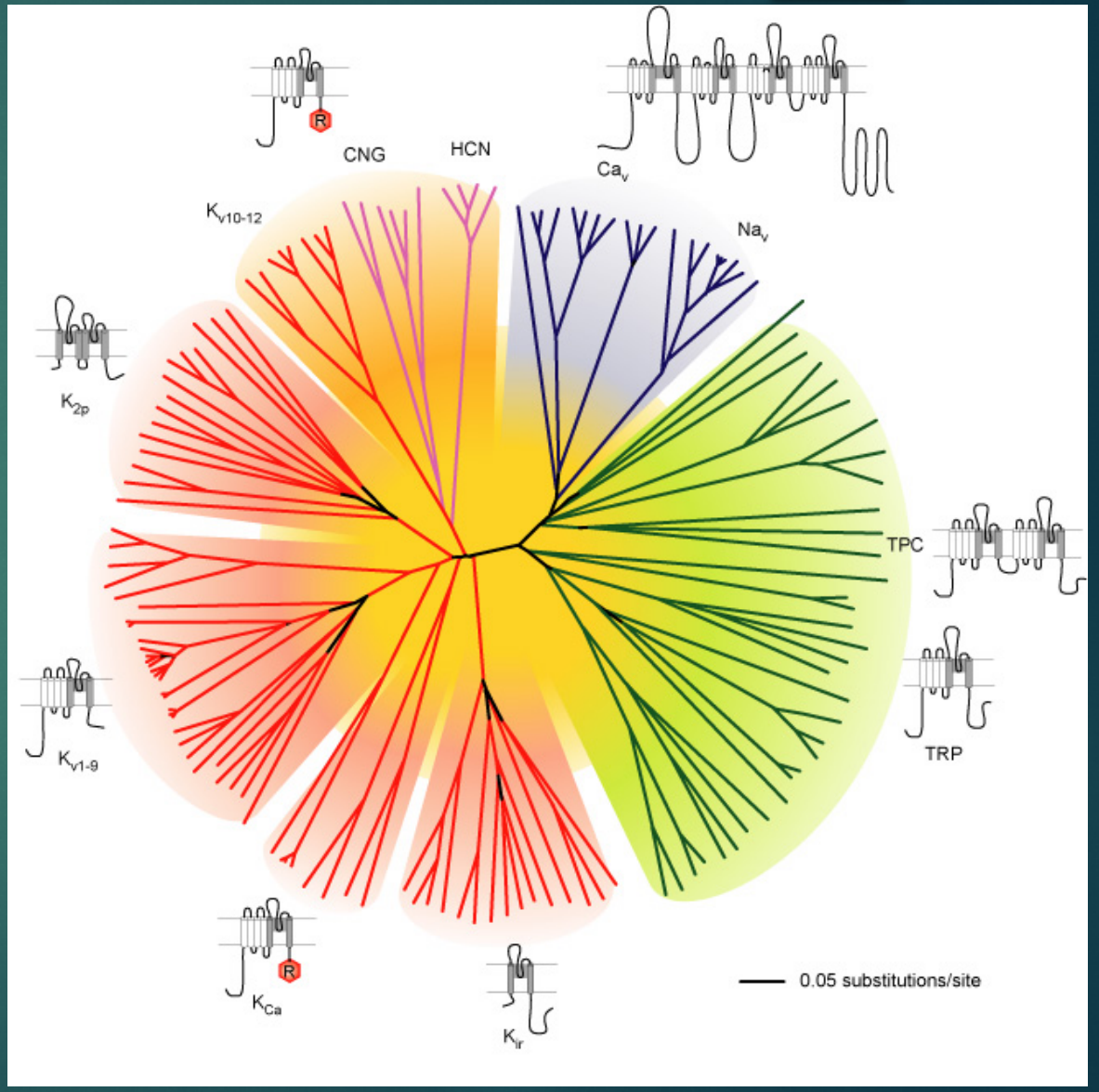
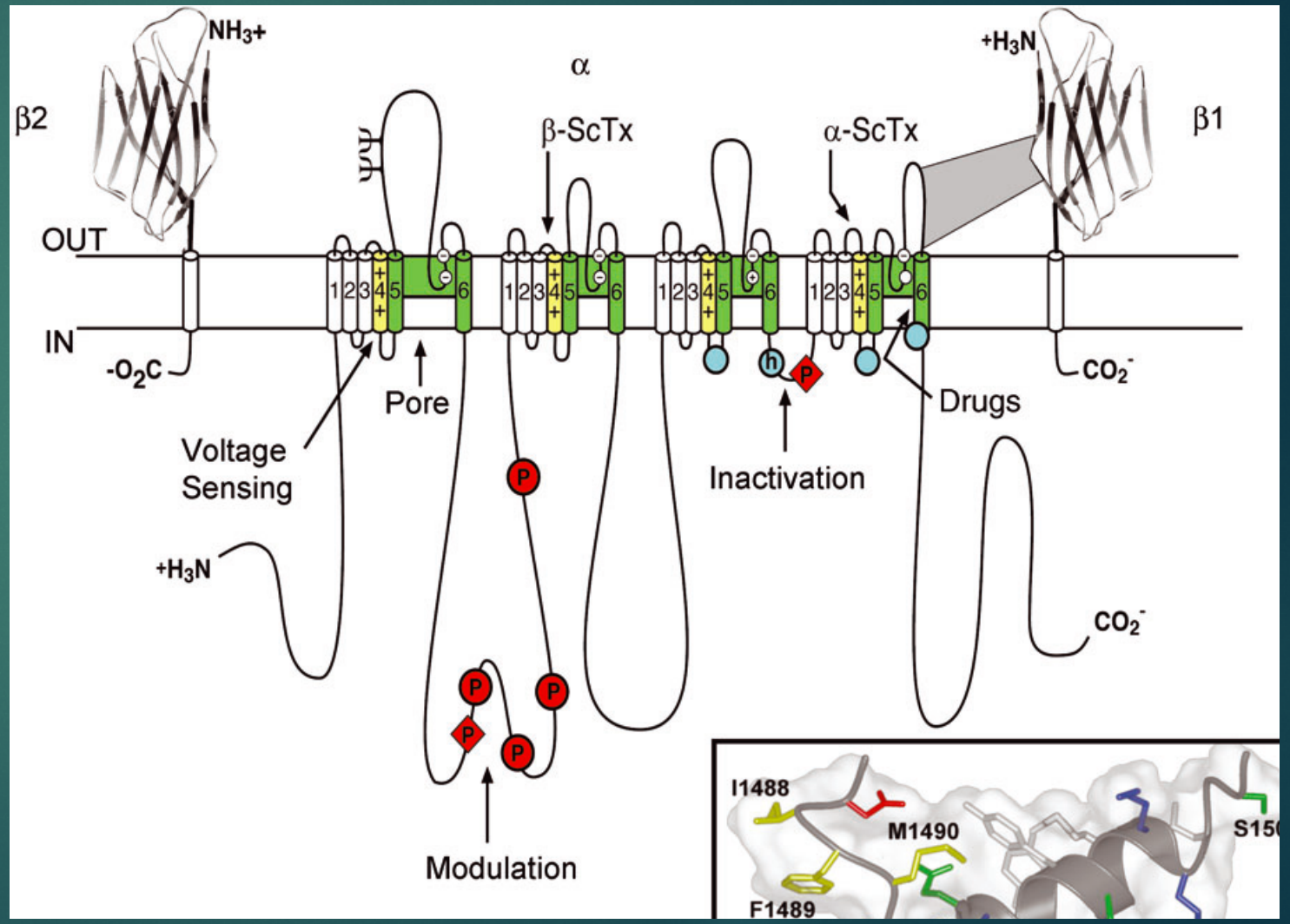
Ion Channels

