
بسم الله الرحمن الرحيم

- **Disorders of calcium sensing receptor**

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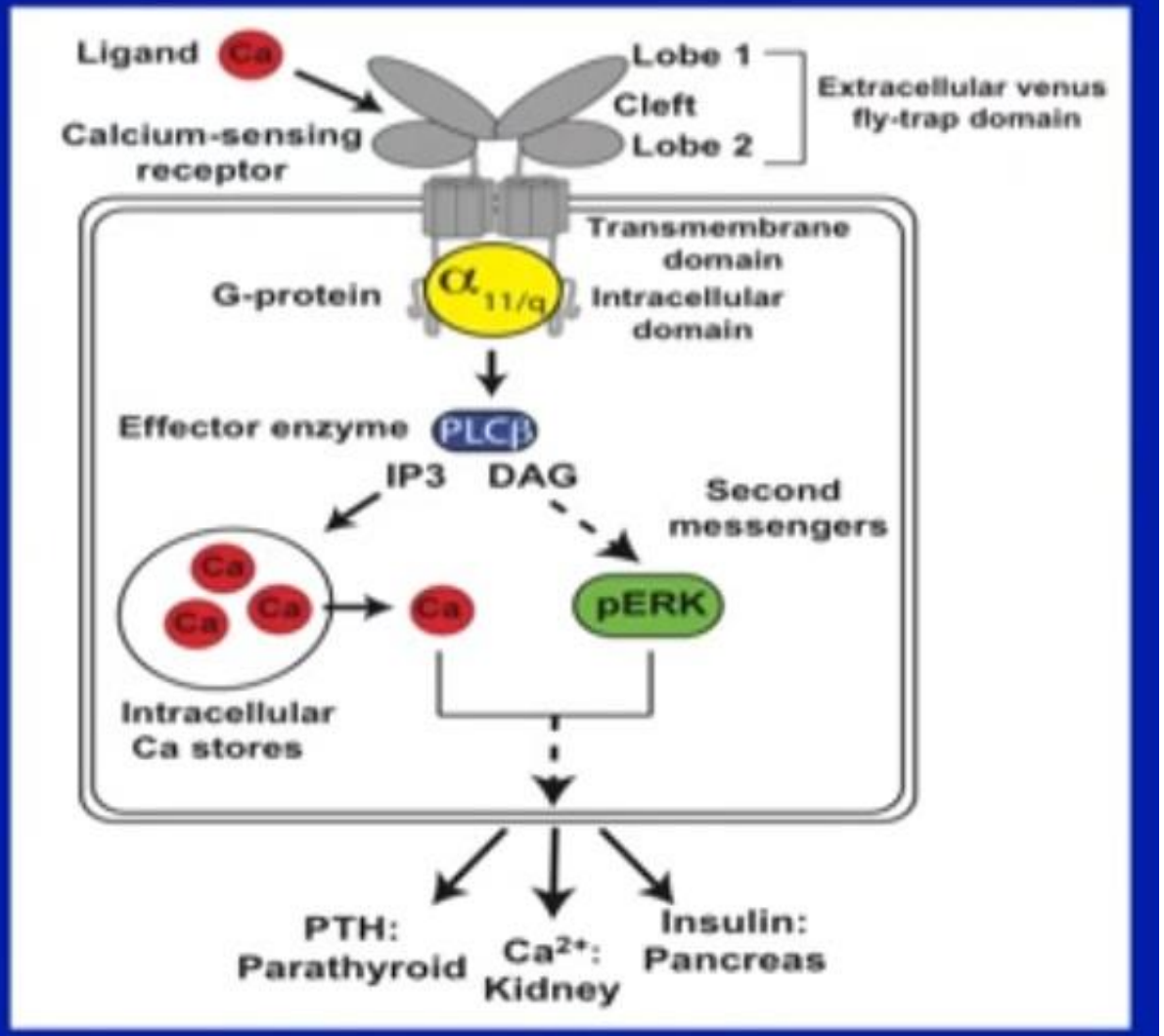
Calcium-Sensing Receptor (CaSR) – a GPCR

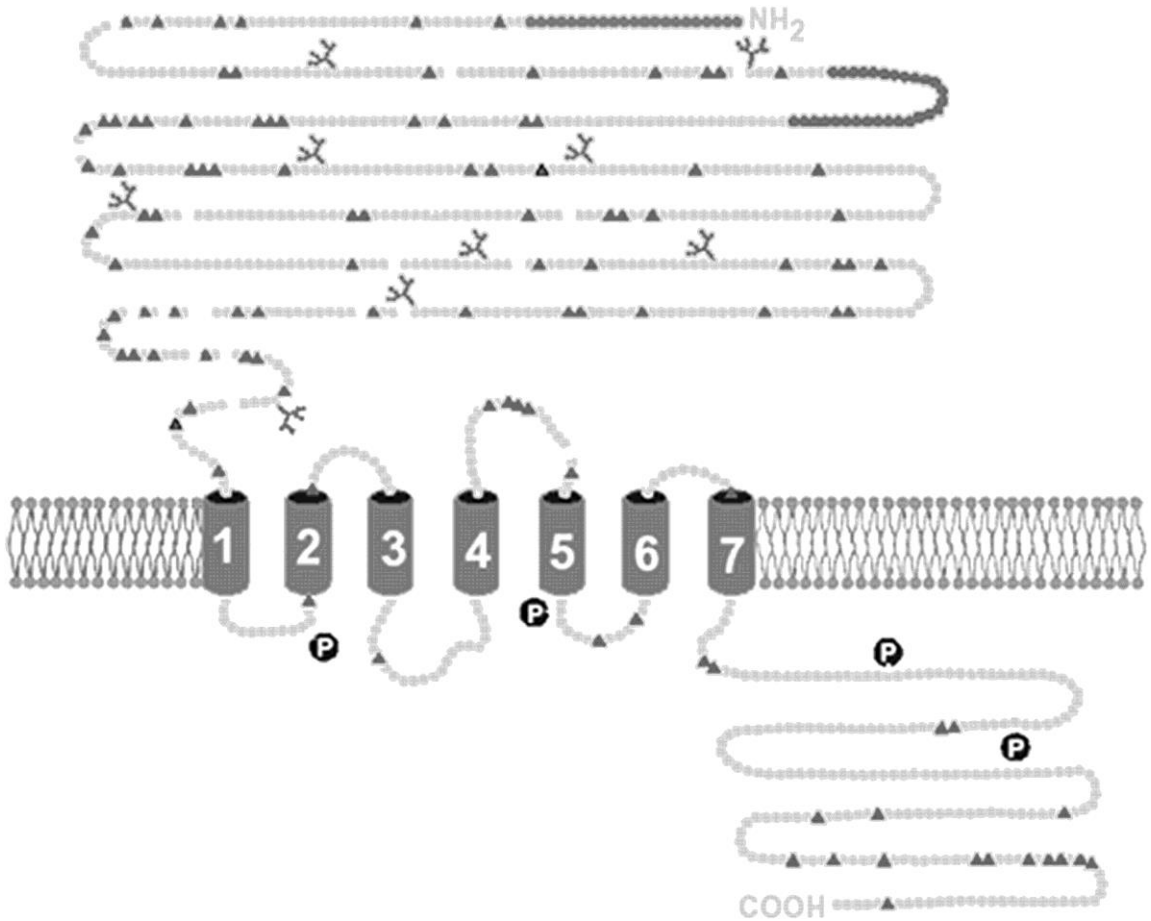
- GPCR with 3 domains : ECD (VFTD), 7 TDMs, and an ICD; forms dimers
- Ligands are cations e.g. Ca^{2+}
- Signals via Gα-proteins G11/q, G12/13, Gi/o and Gs, and different pathways e.g. IP_3 pathway to increase Ca_i^{2+} and decrease PTH expression and secretion
- Pivotal role Ca homeostasis
- Widely expressed, including parathyroids and kidneys
- Calcitrophic and non-calcitrophic roles (kidney, CNS, eye, lung, & cancers - breast, prostate and colon)



