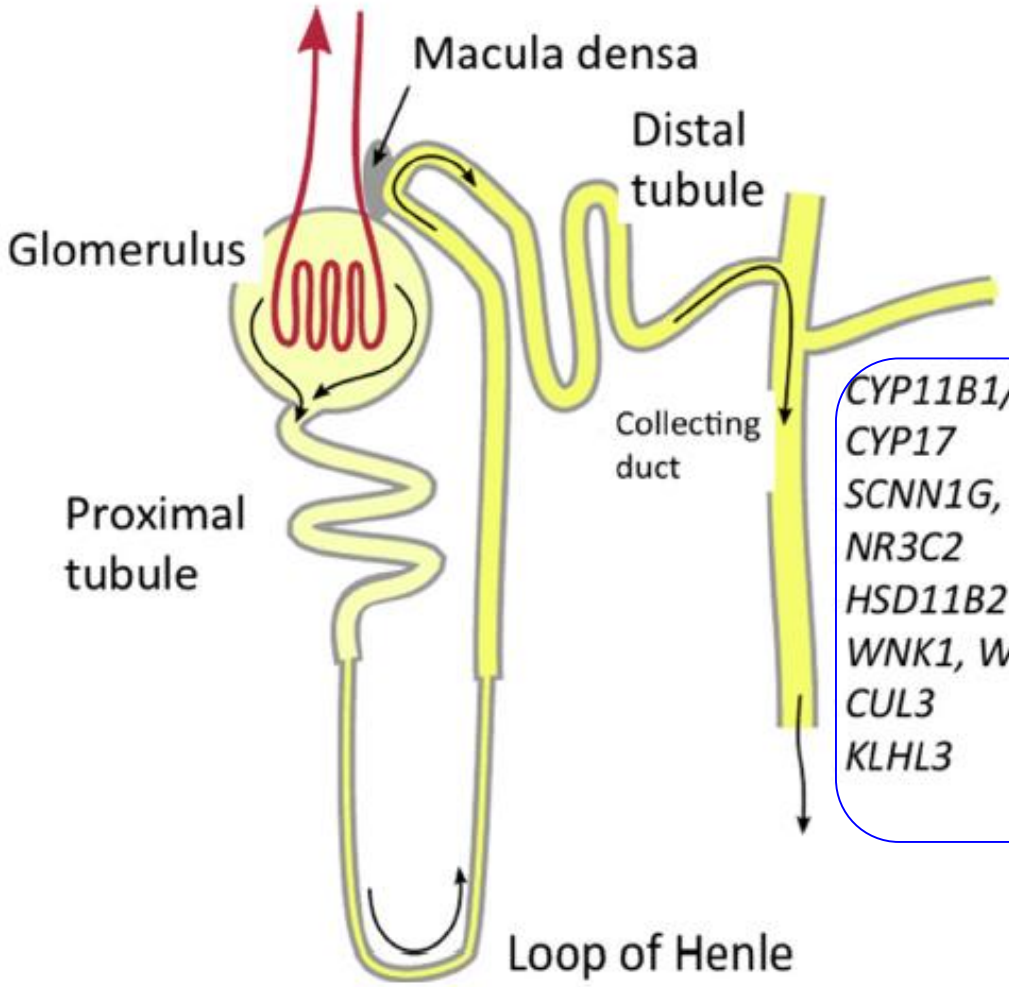
How do CNIs cause hypertension and type-4 RTA?

NCC/ENaC is the potassium switch



Hoorn E.J. et al. Nat Med 2011

The salt syndromes



GRA

ENaC
β und γ

MR

AME

WNKs

Ubiquitin Ligases

All Electrolyte and Acid-Base
All PRA/PA disturbances

11-β Hydroxylase-aldosterone-synthase chimeric gene syndromes
ENaC subunits, MR receptor-activating mutation, licorice syndrome,
the “with-no-lysine” kinases, Cullin und Kelch ubiquitin ligases that regulate WNKs

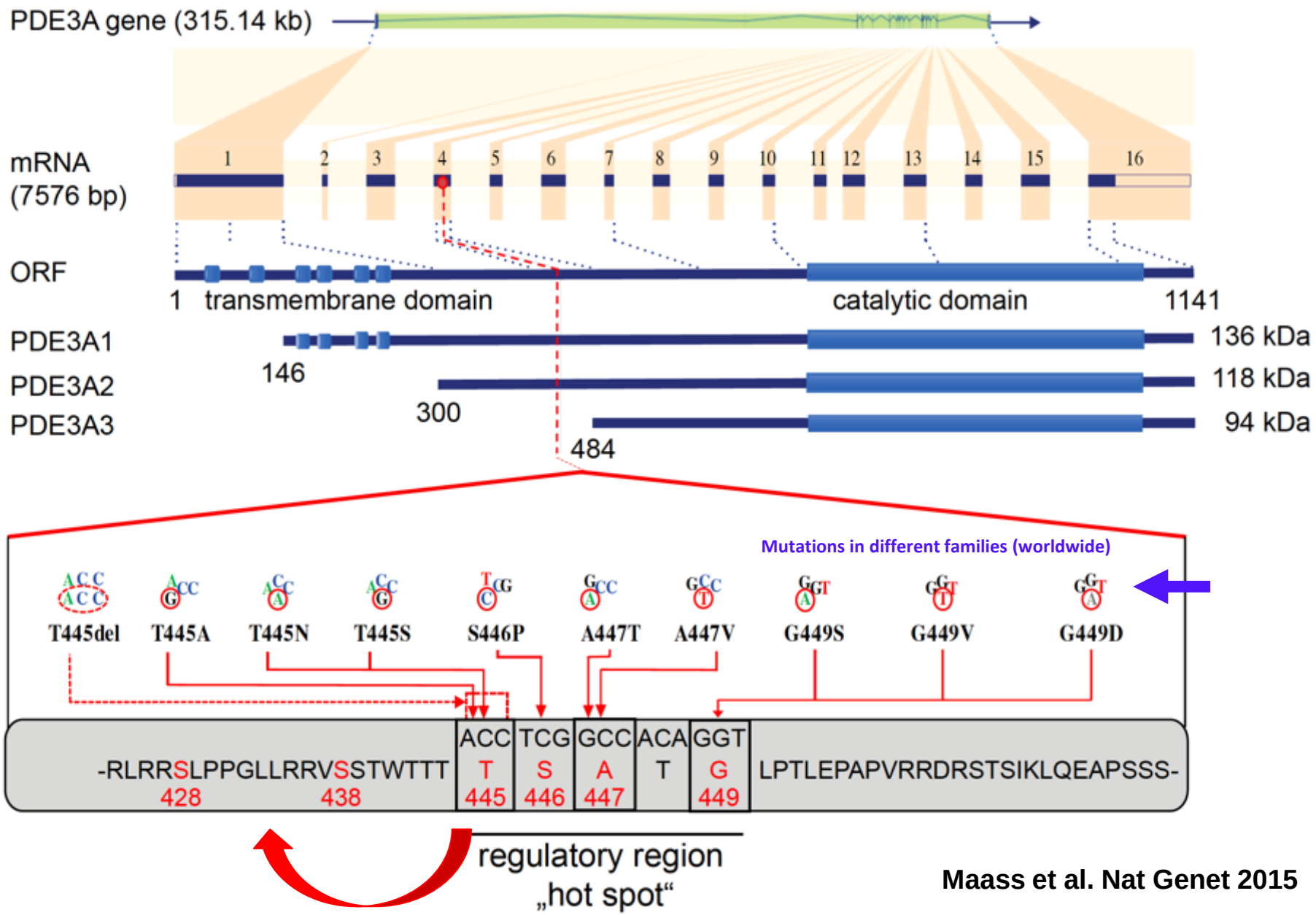
Luft. Med Clin North Am. 2017; Luft. Curr Op Cardiol 2020

1960. Kemal stood at the grave of is uncle Mehmet. An expert came from Ankara and defined the phenotypes.

- pH 7.40, $PaCO_2$ 40, PaO_2 100 (mm Hg), Na 140, K 4.7, Cl 106, HCO_3 24 (mmol/l).
- Crea 80 µmol/l, UpH 5.3, UNa 53, UK 34, UOsm 450.
- Thyroid, PRA, PA, cortisol etc were all OK.
- Family tree of 60 people, half had hypertension, men and women equally affected, inheritance from father to son.
- All of the hypertensive persons had short fingers.