STRUCTURE AND FUNCTION



Voltage gated Na+ channels

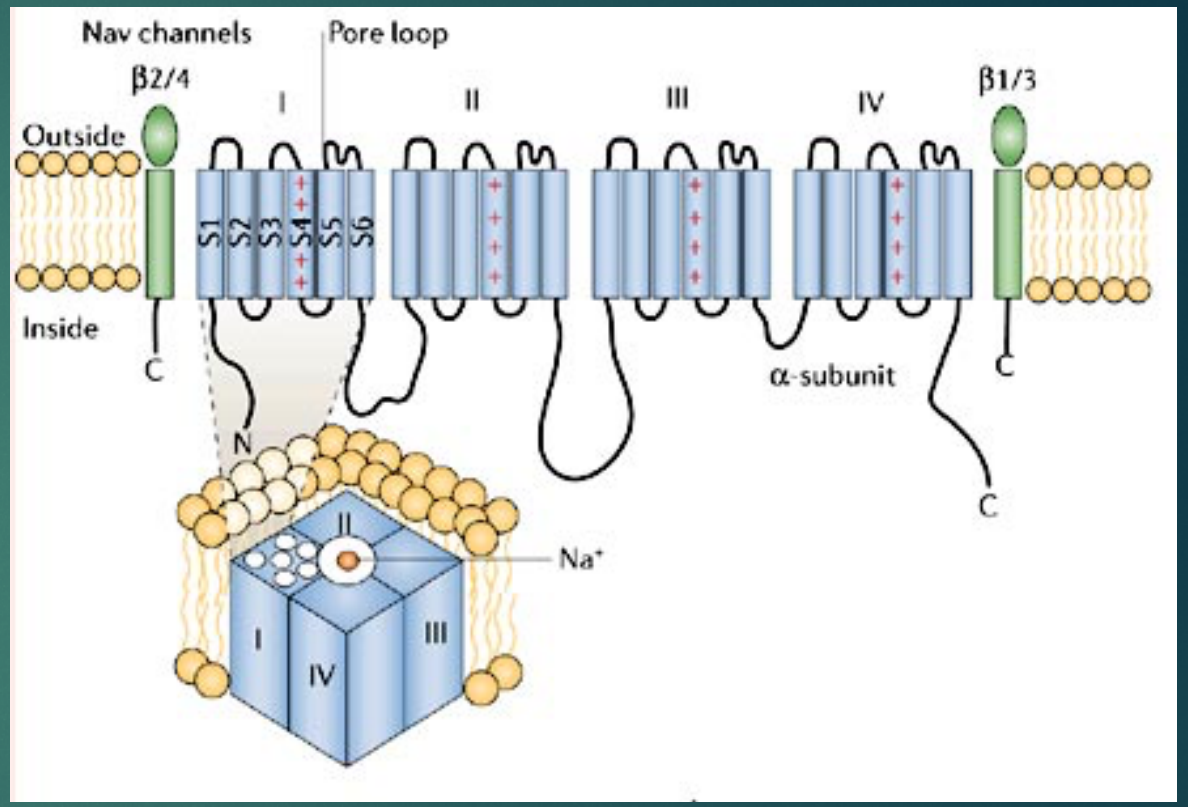
Sodium channel α subunits are composed of approximately 2000 amino acid residues organized in four homologous domains, which each contains six transmembrane segments. Later biochemical analyses and cDNA cloning showed that sodium channel β subunits are composed of an N-terminal extracellular immunoglobulin-like fold, a single transmembrane segment, and a short intracellular segment.



Catterall W, J Physiol 2012

Voltage gated Na+ channels

These subunits are thought to form heterodimeric and heterotrimeric complexes composed of a single α subunit and one or two β subunits in excitable cell membranes, and co-expression of β subunits modulates the kinetics and voltage dependence of sodium channel activation and inactivation.



Catterall W, J Physiol 2012

Voltage gated Na+ channels

Sodium channel **α subunits are encoded by 10 genes**, which are expressed in different excitable tissues.

- NaV1.1, 1.2, 1.3 and 1.6 are the primary sodium channels in the central nervous system.
- NaV1.7, 1.8 and 1.9 are the primary sodium channels in the peripheral nervous system.
- NaV1.4 is the primary sodium channel in skeletal muscle, whereas
- NaV1.5 is primary in heart.