Brown et al. Nat 1993,

Brown and Macleod Phys Rev 2001

Calcium sensing receptor



Drugs - CaSR allosteric modulators

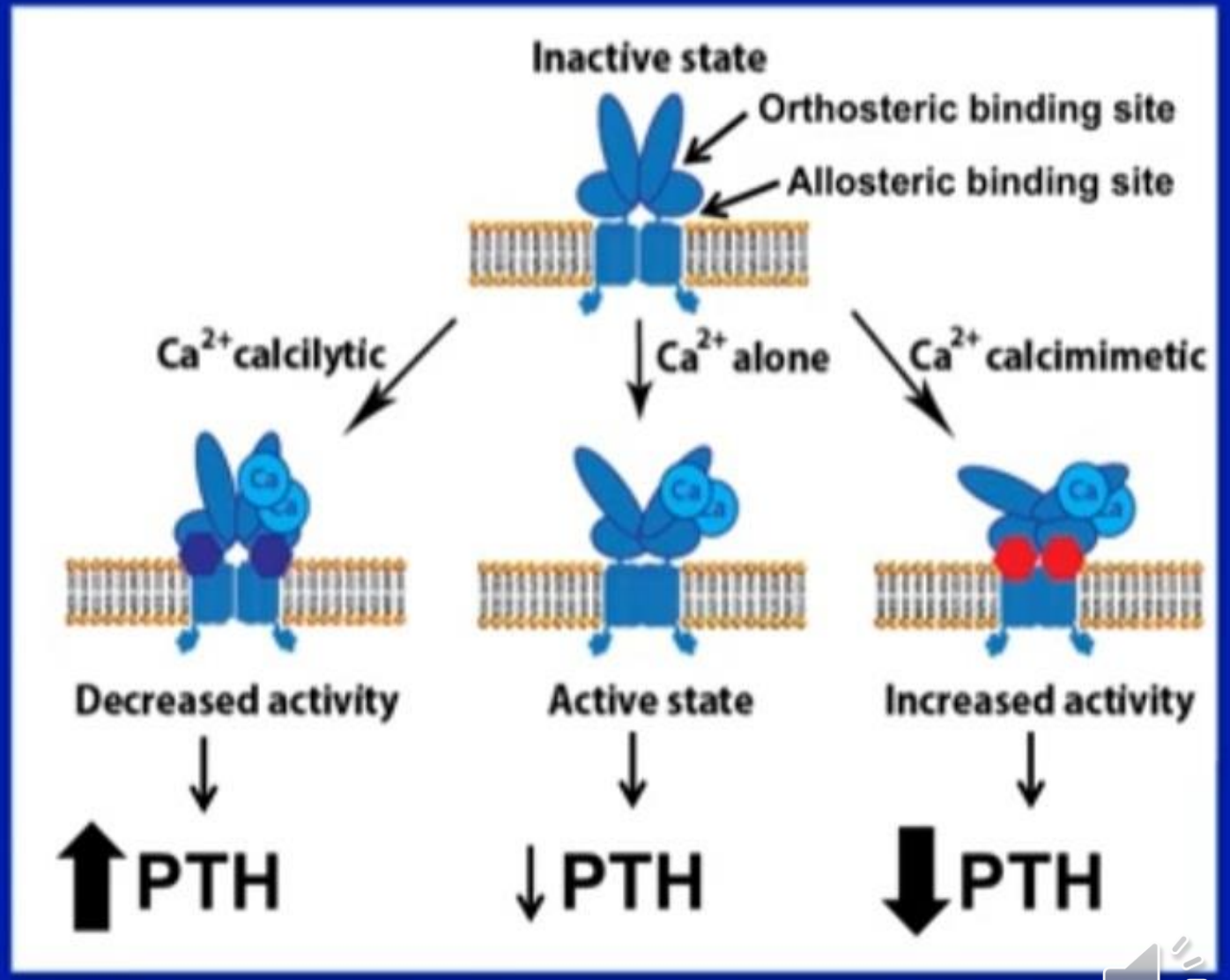
- CaSR allosteric modulators bind to the extracellular aspect of the transmembrane domain
- 2 Classes:

Positive - Calcimimetics e.g. *Cinacalcet*

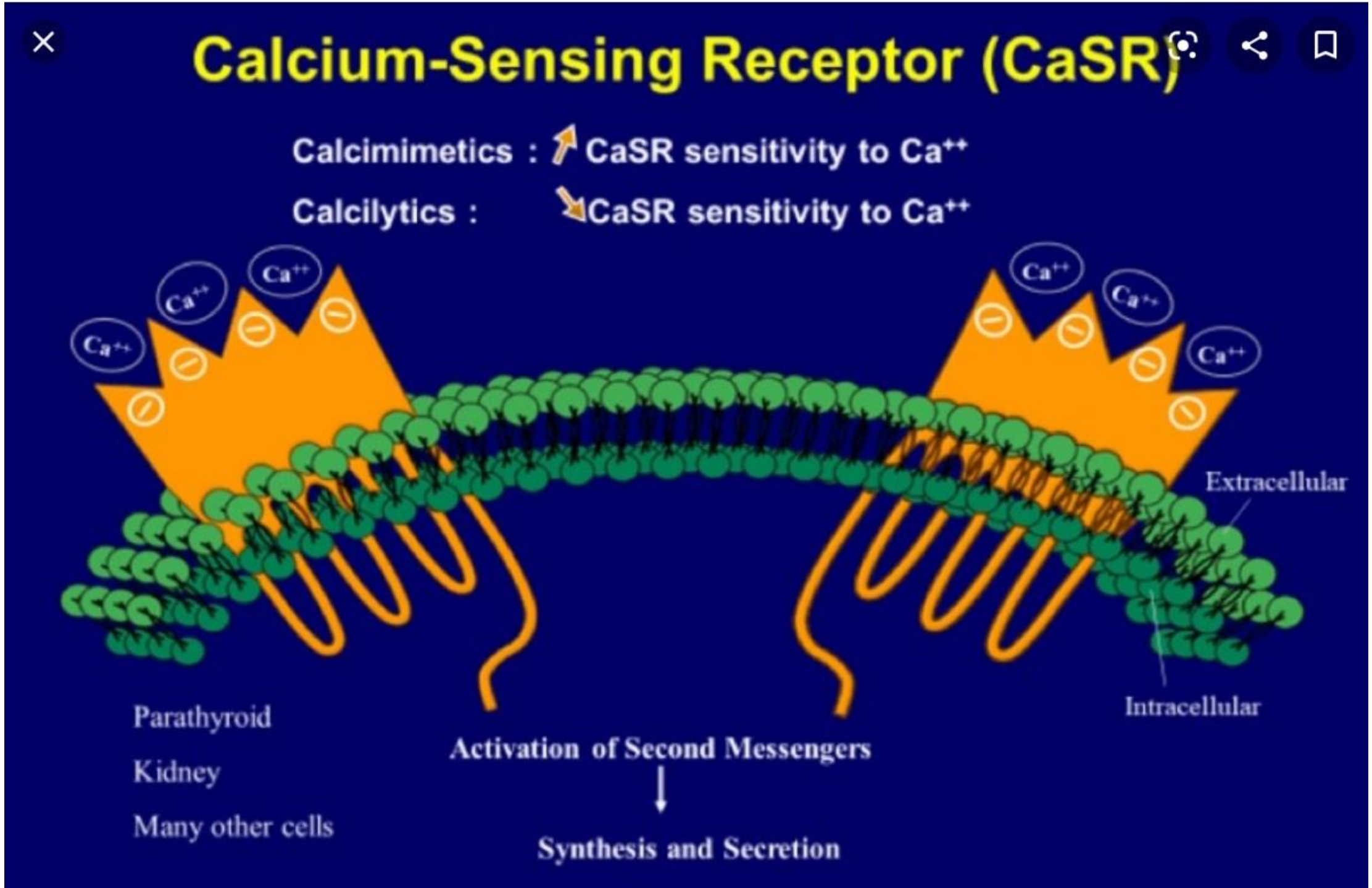
- Decrease PTH and plasma calcium
- Therapy for secondary hyperparathyroidism and parathyroid carcinoma