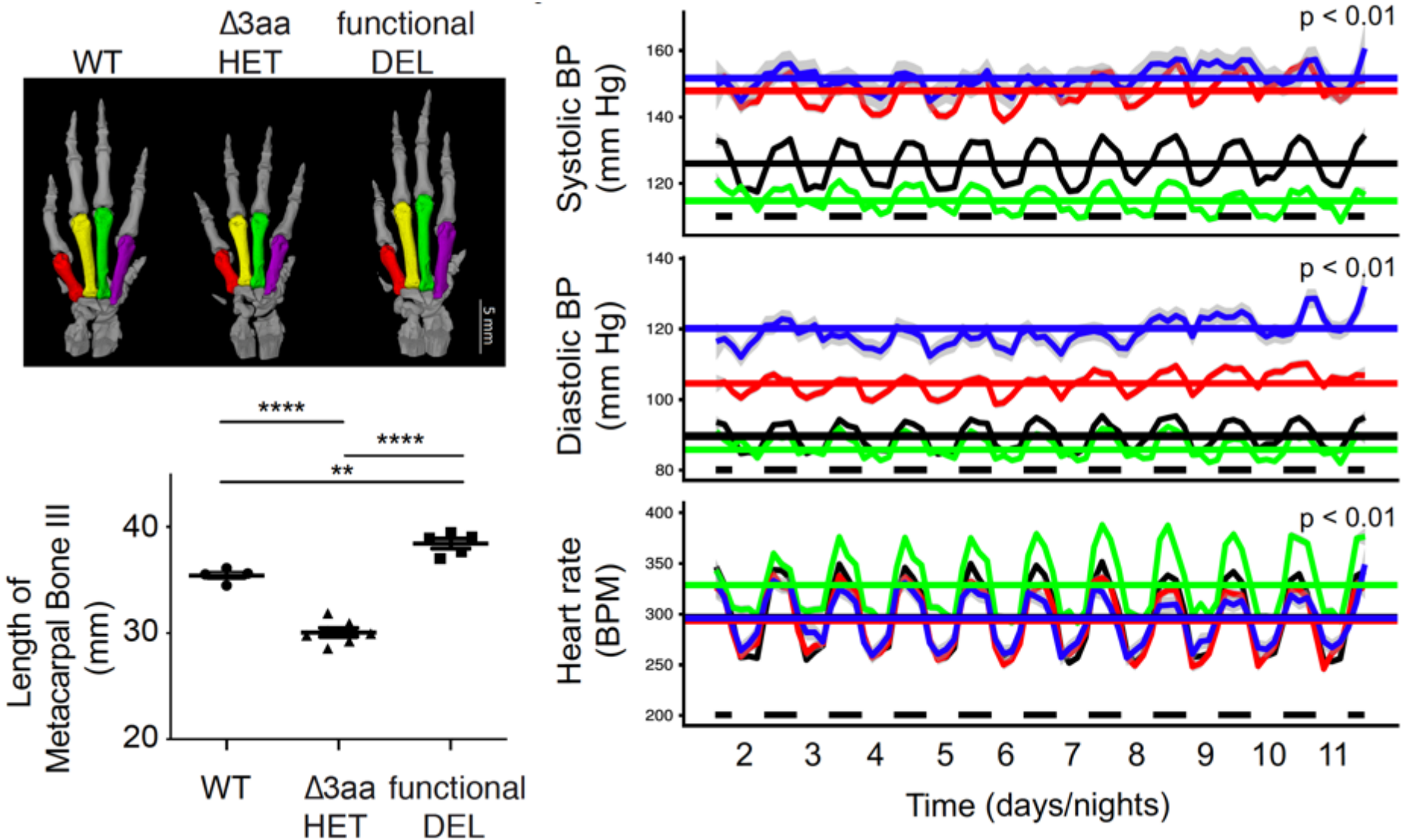
Bilginturan et al. J Med Genet 1973.

Many families worldwide



Maass et al. Nat Genet 2015

Crispr-Cas9 Rats recapitulate the syndrome!



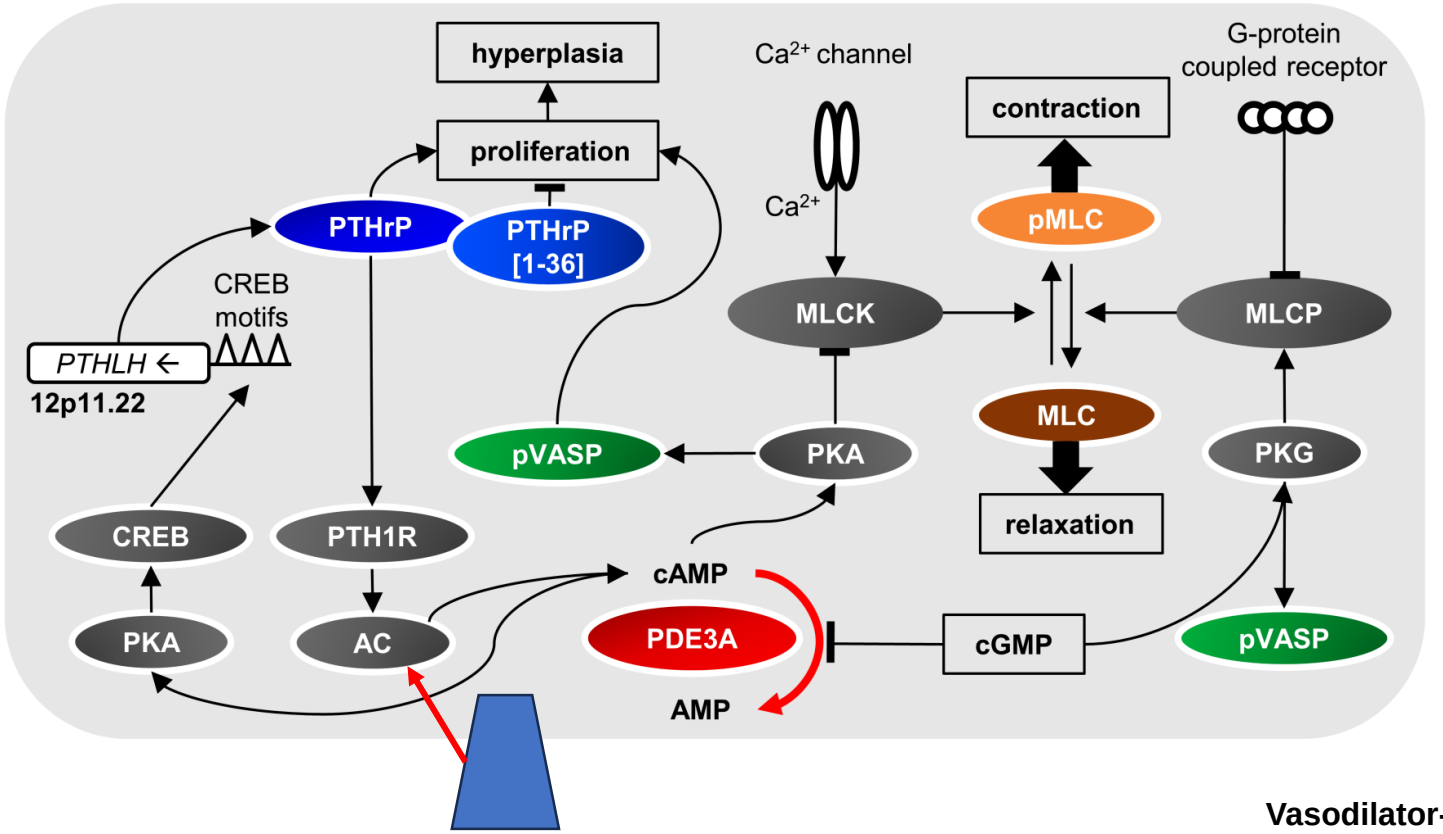
Ercu et al. Circulation 2020

How might this work?

cAMP responsive element binding protein regulates PTHrP.