Most of these sodium channels also have significant levels of expression outside of their primary tissues.

- The 10th sodium channel protein is not voltage-gated and is involved in salt sensing.

Table 1. Mammalian sodium channel α subunits

Type	Gene symbol	Chromosomal location	Primary tissues
$Na_v1.1$	*SCN1A*	Mouse 2 Human 2q24	CNS neurons
$Na_v1.2$	*SCN2A*	Mouse 2 Human 2q23–24	CNS neurons
$Na_v1.3$	*SCN3A*	Mouse 2 Human 2q24	CNS neurons
$Na_v1.4$	*SCN4A*	Mouse 11 Human 17q23–25	SkM
$Na_v1.5$	*SCN5A*	Mouse 9 Human 3p21	Uninnervated SkM, heart
$Na_v1.6$	*SCN8A*	Mouse 15 Human 12q13	CNS neurons
$Na_v1.7$	*SCN9A*	Mouse 2 Human 2q24	PNS neurons
$Na_v1.8$	*SCN10A*	Mouse 9 Human 3p22–24	DRG neurons
$Na_v1.9$	*SCN11A*	Mouse 9 Human 3p21–24	DRG neurons
Na_x	*SCN7A* *SCN6A*	Mouse 2 Human 2q21–23	uterus, astrocytes, hypothalamus

Catterall W, J Physiol 2012

Voltage gated Na+ channels

β subunits have been identified by genomic analyses and cDNA cloning to give a small family of four NaVβ subunits in total.

β1 and β3 are associated non-covalently with α subunits and resemble each other most closely in amino acid sequence, whereas β2 and β4 form disulfide bonds with α subunits and also resemble each other closely.

The structure of Navβ subunits resembles the family of cell adhesion molecules, and increasing evidence supports their role in localization and immobilization of sodium channels in specific locations in excitable cells.

Voltage gated Ca+ channels

Ca2+ entering the cell through voltagegated Ca2+ channels serves as the second messenger of electrical signaling, initiating many different cellular events:

- In cardiac and smooth muscle cells, activation of Ca2+ channels initiates contraction directly by increasing cytosolic Ca2+ concentration and indirectly by activating calcium-dependent calcium release by ryanodine-sensitive Ca2+ release channels in the sarcoplasmic reticulum.
- In skeletal muscle cells, voltage-gated Ca2+ channels in the transverse tubule membranes interact directly with ryanodine-sensitive Ca2+ release channels in the sarcoplasmic reticulum and activate them to initiate rapid contraction.

Voltage gated Ca+ channels

- In endocrine cells, voltage-gated Ca2+ channels mediate Ca2+ entry that initiates secretion of hormones.
- In neurons, voltage-gated Ca2+ channels initiate synaptic transmission.
- In many different cell types, Ca2+ entering the cytosol via voltage-gated Ca2+ channels regulates enzyme activity, gene expression, and other biochemical processes

Voltage gated Ca+ channels

Table 1. Subunit composition and function of Ca^{2+} channel types

Ca^{2+} current type	α1 Subunits	Specific blocker	Principal physiological functions	Inherited diseases
L	$Ca_v1.1$	DHPs	Excitation-contraction coupling in skeletal muscle, regulation of transcription	Hypokalemic periodic paralysis
	$Ca_v1.2$	DHPs	Excitation-contraction coupling in cardiac and smooth muscle, endocrine secretion, neuronal Ca^{2+} transients in cell bodies and dendrites, regulation of enzyme activity, regulation of transcription	Timothy syndrome: cardiac arrhythmia with developmental abnormalites and autism spectrum disorders
	$Ca_v1.3$	DHPs	Endocrine secretion, cardiac pacemaking, neuronal Ca^{2+} transients in cell bodies and dendrites, auditory transduction	
	$Ca_v1.4$	DHPs	Visual transduction	Stationary night blindness
N	$Ca_v2.1$	ω-CTx-GVIA	Neurotransmitter release, Dendritic Ca^{2+} transients	
P/Q	$Ca_v2.2$	ω-Agatoxin	Neurotransmitter release, Dendritic Ca^{2+} transients	Familial hemiplegic migraine, cerebellar ataxia
R	$Ca_v2.3$	SNX-482	Neurotransmitter release, Dendritic Ca^{2+} transients	
T	$Ca_v3.1$	None	Pacemaking and repetitive firing	
	$Ca_v3.2$		Pacemaking and repetitive firing	Absence seizures
	$Ca_v3.3$			

Abbreviations: DHP, dihydropyridine; ω-CTx-GVIA, ω-conotoxin GVIA from the cone snail *Conus geographus*; SNX-482, a synthetic version of a peptide toxin from the tarantula *Hysterocrates gigas*.