Negative - Calcilytics e.g. *NPS2143*

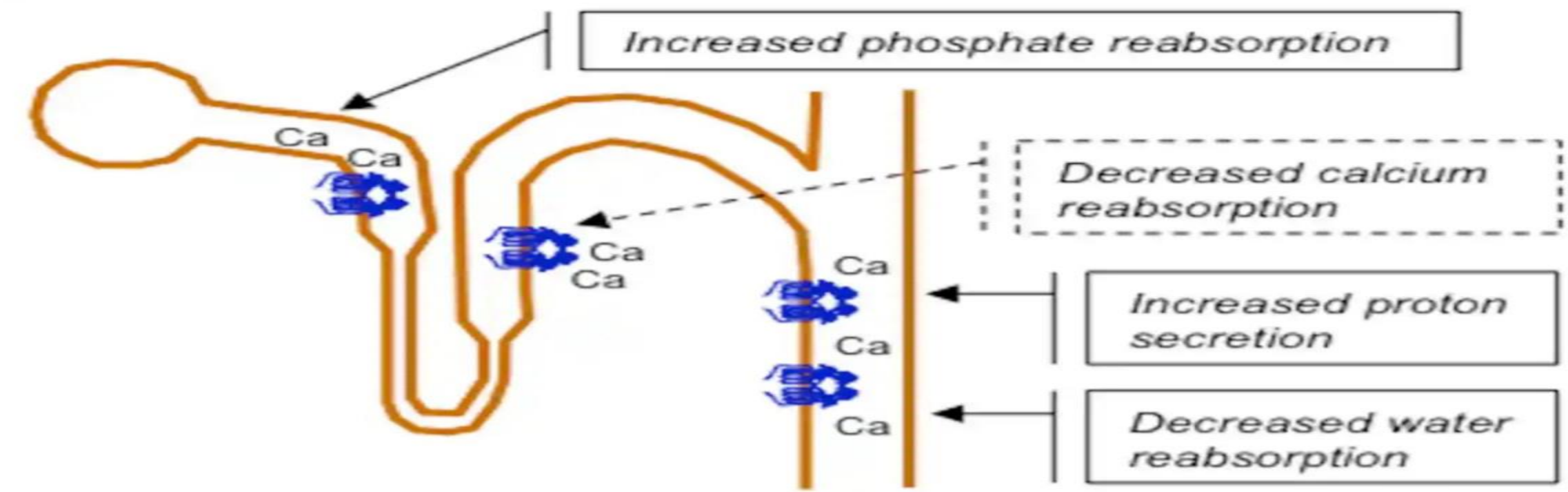
- Increase PTH and plasma calcium
- Potential therapy for hypocalcaemia of ADH(H)



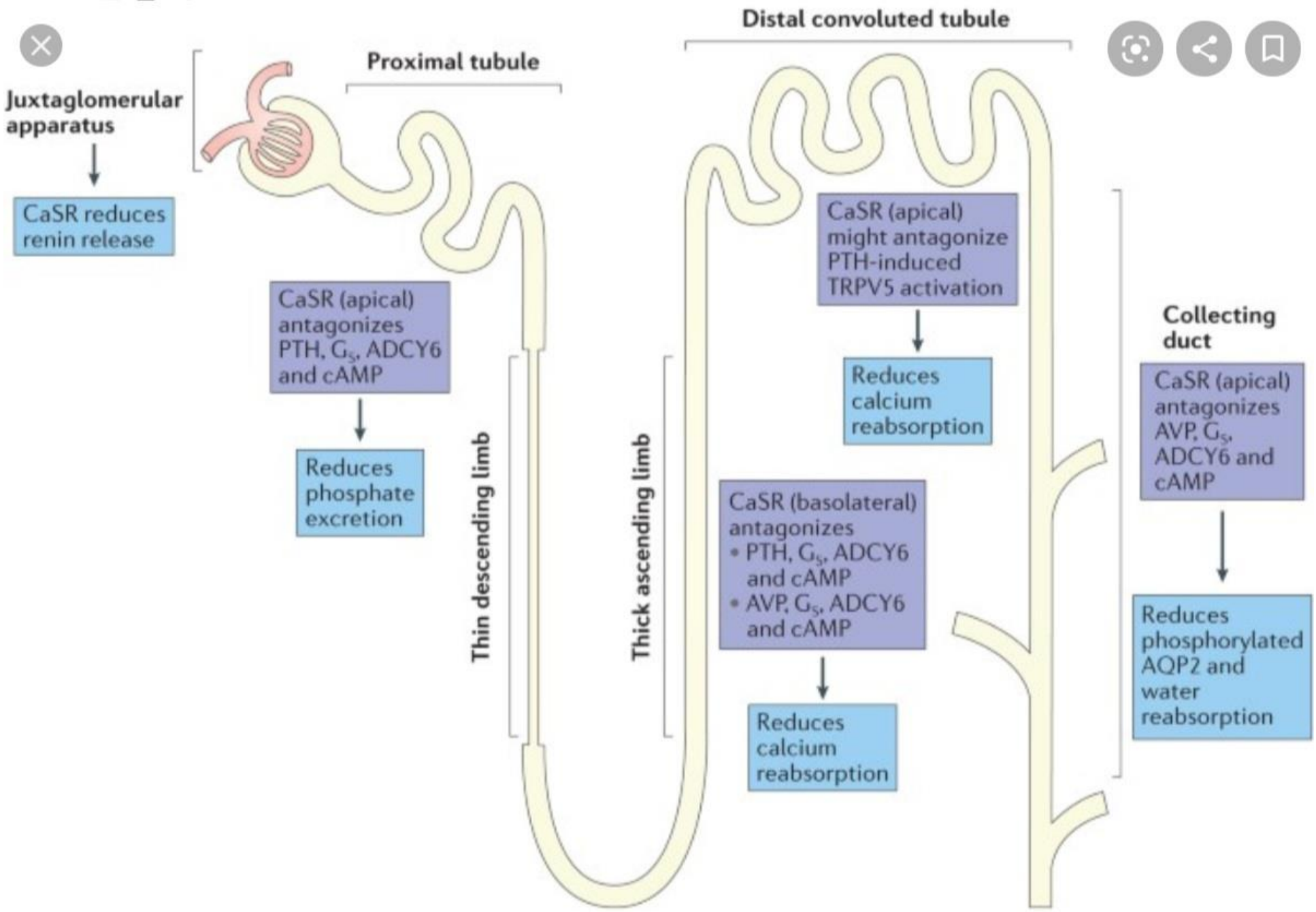
Gowen et al. J Clin Invest 2000, Goodman et al. J Am Soc Nephrol 2002, Hannan et al Endocrinology 2015