PTHrP is responsible for the short fingers.

***PTHLH* encodes PTHrP.**



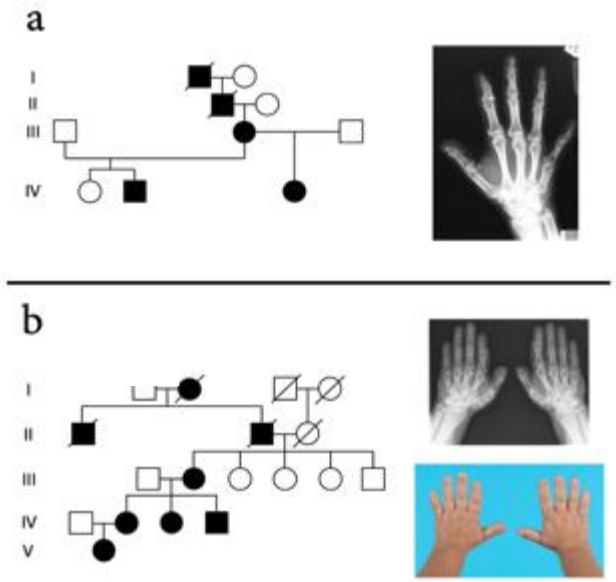
PKA, a serine—threonine kinase, resides on „anchoring proteins".

β-2 adrenergic receptor

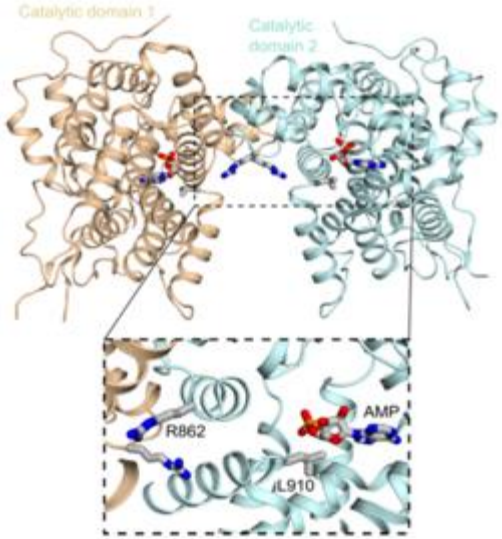
Vasodilator-stimulated phosphoprotein

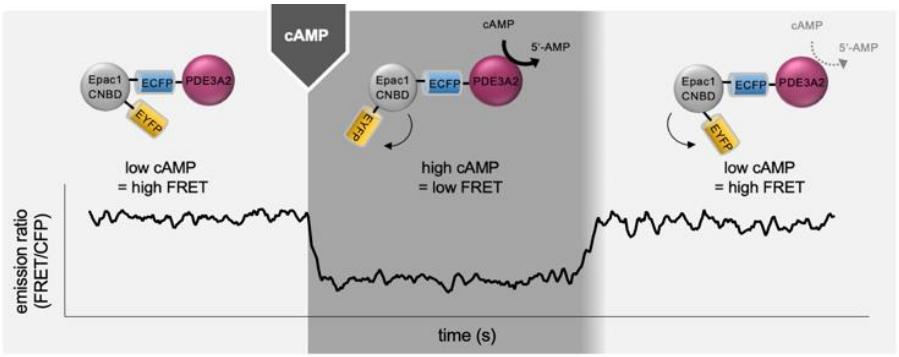
Maass et al. Nat Genet 2015

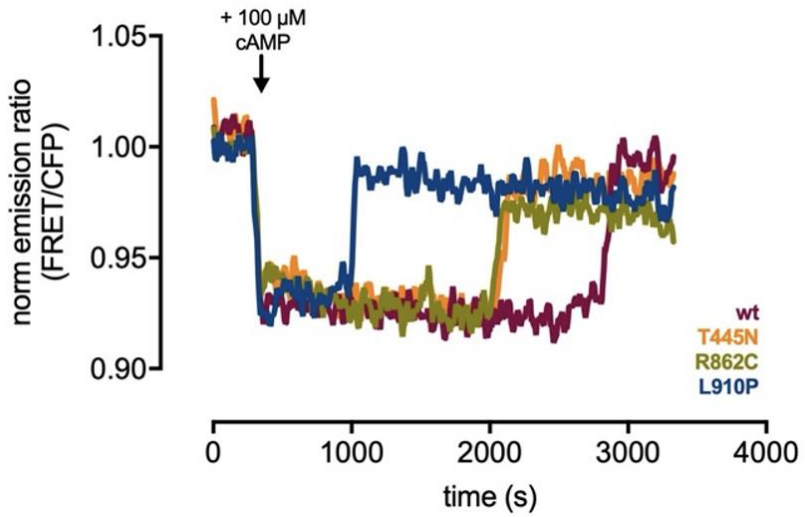
Now, to convince you!