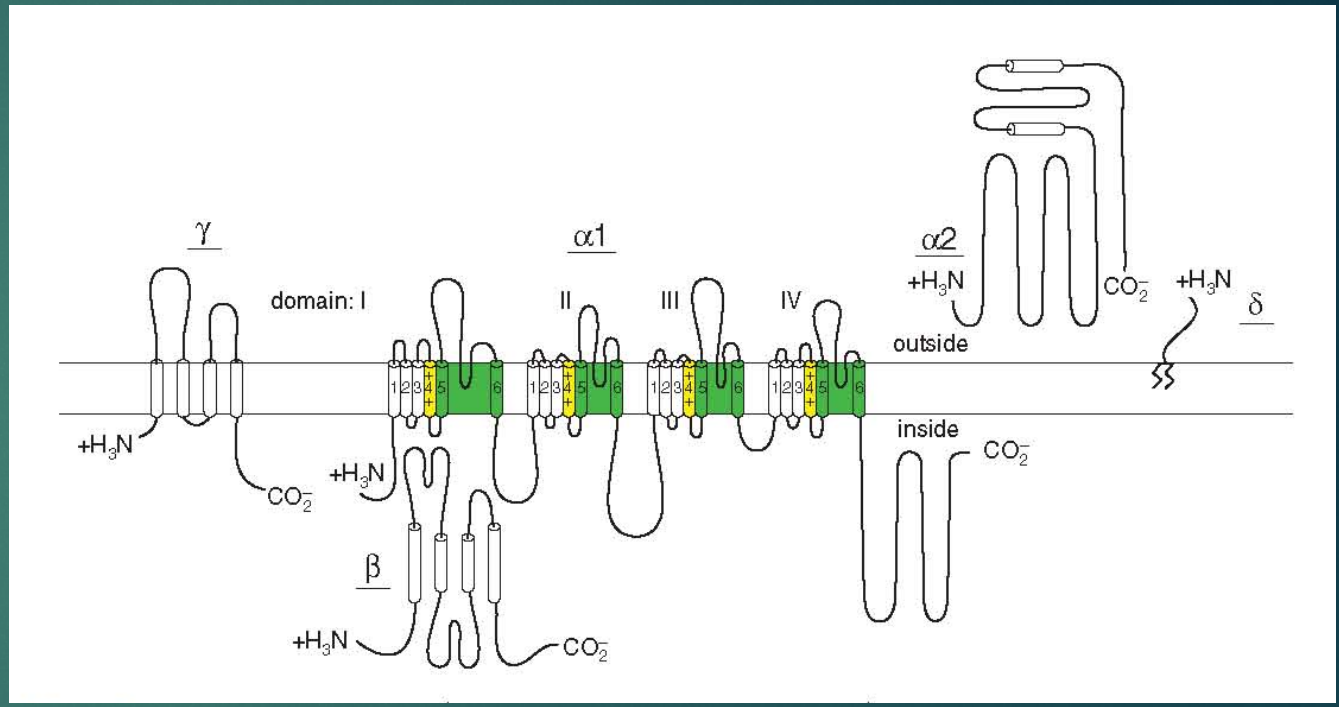
Catterall W, CSH persp, 2011

Voltage gated Ca+ channels

Ca2+ channels purified from skeletal muscle transverse tubules are complexes of α1, α2, β, γ, and δ subunits.

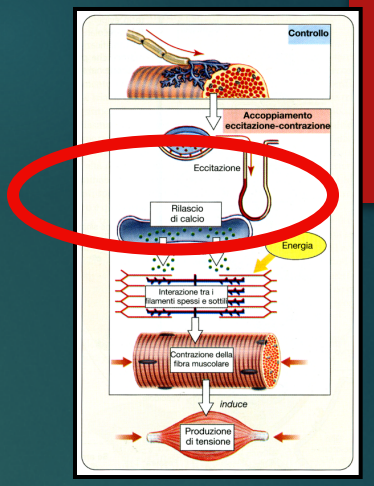
The principal transmembrane α1 subunit of 190 kDa in association with a disulfide-linked α2δ dimer of 170 kDa, an intracellular phosphorylated β subunit of 55 kDa, and a transmembrane γ subunit of 33 kDa.

The α1 subunit is a protein of about 2000 amino acid residues in length with an amino acid sequence and predicted **transmembrane structure like the previously characterized, pore-forming a subunit of voltage-gated sodium channels**