Calcium-Sensing Receptor (CaSR)
Calcimimetics : CaSR sensitivity to Ca++
Calcilytics : CaSR sensitivity to Ca++
Ca++
Ca++
Ca++
Ca++
Ca++
Ca++
Extracellular
Intracellular
Parathyroid
Kidney
Many other cells
Activation of Second Messengers
Synthesis and Secretion

Calcium sensing receptor and kidney



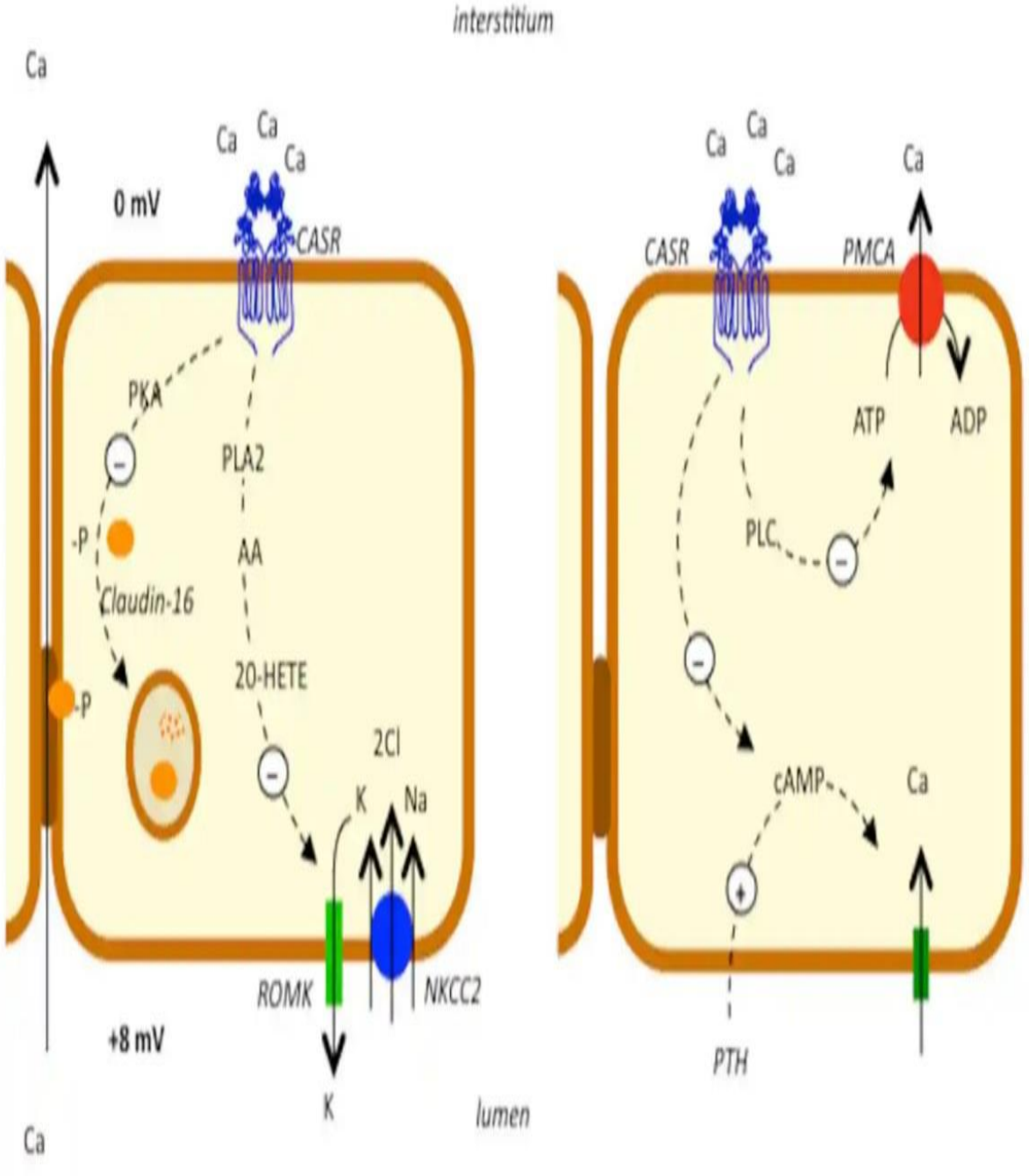
CaSR is located on the basolateral membrane of the tubular cells in the thick ascendent limb of Henle loop and in the distal convoluted cortical tubule, where it senses serum calcium and reduces calcium reabsorption when activated by serum calcium increase. In proximal tubules and collecting ducts it is located on the apical membrane and its activation by the increase in tubular fluid calcium may protect against calcium-phosphate precipitation.