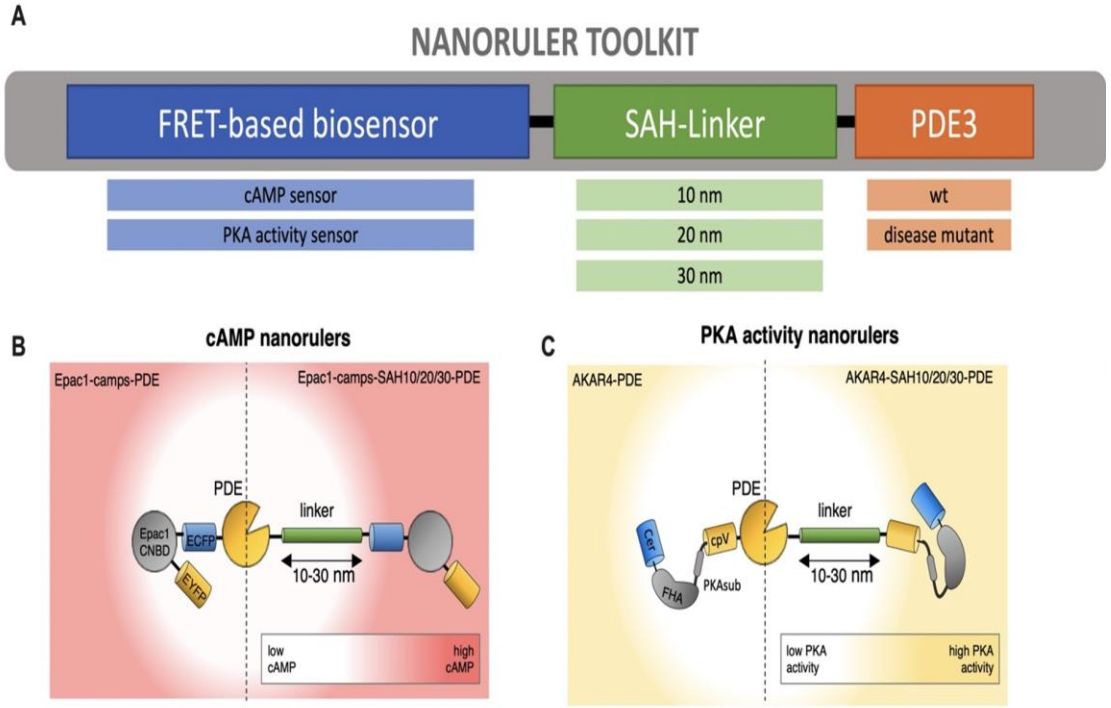


Enzymatic-pocket mutations
cause the same phenotype

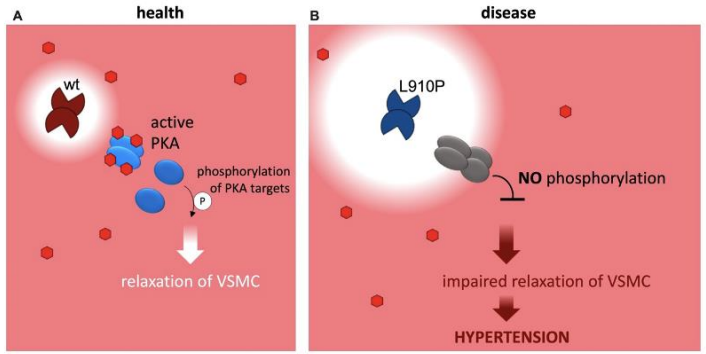


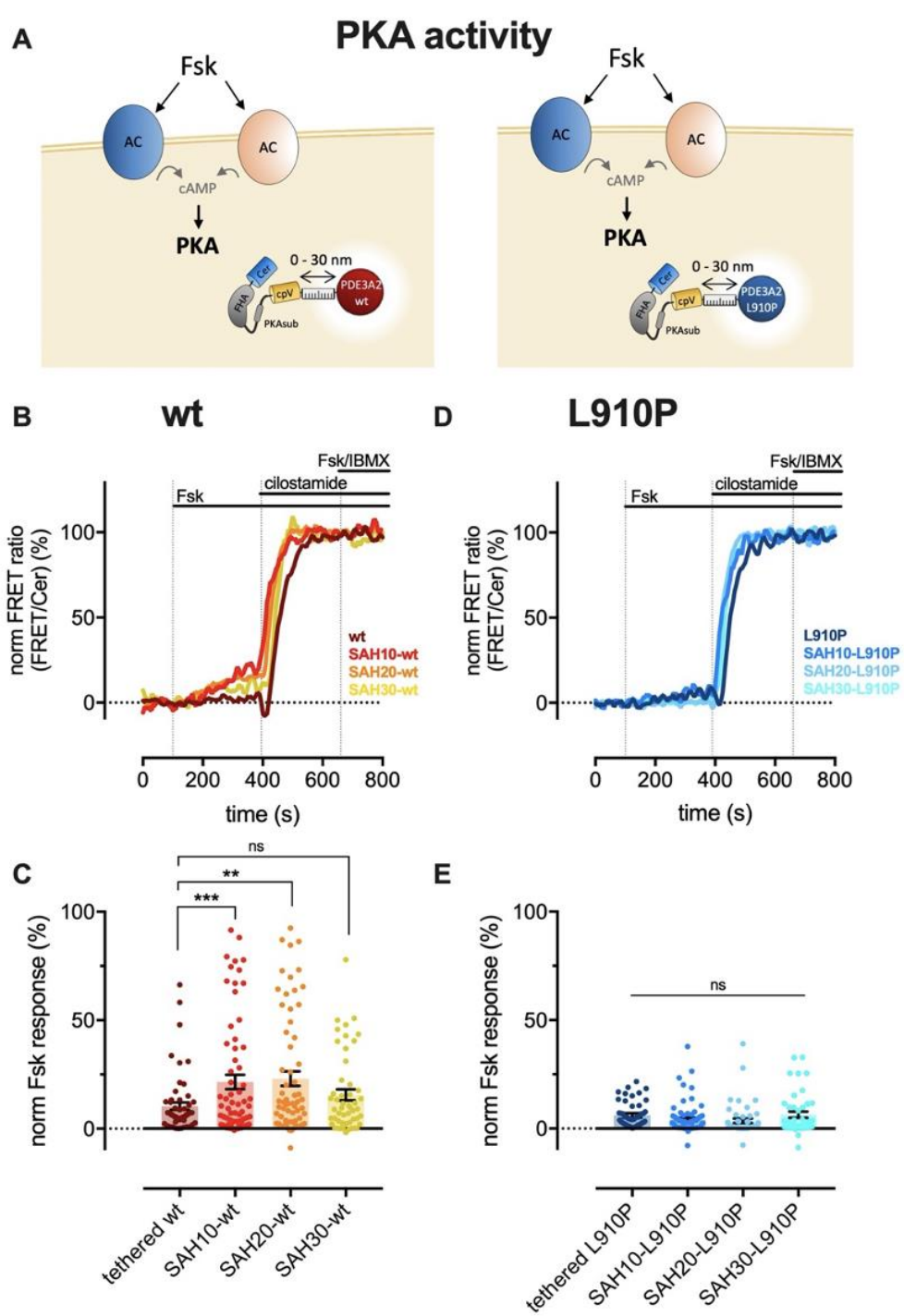






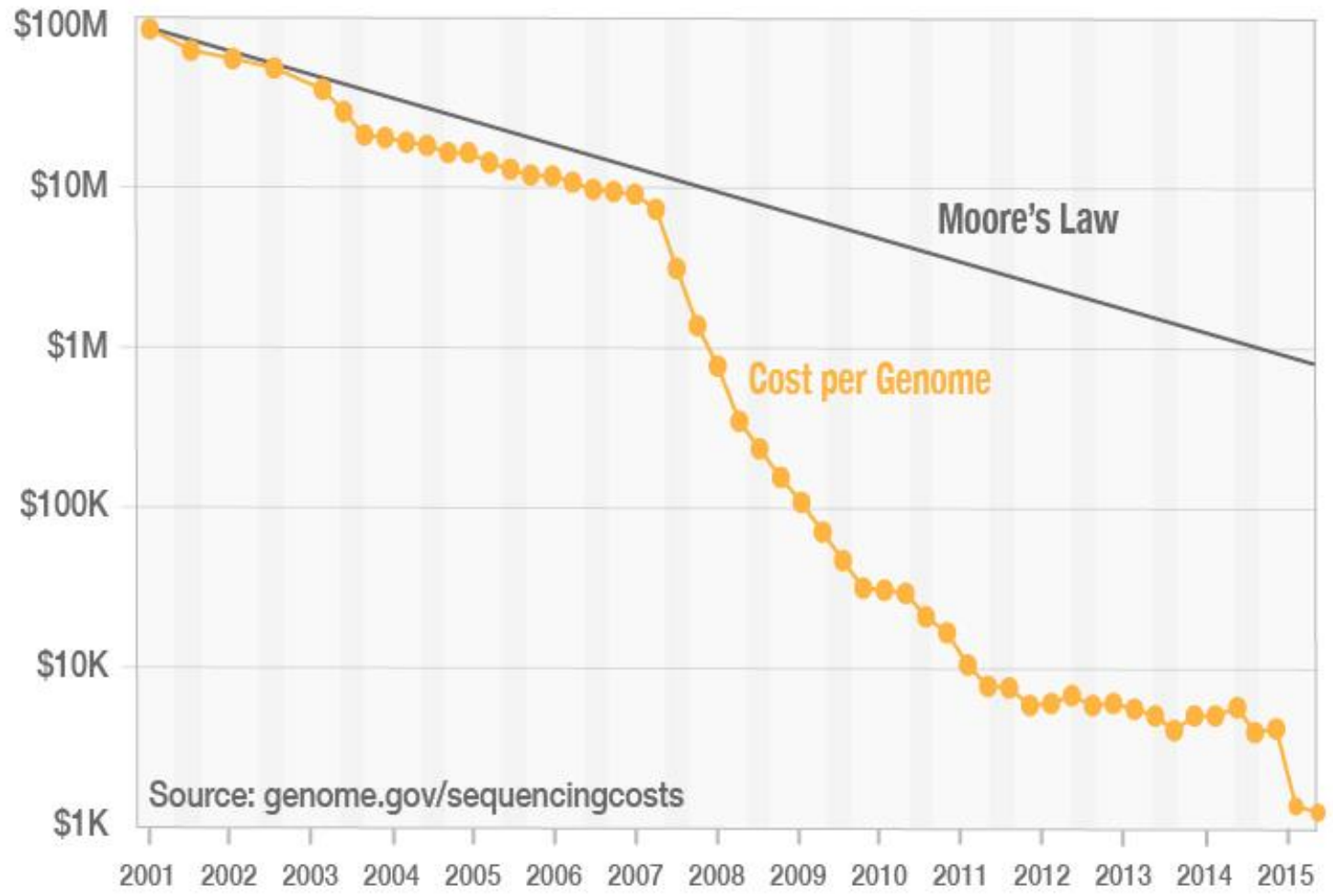
Conclusion: Altered cAMP/PKA signaling, at the nanometer scale, causes the hyper-contractile vascular phenotype and increased SVR in HTNB patients by decreasing local PKA activity in direct vicinity of L910P mutated PDE3A.