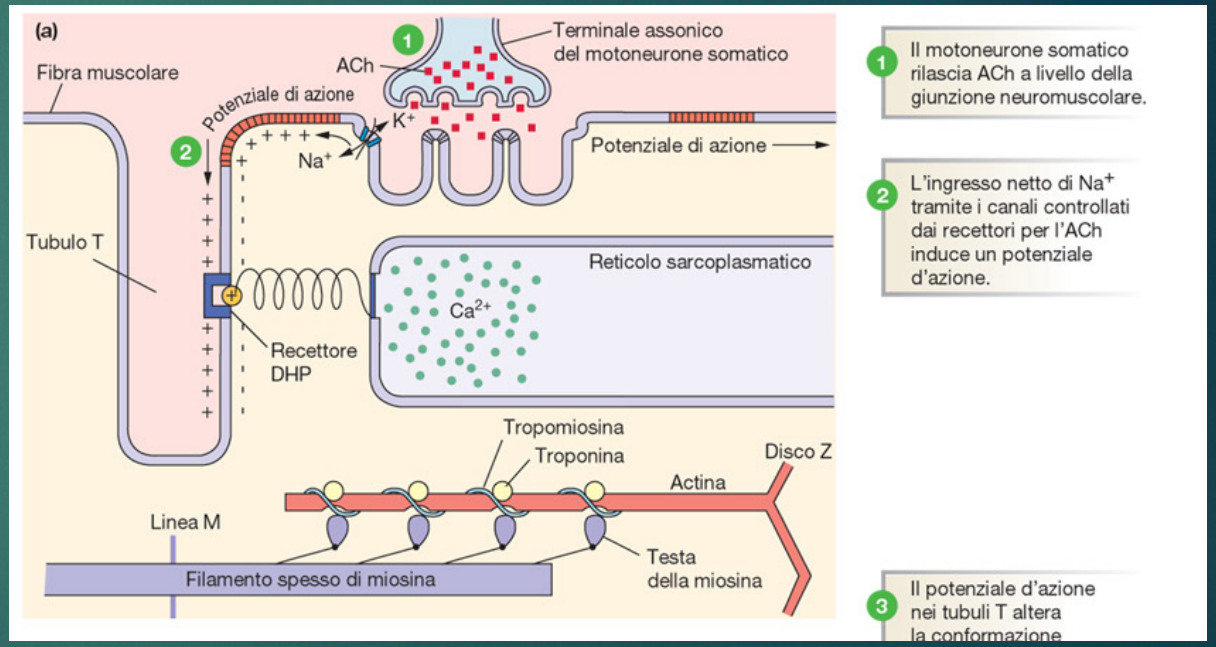


Catterall W, CSH persp, 2011

Excitation-contraction coupling in skeletal muscle

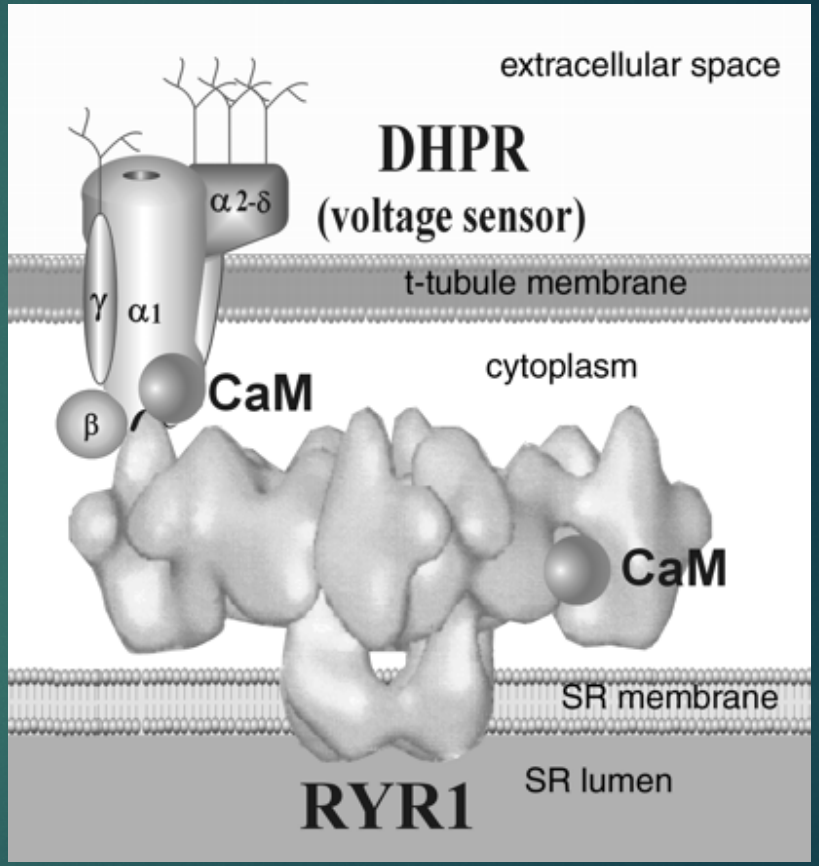


CaV1.1 channels in the transverse tubules are thought to interact directly with the ryanodine-sensitive Ca2+ release channels (RyR1) of the sarcoplasmic reticulum, as observed in high-resolution electron microscopy, and the voltage-driven conformational changes in their voltage-sensing domains are thought to directly induce activation of RyR1.



In skeletal muscle, entry of external Ca2+ is not required for initiation of contraction

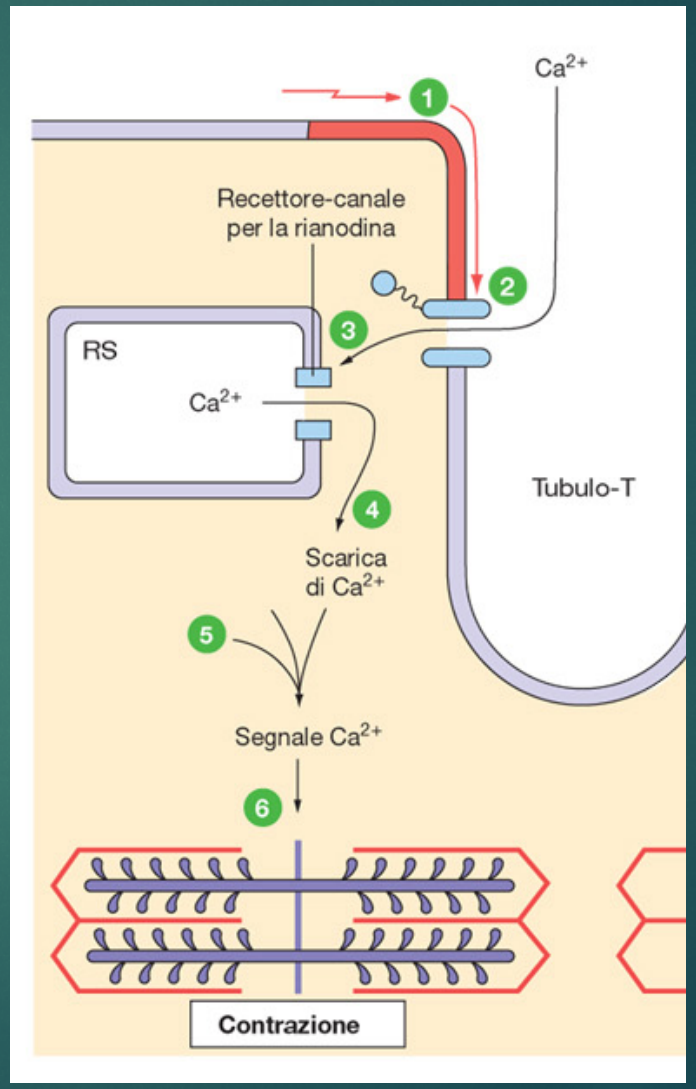
This is because the cytoplasmic domain of these channels is physically coupled to ryanodine receptor (RyR1) Ca2+ release channels on internal membranes. Even though Cav1.1 proteins can act as *bona fide Ca2+ channels, they also function as* voltage sensors that directly produce conformational changes in the ryanodine receptor/Cav1.1 complex, resulting in the release of Ca2+ from internal stores.