Nature Reviews | Nephrology

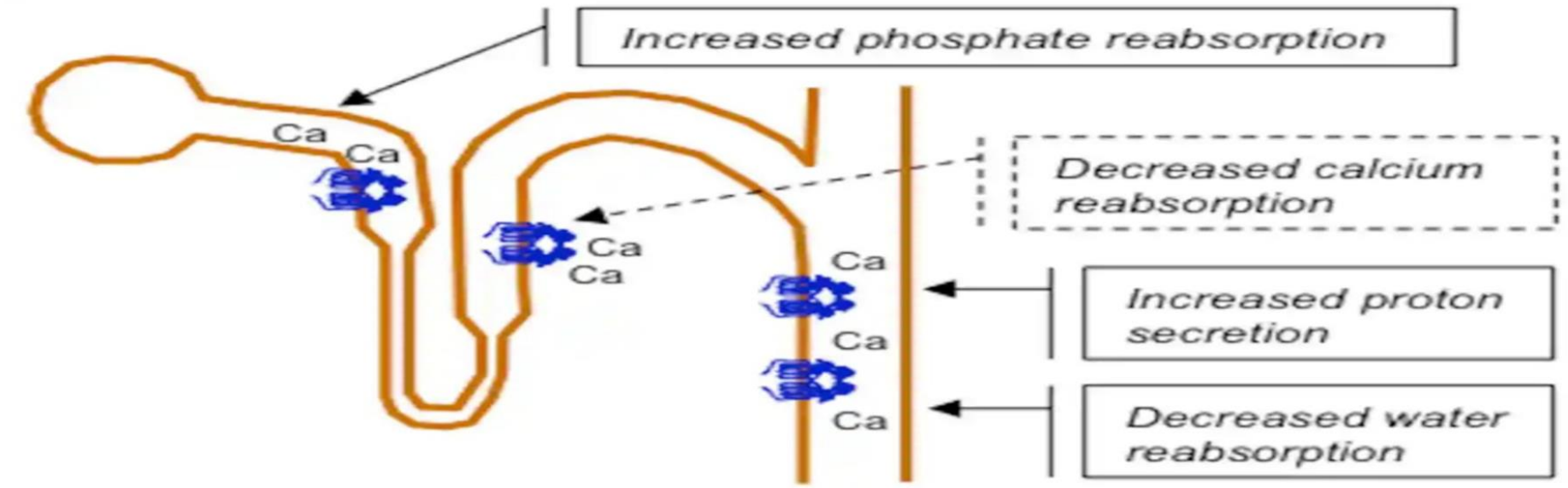
Calcium sensing receptor and kidney

Region	Cellular Target	Biological Effects
PCT/PST	↓PTH1R	↓P_i transport
	↑1-Hydroxylase activity	↑1,25$(OH)_2D_3$ synthesis
	↑p38 MAPK	↑VDR expression
MTAL	↑H^+-K^+-ATPase	↑Urine acidification
	↓Calcitonin- and AVP-induced cAMP production	↓NaCl/Ca^{2+}/Mg^{2+} transport
TAHL	↓CLDN-16	↓Ca^{2+}/Mg^{2+} transport
	↓NKCC2	↓NaCl/Ca^{2+}/Mg^{2+} transport
	↓ROMK	↓NaCl/Ca^{2+}/Mg^{2+} transport port by **inhibiting K channel**
	↓PTH-induced second messenger production	↓Transcellular Ca^{2+} transport
DCT/CNT	↑TRPV5	↑Ca^{2+} reabsorption
CCD/OMCD	↑H^+-ATPase	↑Urine acidification
OMCD/IMCD	↓AVP-dependent AQP2 apical insertion	↓Urine concentration
JG cells	↓AC-V, renin gene expression	↓Renin secretion



CaSR is expressed on the basolateral membrane in the thick ascending limb of Henle loop (left panel) and in the cortical distal convoluted tubule (right panel). In the thick ascending limb of Henle loop, CaSR inactivates the luminal potassium recycling through the ROMK channels. This effect dissipates the positive luminal electrical potential that is the driving force for passive paracellular calcium reabsorption. In this nephron segment, CaSR also inhibits the phosphorilation of caludin-16, which can be expressed in tight junctions only after phosphorilation. The absence of claudin-16 reduces the tight junction permeability to calcium and magnesium and, as a consequence, passive calcium reabsorption.In the cortical distal convoluted tubule, CaSR reduces calcium pump activity (PMCA) and calcium active reabsorption.PLA2 is phospholipase A2, AA is arachidonic acid, 20-HETE is 20-hydroxi-eicosa-tetraenoic-acid, PLC is phospholipase C, PKA is protein kinase A. Split lines express the enhancing (plus sign) or inhibitory (minus sign) pathways activated by CaSR.

Calcium sensing receptor and calcium kidney stones



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Inactivating mutations of the CaSR cause human disease

Familiar Hypocalciuric Hypercalcemia

- Heterozygous, AD
- Hypercalcemia
- Relative hypocalciuria (Ca/CrCl< 0.01)
- Normal or high plasma PTH (may be confused with 1o hyperparathyroidism)
- Asymptomatic (rare pancreatitis)

Neonatal severe hyperparathyroidism

- Homozygous for inactivating mutations of CaSR
- Severe hypercalcemia
- Severe bone demineralization
- Hyperparathyroidism
- Requires parathyroidectomy

Activating mutations of the CaSR cause human disease

Hypercalciuric hypocalcaemia syndrome type 1

- Hypocalcemia
- Relative hypercalciuria
- Manifestation of the Bartter syndrome
- Nephrocalcinosis
- Ectopic calcifications
- Nephrolithiasis

Bartter syndrome

- Type 5 Bartter syndrome due to mutations of the calcium sensing receptor
- Intrarenal defect in the TAHL
- Normally CaSR inhibits sodium transport
- Activating mutation leads to salt wasting
- Autosomal dominant

Calcium sensing receptor (CaSR) and its Seven Disorders

	Clinical Phenotype	Serum Ca^{2+}	Urinary Ca^{2+}
1	Familial benign hypocalciuric hypercalcaemia (FBHH) ^	↑	↓
2/3	Adult and Neonatal severe primary hyperparathyroidism (NSHPT) ^*	↑	↑↓
4	Autosomal dominant hypercalciuric hypocalcaemia (ADHH) ^	↓	↑
5	Bartter syndrome Type V ^	↓	↑
6	Autoimmune hypocalciuric hypercalcaemia (AHH)	↑	↓
7	Autoimmune hypoparathyroidism (AH)	↓	?