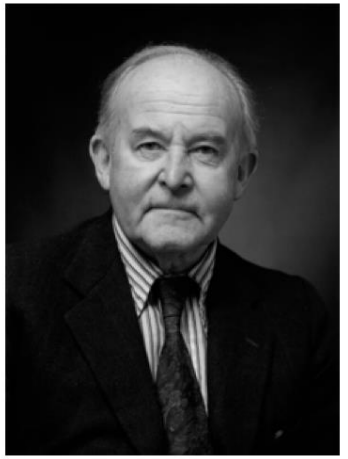




To help Sir George, a revolution next occurred.

$1000 M

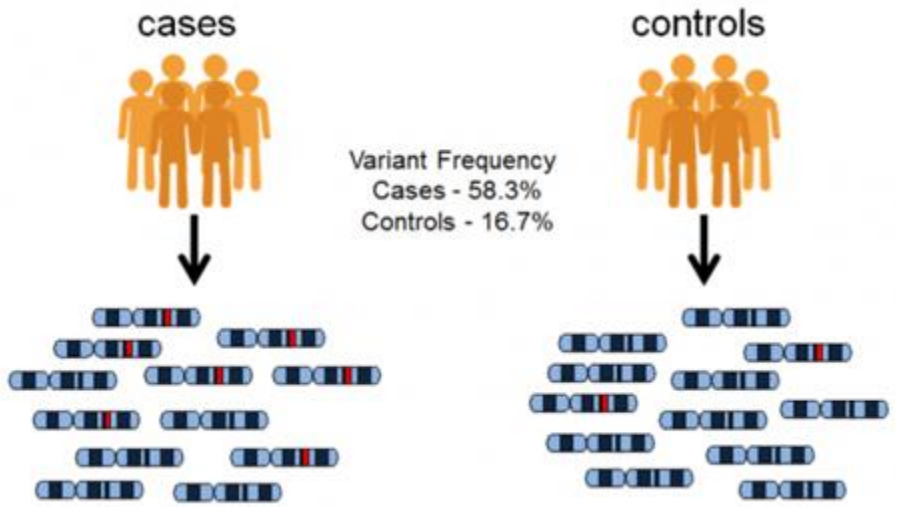


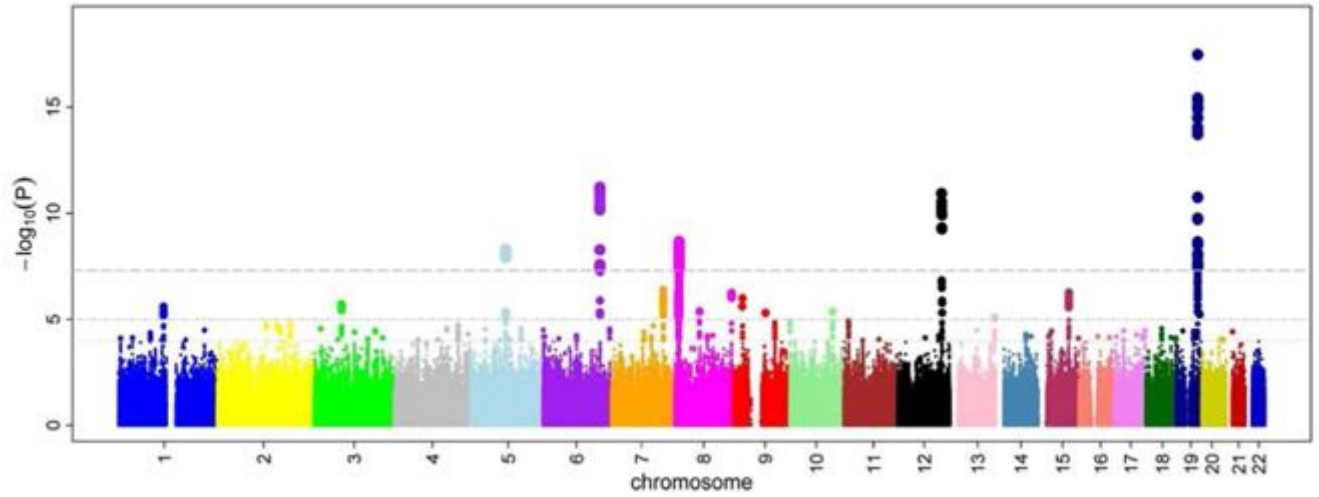


<$1000

Sequencing costs dropped „1,000,000-fold“

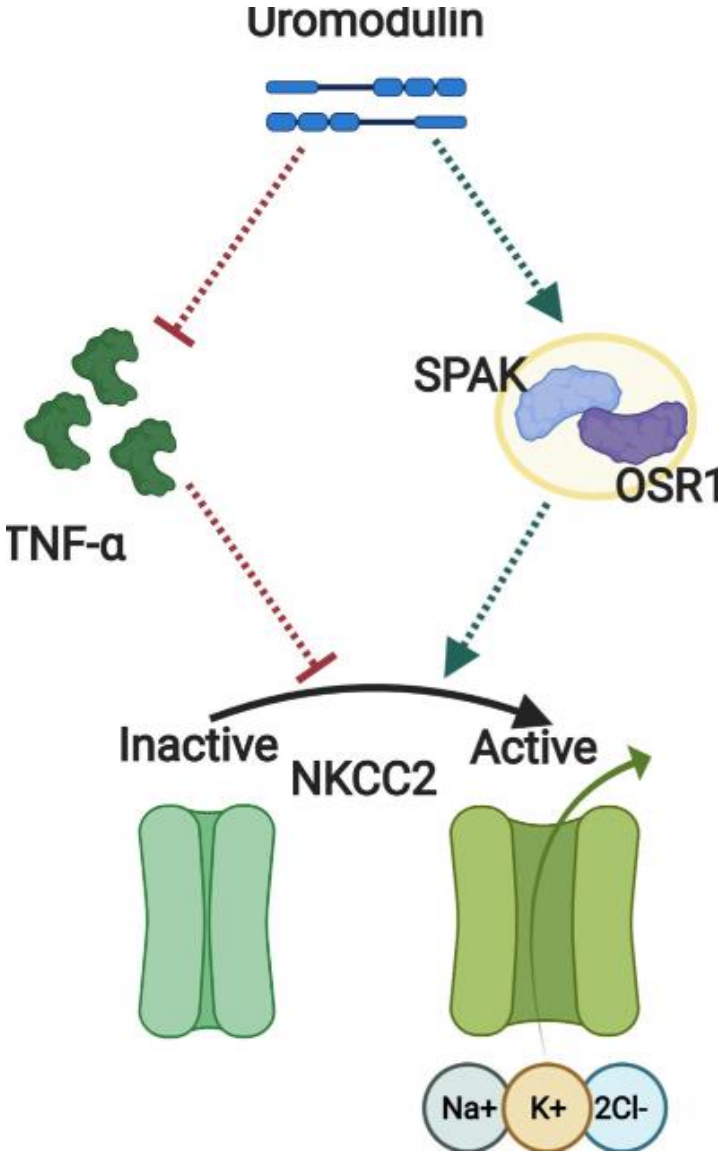
Cheap sequencing allowed for massive GWAS.





Sequencing the genome in 2000 = $1,000,000,000. In 2020 the costs were = $1,000 (1 millionth of the initial cost)

Occasionally, a relevant gene “fell out”.