Excitation-contraction coupling in cardiac muscle

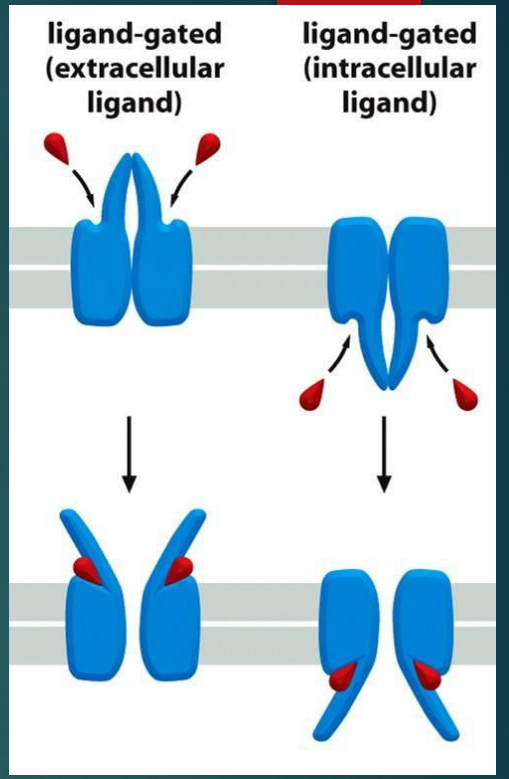
In contrast to skeletal muscle, entry of **Ca2+ is required for excitation-contraction coupling in cardiac myocytes**, and Ca2+ entry via CaV1.2 channels triggers activation of the RyR2 and initiates Ca2+-induced Ca2+-release, activation of actomyosin, and contraction

Release of Ca2+ from the sarcoplasmic reticulum via RyR2 greatly amplifies the cellular Ca2+ transient and is required for effective initiation of contraction. All three steps in the cascade of Ca2+ transport processes—**Ca2+ entry via CaV1.2 channels**, **Ca2+ release via RyR, and Ca2+ uptake into the sarcoplasmic reticulum by SERCA Ca2+ pumps**—are tightly regulated by second messenger signaling networks



1. Un potenziale d'azione invade la membrana cellulare provenendo da una cellula adiacente.
2. I canali voltaggio-dipendenti per il Ca^{2+} si aprono. Il Ca^{2+} entra nella cellula.
3. L'ingresso di Ca^{2+} innesca il rilascio di altro Ca^{2+} dal reticolo sarcoplasmatico attraverso i recettori-canali della rianodina (RyR).
4. Il rilascio localizzato di calcio provoca la «scarica» di Ca^{2+}.
5. Le scariche di Ca^{2+} si sommano per produrre un segnale di Ca^{2+}.
6. Gli ioni calcio si legano alla troponina e inizia la contrazione.

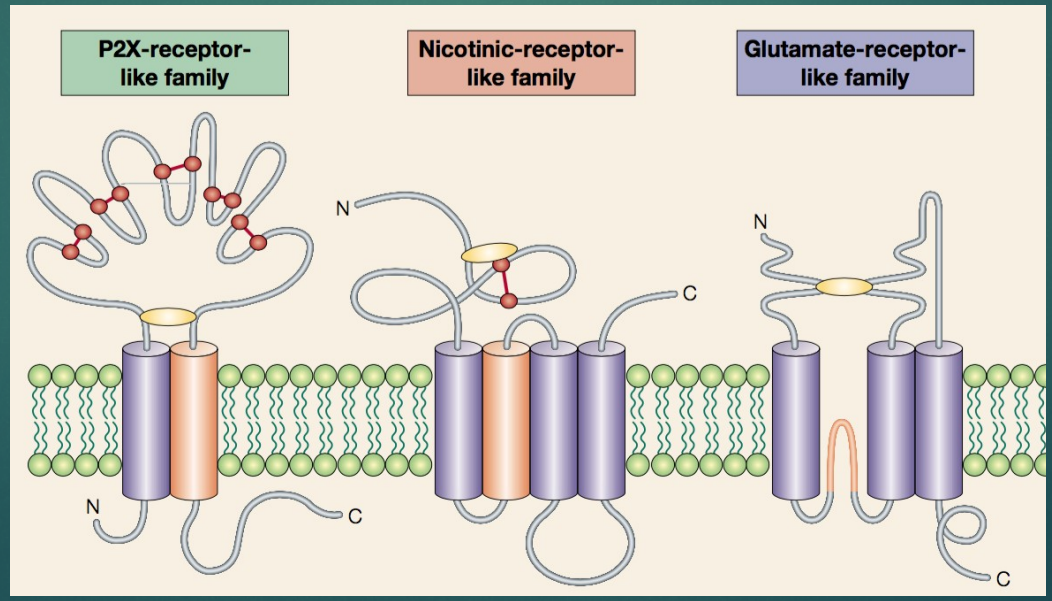
Ligand gated ion channels



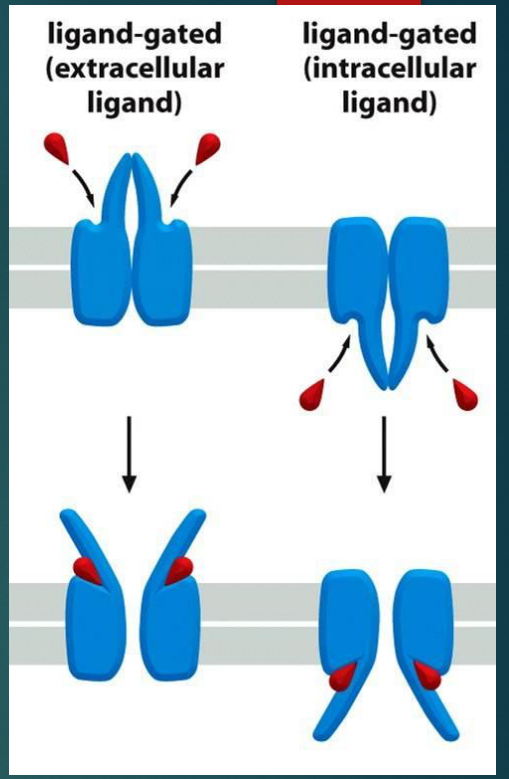
This is a highly heterogenous family of channels tha includes several families

- the **extracellular ligand-activated channels** which includes channels such as **GABA and glycine receptor channels**, most of which are regulated by ligands that are "neurotransmitters". These channels are often named according to the ligand they bind to. Other examples are: **nicotinic receptors ; P2X receptors**