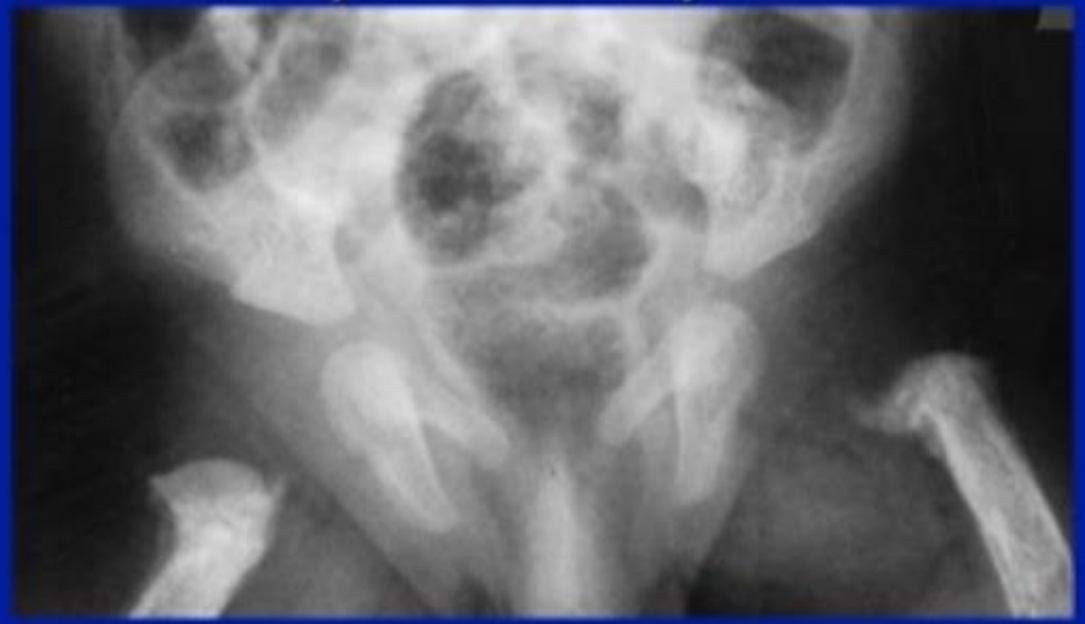
*CaSR Mutations : ^ = +/- ; * = -/-*

Thakker Cell Calcium 2004

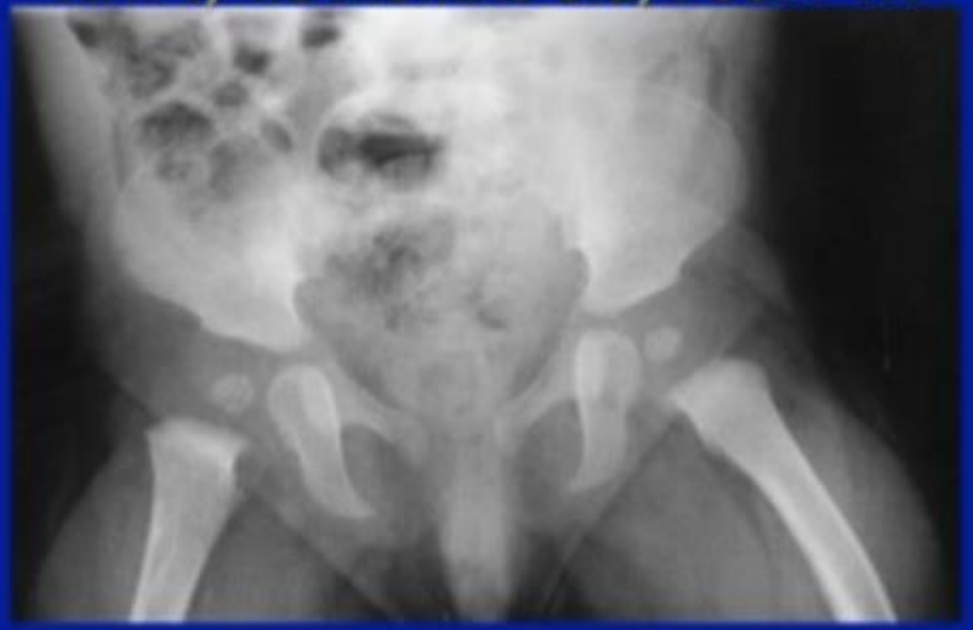
Calcium-Sensing Receptor (CaSR) Mutations cause Hypercalcaemic and Hypocalcaemic Disorders

- ***Loss-of-function*** CaSR mutations cause Familial Hypocalciuric Hypercalacemia (FHH) and Neonatal Hyperparathyroidism (NHPT), a life threatening hypercalcaemic disorder, with bone demineralisation, fractures, hypotonia and respiratory distress.

NHPT, 3 weeks old, Pre PTX



NHPT, 4.5 months old, Post PTX



- ***Gain-of-function*** CaSR mutations cause Autosomal Dominant Hypocalcaemia (ADH) with Hypercalciuria

J Clin Invest 1995, 1996,1997; NEJM 1996

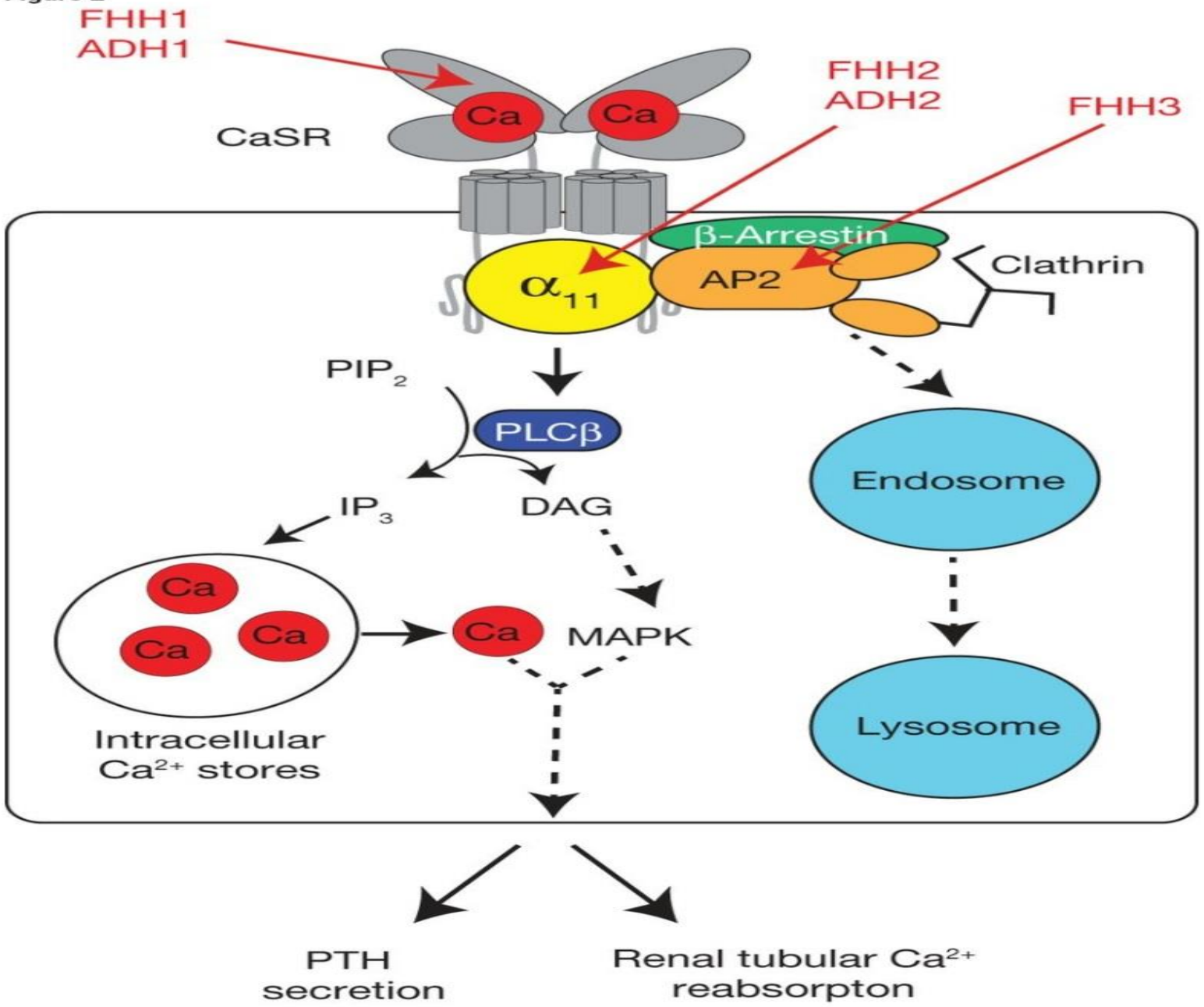
CaSR Mutations found in ≅65% FHH and ≅70% ADH Patients– Genetic Heterogeneity

FHH is genetically heterogeneous with 3 defined types - FHH1, FHH2 and FHH3 - with chromosomal locations of 3q21.1, 19p and 19q13.3, respectively.