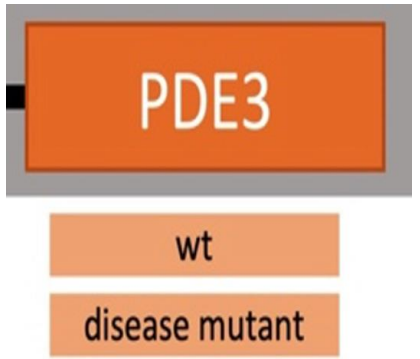
GWAS found the UMOD promoter. Peripheral evidence from Mendelian randomization studies support the relevance of UMOD. A UMOD overexpressing mouse has an increase in blood pressure.

Uromodulin regulates salt reabsorption via NKCC2 (Na+-K+-2Cl− cotransporter).

An animal model is available.

Mary et al. Hypertension 2022

Also appearing in GWAS is PDE3A



The polygenic risk score could determine the future

A bunch of SNPs

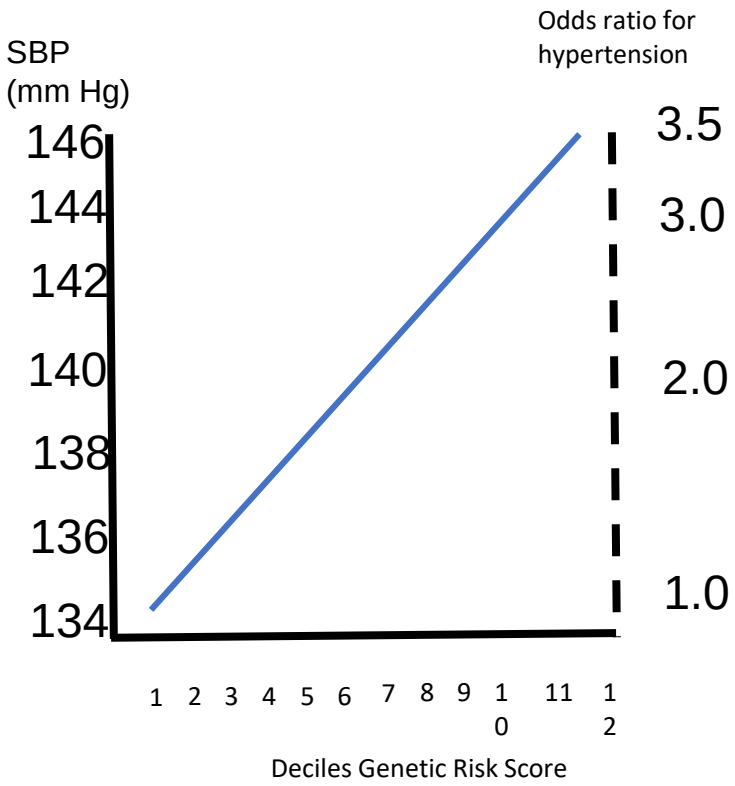
*SNP	Target allele	Allele 1	Allele 2	Genotypic score	Correlation with trait	Weighted genotypic score
SNP1	T	A	T	1	0.005	0.005
SNP2	C	G	G	0	0.004	0.000
SNP3	A	A	A	2	0.003	0.006
SNP4	G	C	G	1	0.003	0.003
SNP5	G	C	C	0	0.003	0.000
SNP6	T	A	T	1	0.002	0.002
SNP7	C	C	G	1	0.002	0.002
SNP8	A	A	A	2	0.002	0.004
SNP9	A	T	T	0	0.001	0.000
SNP10	C	C	G	1	0.001	0.001
Polygenic score				9		0.023

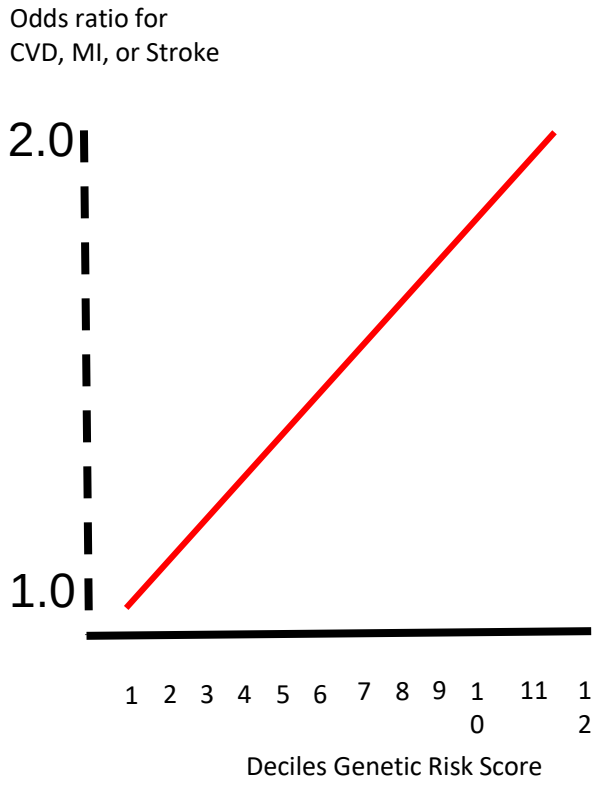
*Single nucleotide polymorphisms being included in a polygenic risk score

Polygenic risk scores can be generated for most „anything you like“.
With >3,000,000 SNPs, associations will be found.

There were about a million persons in this study

A bunch of SNPs



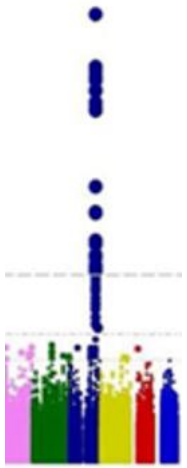


Sequencing the genome in 2000 = $1,000,000,000. In 2020 the costs were = $1,000

In your generation, everyone will be sequenced.

Evangelou et al. Nat Genet 2018 (>1,000,000 subjects and >500 loci)

Could CRISPR Purturbation Screening Offer An Answer?



A bunch of SNPs

Genomics: Genome-wide Association Studies

- >300 loci associated with CAD, >500 associated with hypertension
- Enrichment of variants at enhancers in HUVECs and aortic tissue

Transcriptomics and Epigenomics

- SNP → Gene expression (eQTLs)
- Enhancer → Gene expression (Activity-by-Contact, ABC)

CRISPR Perturbation Screens

Genetic mechanisms of EC Dysfunction

- Angiogenesis
  - PRDM16
  - FLT1
  - ARHGEF26
- Vascular tone
  - NOS3
  - EDN1
- Barrier function
  - JCAD
  - PLPP3
  - CCM2

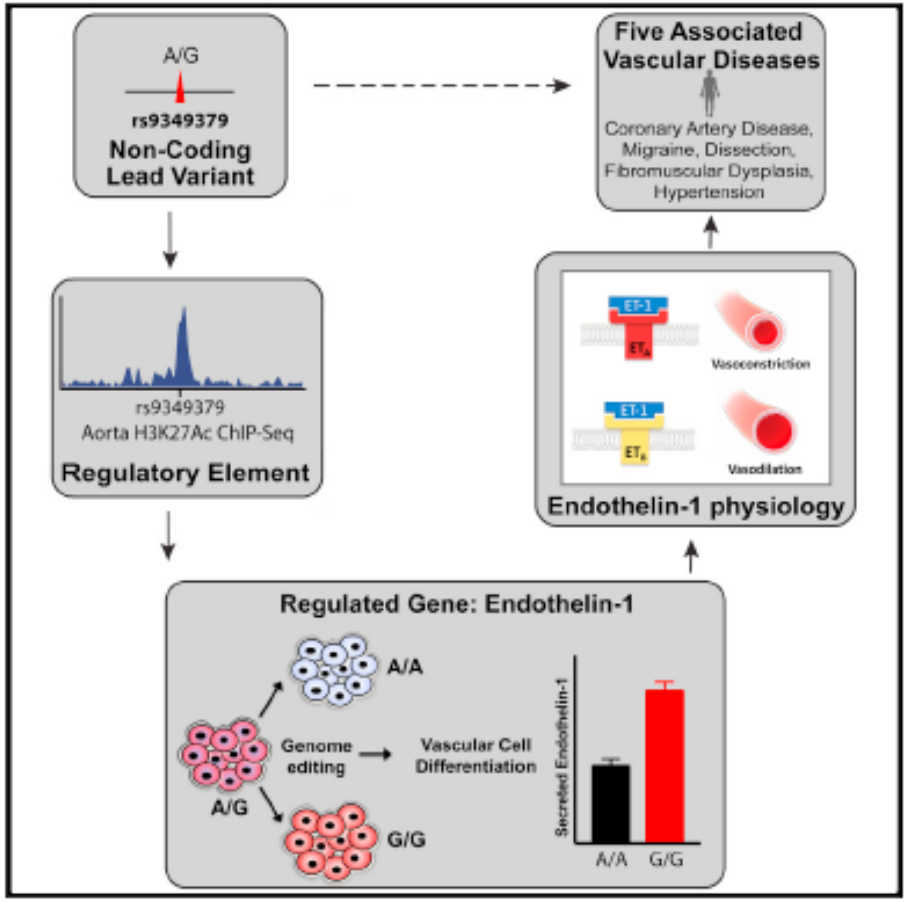
Crispr indentifies causative mechanisms in endothelial cells derived from SNPs.