We will discuss different examples in the course



Ligand gated ion channels

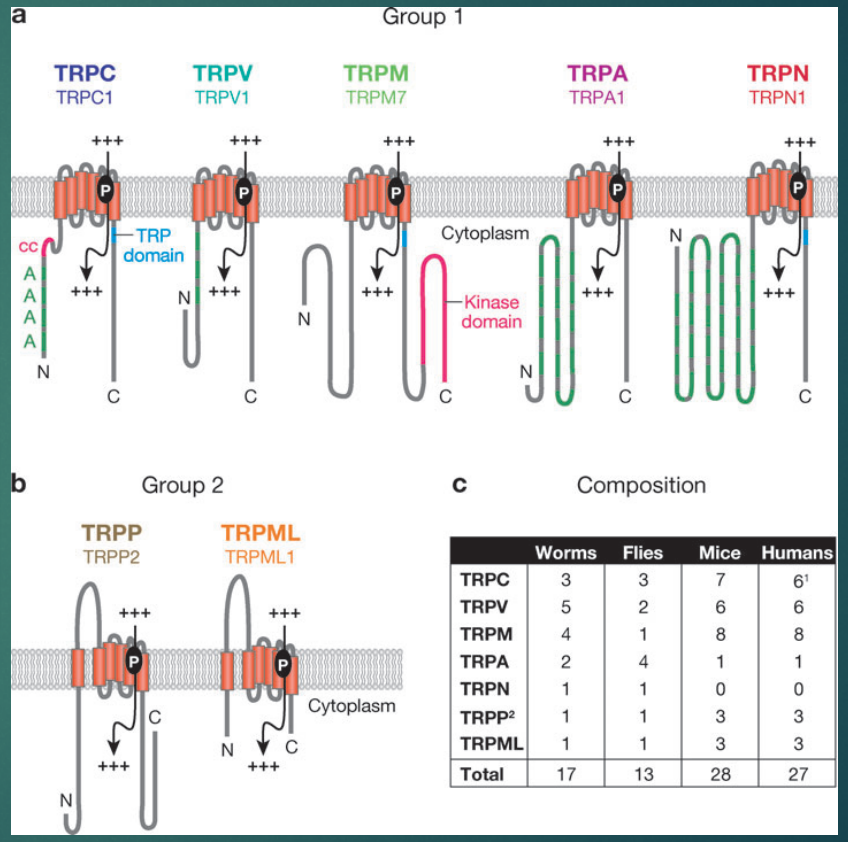


This is a highly heterogenous family of channels tha includes several families

- **Intracellular ligand-gated ion channels**. These include **CFTR** and some other ABC family members as well as ion channels involved in sense perception; **TRP channels; CNGC**; **These are often activated indirectly by GCPRs**. Other common intracellular ligands which activate these kinds of channels include calcium ions, **ATP**, **cyclic AMP and GMP as well as phosphadidyl inositol (PI)**. There are additional systems of nomenclature which have joined the second and third groups into the "chemically activated" or just simply "ligand gated" ion channels.

TRP family of channels

- Cation channels
- Non voltage-dependent
- Diversity in activation mechanisms
- Implication in diverse physiological functions