- FHH1 is caused by loss-of-function mutations of the calcium-sensing receptor (*CaSR*) GPCR.
- FHH2 is caused by a mutation of G protein alpha 11 (*GNA11*)
- FHH3 – Due to Adaptor protein 2 sigma 2 subunit , AP2σ2,(AP2S1) mutations

Nesbit et al New Engl J Med 2013; Nesbit et al. Nature Genetics 2013

Figure 2

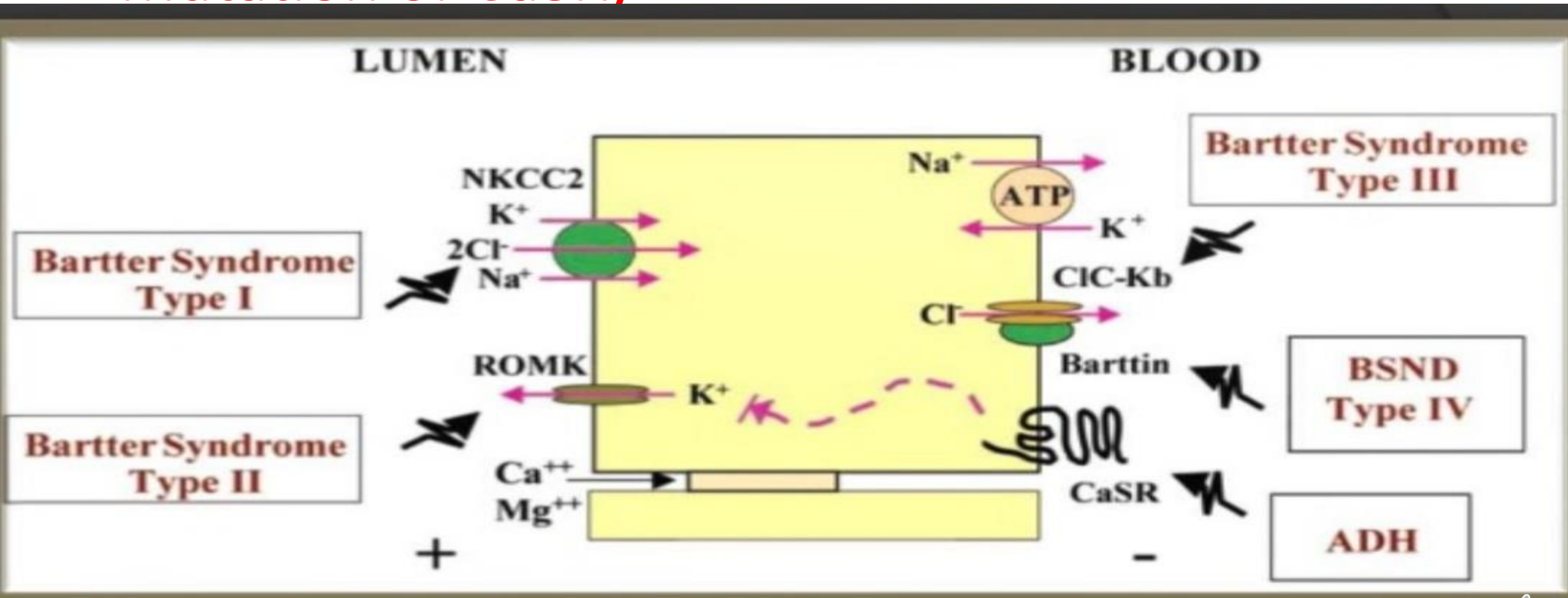

FHH1
ADH1
FHH2
ADH2
FHH3
CaSR
Ca
Ca
β-Arrestin
AP2
Clathrin
α11
PIP2
PLCβ
IP3
DAG
Endosome
Ca
Ca
Ca
Ca
MAPK
Intracellular
Ca2+ stores
Lysosome
PTH
secretion
Renal tubular Ca2+
reabsorpton

Bartter syndrome

Type	Inheritance*	Gene	Affected Protein
1	AR	SLC12A1	Sodium-potassium-chloride cotransporter (NKCC2)
2	AR	KCNJ1	Rectified potassium channel (ROMK)
3	AR	CLCNKB	Chloride channel Kb (ClC-Kb)
4	AR	BSND	Barttin protein
5	AD	CASR	Calcium-sensing receptor (CaSR)

*AR, autosomal recessive inheritance; AD, autosomal dominant inheritance.

Bartter syndrome typeV (gain of function mutation of CaSR)



Hypokalemic salt-losing tubulopathies Zelikovic Nephrology Dialysis Transplant 2003

Autosomal dominant hypocalcemia with BS

Gain-of-function mutations of *CASR* cause hypocalcemia, hypercalciuria, and salt wasting. Hypokalemia, metabolic alkalosis with hyperreninemia, as well as hyperaldosteronism are all common findings.

In the kidney, CaSR is expressed on the basolateral cell surface of the TAL. Several mechanisms have been proposed to explain the inhibitory effect of the active CaSR on paracellular divalent cation transport, which is probably related to the inhibition of NKCC2 phosphorylation and activation. Impairment on divalent calcium reabsorption with consequent renal loss of Ca^{2+} and Mg^{2+} is observed.[57]

Several mutations have been identified in the *CASR* gene, and the phenotype differs according to its type. In physiological conditions, activation of CaSR by increases in serum Ca^{2+} and Mg^{2+} concentrations inhibits divalent calcium reabsorption. The extracellular Ca^{2+} level at which normal CaSR exhibits half of its maximum activity (EC50) is 3.4 mmol/L (13.6 mg/dL). In CaSR with K47N or P221L mutations, the EC50 is around 2.0–2.2 mmol/L (8.0–8.8 mg/dL) and 0.5 mmol/L (2.0 mg/dL) for *CASR* with A843E or C131W mutations. A lower level of EC50 can induce a higher activity of CaSR and a faster onset of the phenotype. This can explain why patients with A843E or C131W mutations have the most severe symptoms of BS (Table 1).

BJP British Journal of Pharmacology
BPS BRITISH PHARMACOLOGICAL SOCIETY

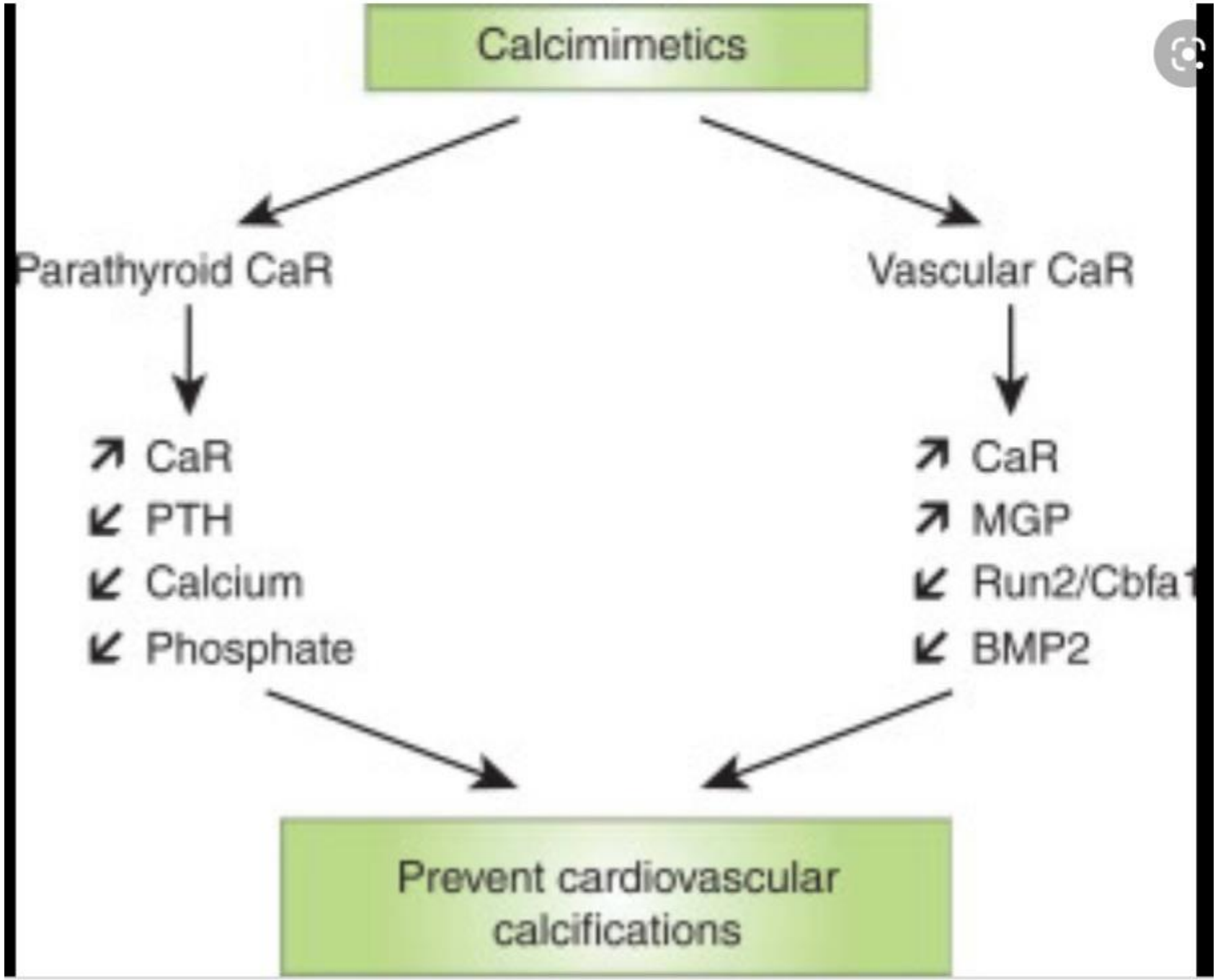
The calcium-sensing receptor and calcimimetics in blood pressure modulation