Gupta et al. Arterioscler Thromb Vasc Biol. 2023;43:600–608.

Cell

A Genetic Variant Associated with Five Vascular Diseases Is a Distal Regulator of Endothelin-1 Gene Expression

Graphical Abstract



Highlights

- A non-coding variant on chromosome 6 associates with five vascular diseases
- Histone acetylation shows a vascular-specific regulatory element at the causal SNP
- Genome editing at the SNP demonstrates distal regulatory effect on endothelin-1
- Endothelin-1 signaling in blood vessels may explain pattern of vascular disease risk

Convergence of coronary artery disease genes onto endothelial cell programs

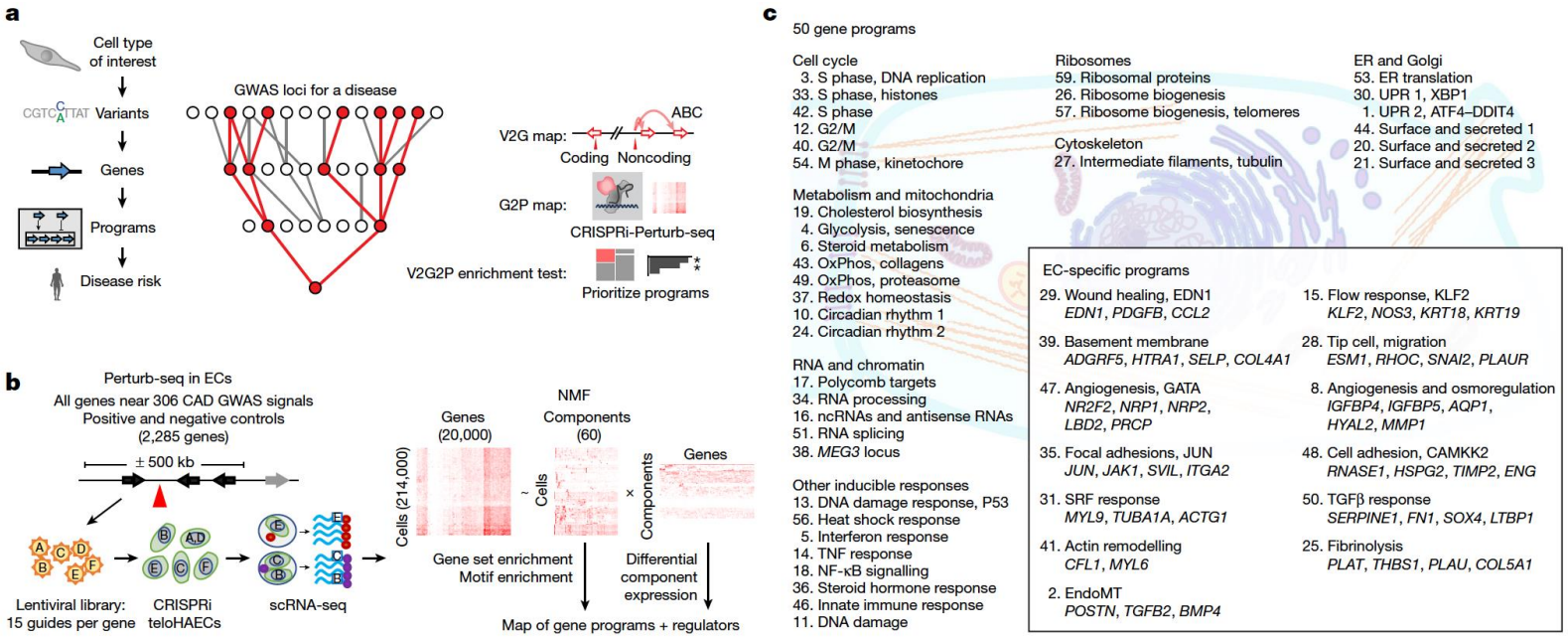
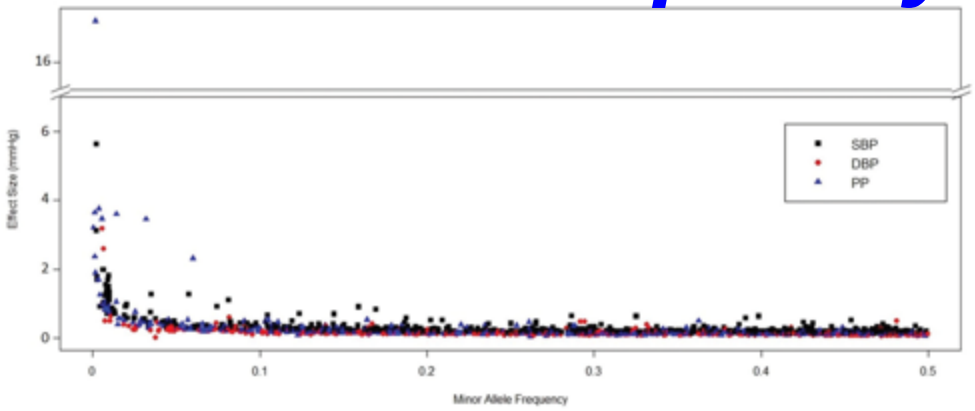
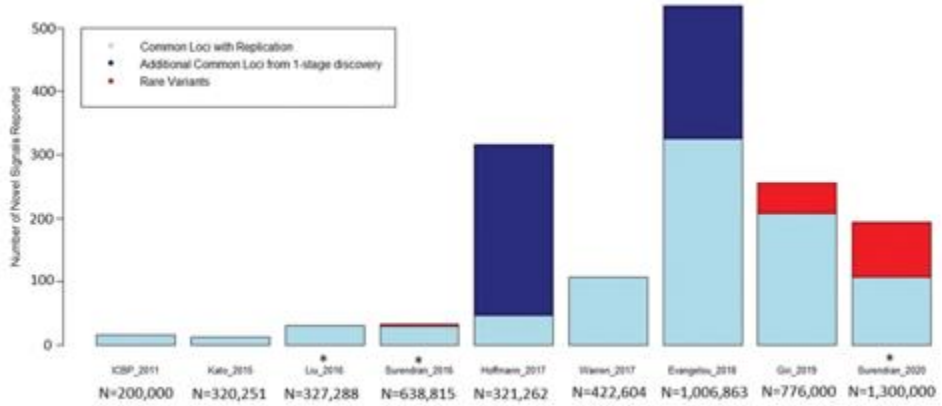


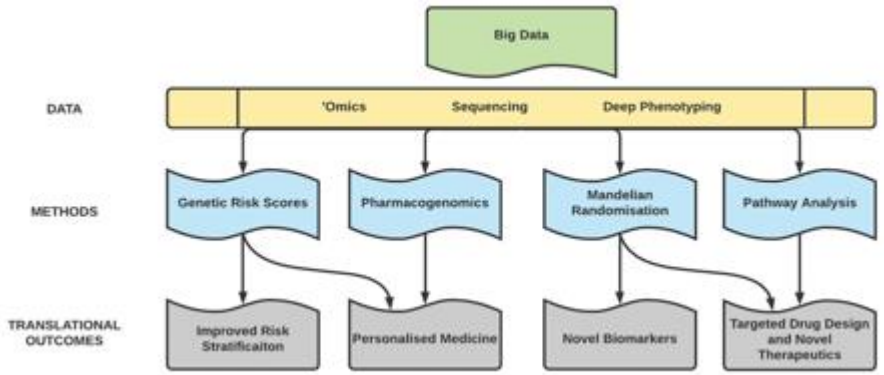
Fig. 1 | Building a map of gene programs in ECs using Perturb-seq. a, Overview of the V2G2P approach. Red nodes outline an example of the predicted convergence of multiple risk variants onto a small number of cell-type-specific programs related to disease. **b**, Diagram of the Perturb-seq G2P approach used to map genes to programs. **c**, The 50 gene programs were identified de novo from Perturb-seq and cNMF. Selected co-regulated program genes are shown in italics for the EC-specific programs.

A bunch of SNPs

Minor allele frequency versus effect size



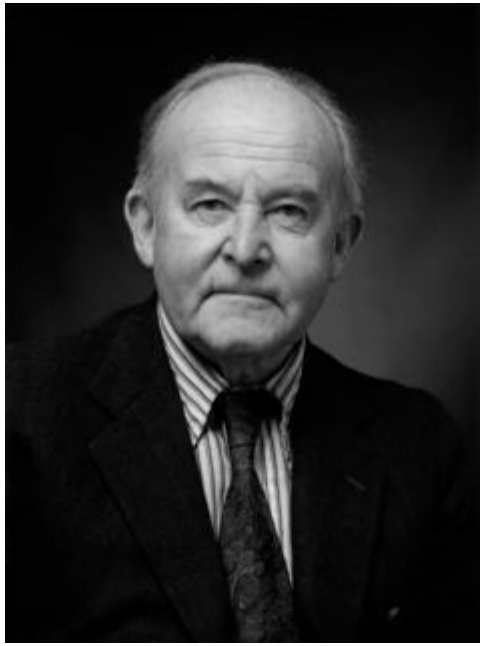




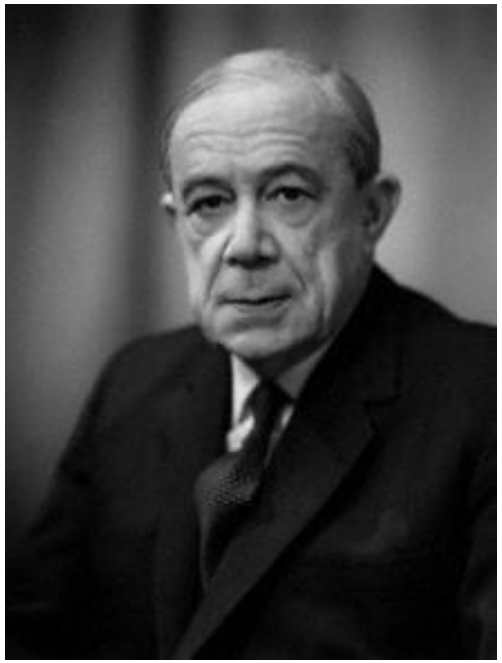
Sir Mark Jonathan Caulfield MD, FRCP, FESC, FPharm, FBHS, FMedSci is a British genomic medicine researcher.

Magavern et al. Hypertension 2021

These guys formulated the genetics of hypertension. They both were right!



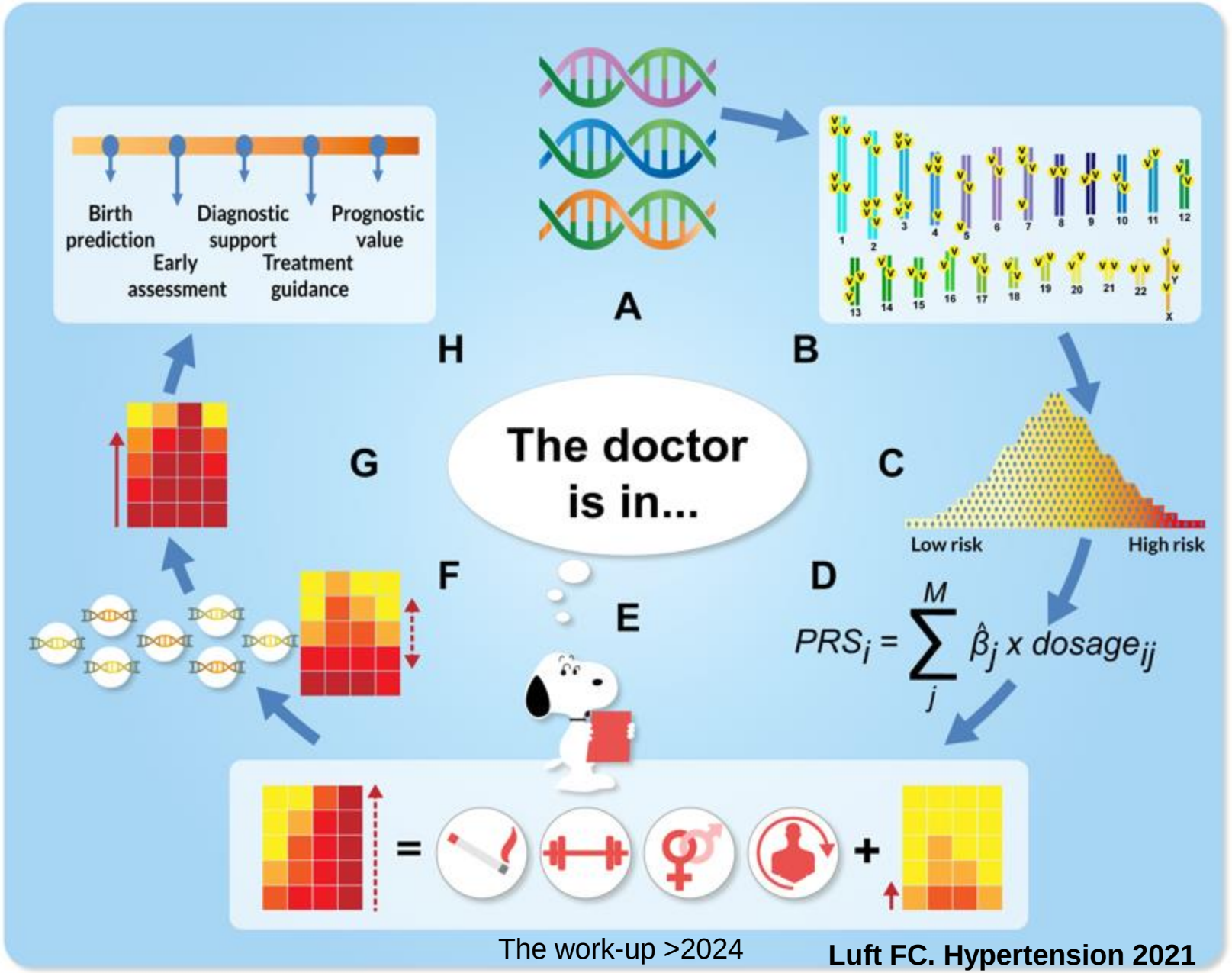
Sir George White Pickering, FRS (26 June 1904 – 3 September 1980) was a British medical doctor and academic.



Sir Robert Platt, FRCP (16 April 1900 – 30 June 1978), 1st Baronet between 1959 and 1967, was a British physician.

Sir Robert was then relegated to the House of Lords

You will not be assessing hypertension the way I did



The work-up >2024

Luft FC. Hypertension 2021