Sanela Smajilovic, Shozo Yano, [...], and Jacob Tfelt-Hansen

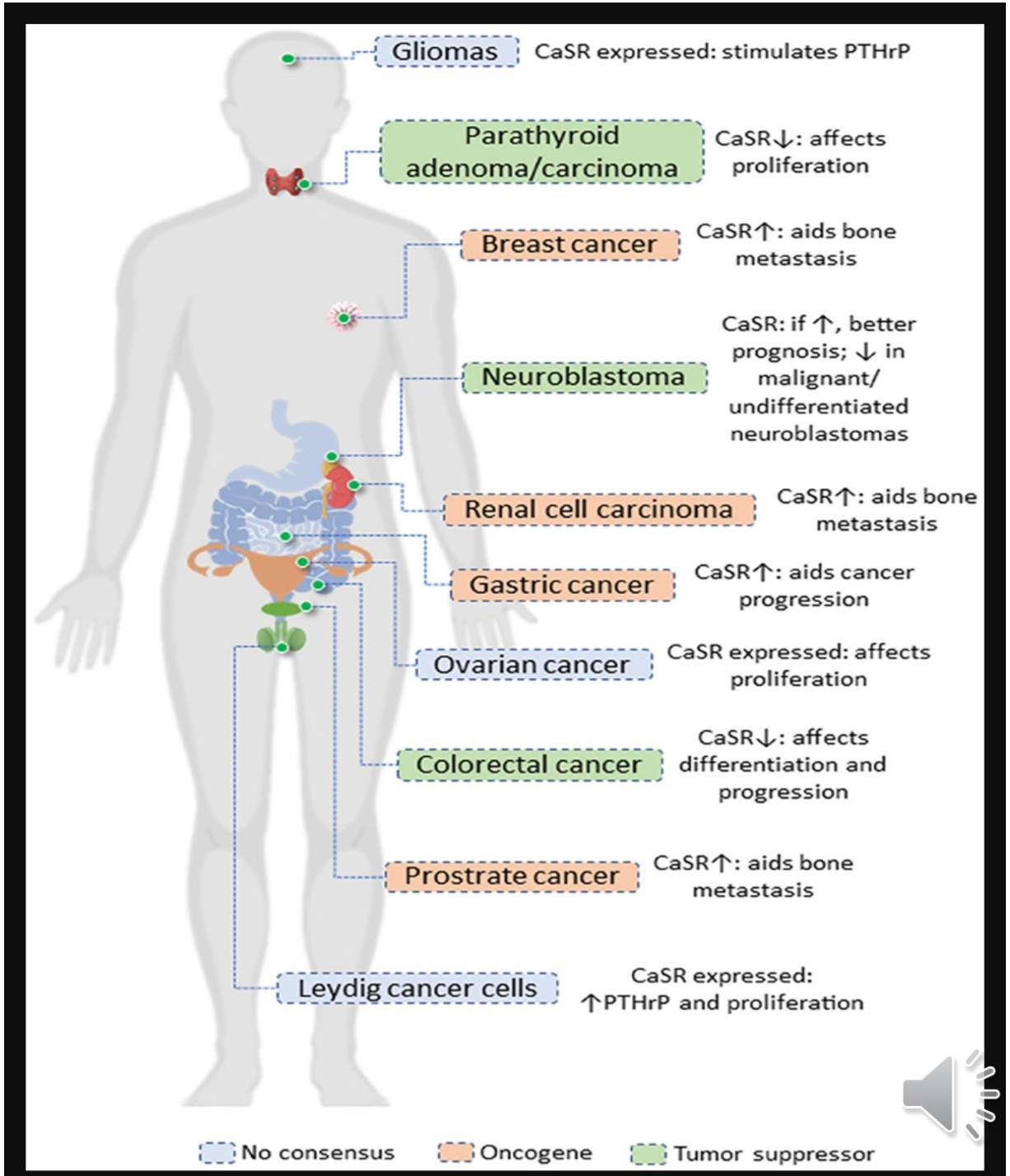
Additional article information

Abstract

Calcium is a crucial second messenger in the cardiovascular system. However, calcium may also be an extracellular first messenger through a G-protein-coupled receptor that senses extracellular concentration (Ca^{2+}_o), the calcium-sensing receptor (CaR). The most prominent physiological function of the CaR is to maintain the extracellular Ca^{2+} level in a very tight range by regulating the circulating levels of parathyroid hormone (PTH). This control over PTH and Ca^{2+} levels is partially lost in patients suffering from primary


Calcimimetics
Parathyroid CaR
Vascular CaR
↗ CaR
↙ PTH
↙ Calcium
↙ Phosphate
↗ CaR
↗ MGP
↙ Run2/Cbfa1
↙ BMP2
Prevent cardiovascular calcifications

CaSR and cancers



summary

- A pivotal role for the renal calcium-sensing receptor (CaSR) in the control of divalent cation excretion in a parathyroid hormone-independent manner has been identified through tissue-specific CaSR ablation and pharmacological studies

- A functional interaction between the CaSR and Claudin-14 in the thick ascending limb permits regulation of paracellular Ca^{2+} reabsorption

- The CaSR fine tunes Ca^{2+}, Mg^{2+}, and Pi transport in the proximal tubule by integrating multiple inputs from divalent cation concentration, osmolarity, and urine acidification

- Calcimimetics would be expected to increase urinary Ca^{2+} excretion by acting on the CaSR in the parathyroid glands and the kidney

- Calcilytics represent a novel, promising avenue for the treatment of hypercalciuria, nephrolithiasis, and nephrocalcinosis