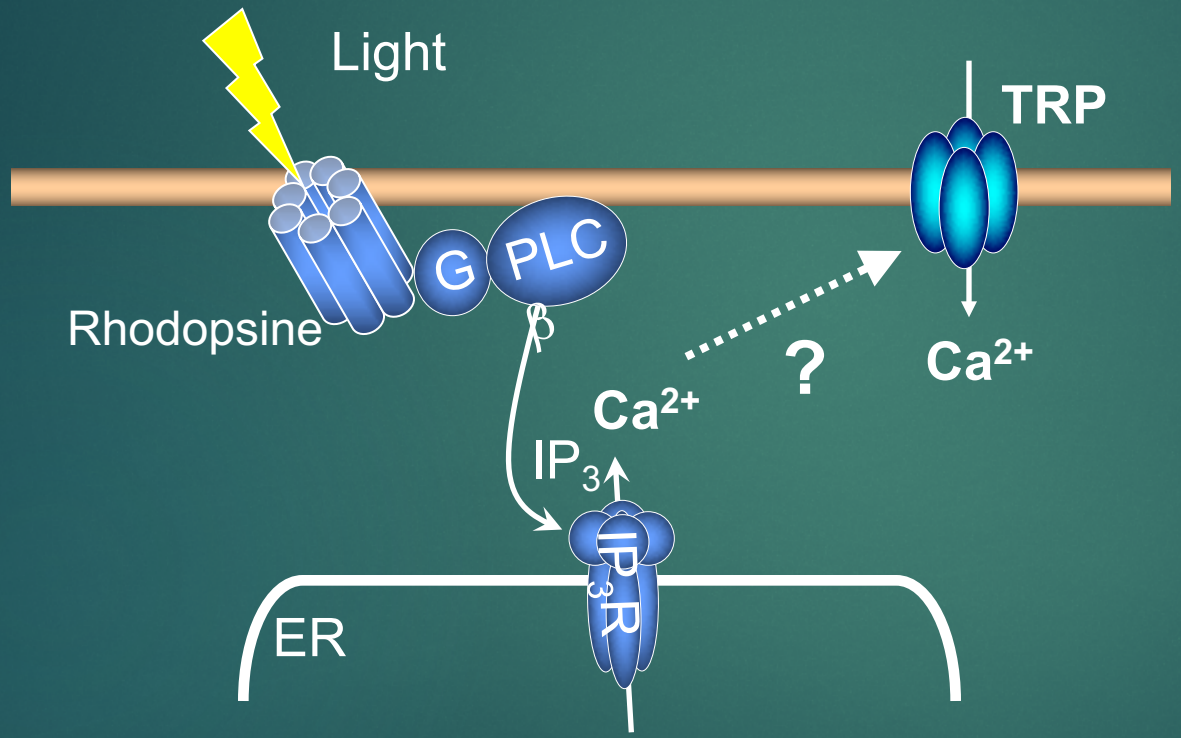


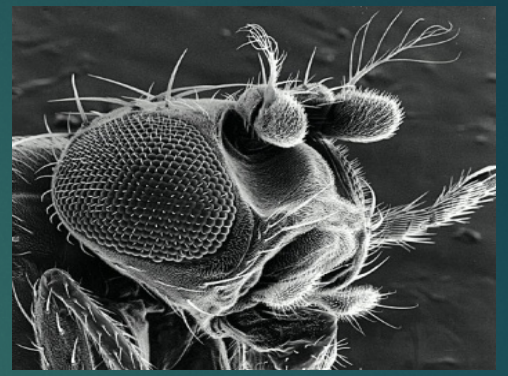
	Worms	Flies	Mice	Humans
TRPC	3	3	7	6[1]
TRPV	5	2	6	6
TRPM	4	1	8	8
TRPA	2	4	1	1
TRPN	1	1	0	0
TRPP[2]	1	1	3	3
TRPML	1	1	3	3
Total	17	13	28	27

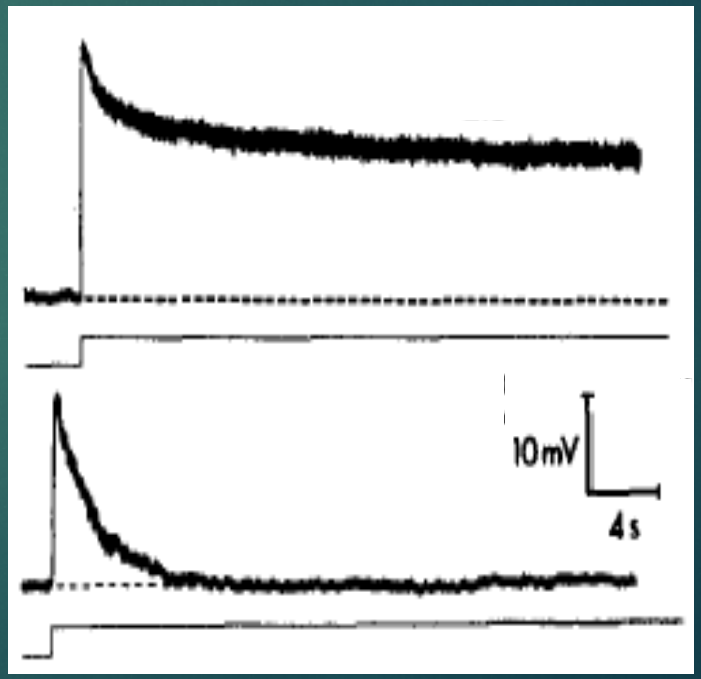
Venkatachalam K & Montell C, *Annual Review of Biochemistry*, 2007

First TRP channel identification

Drosophila melanogaster :

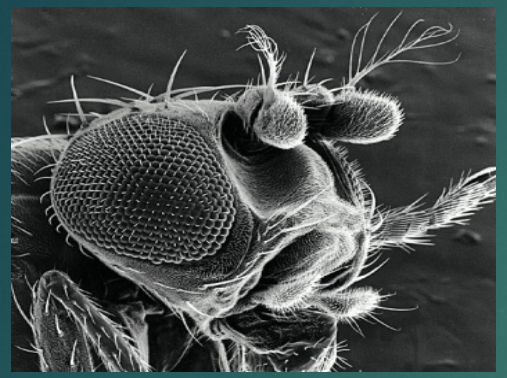


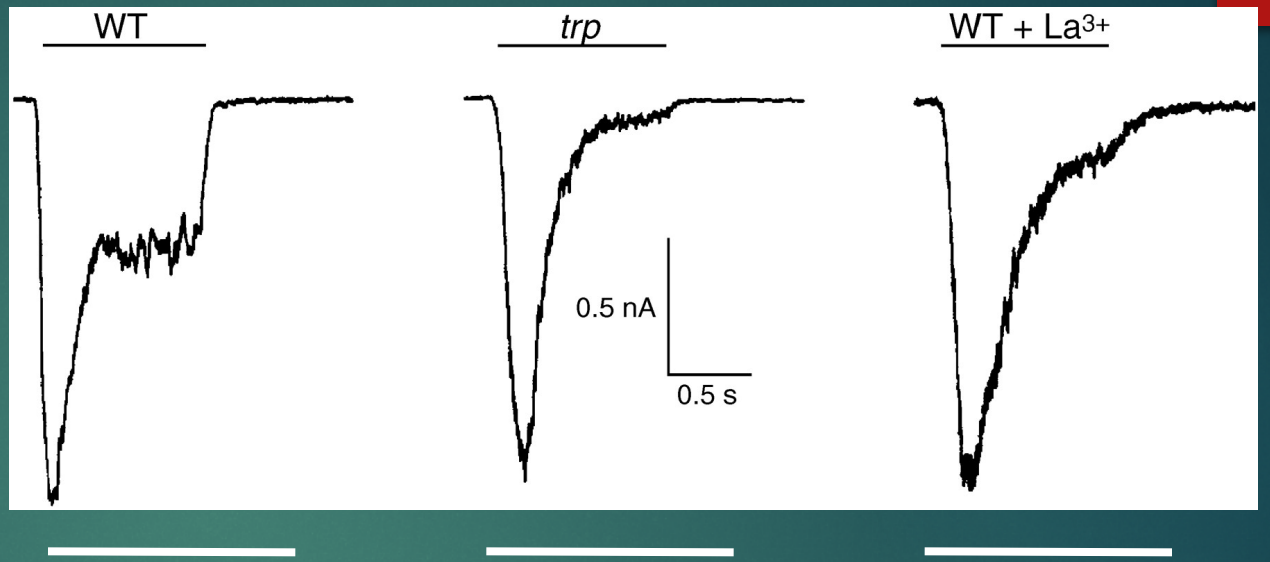




Minke B, Biophys Struct Mech., 1977

Identification du 1er Canal TRP





trp
Transient Receptor Potential → Cationic channel → Conserved from c.Elegans to human

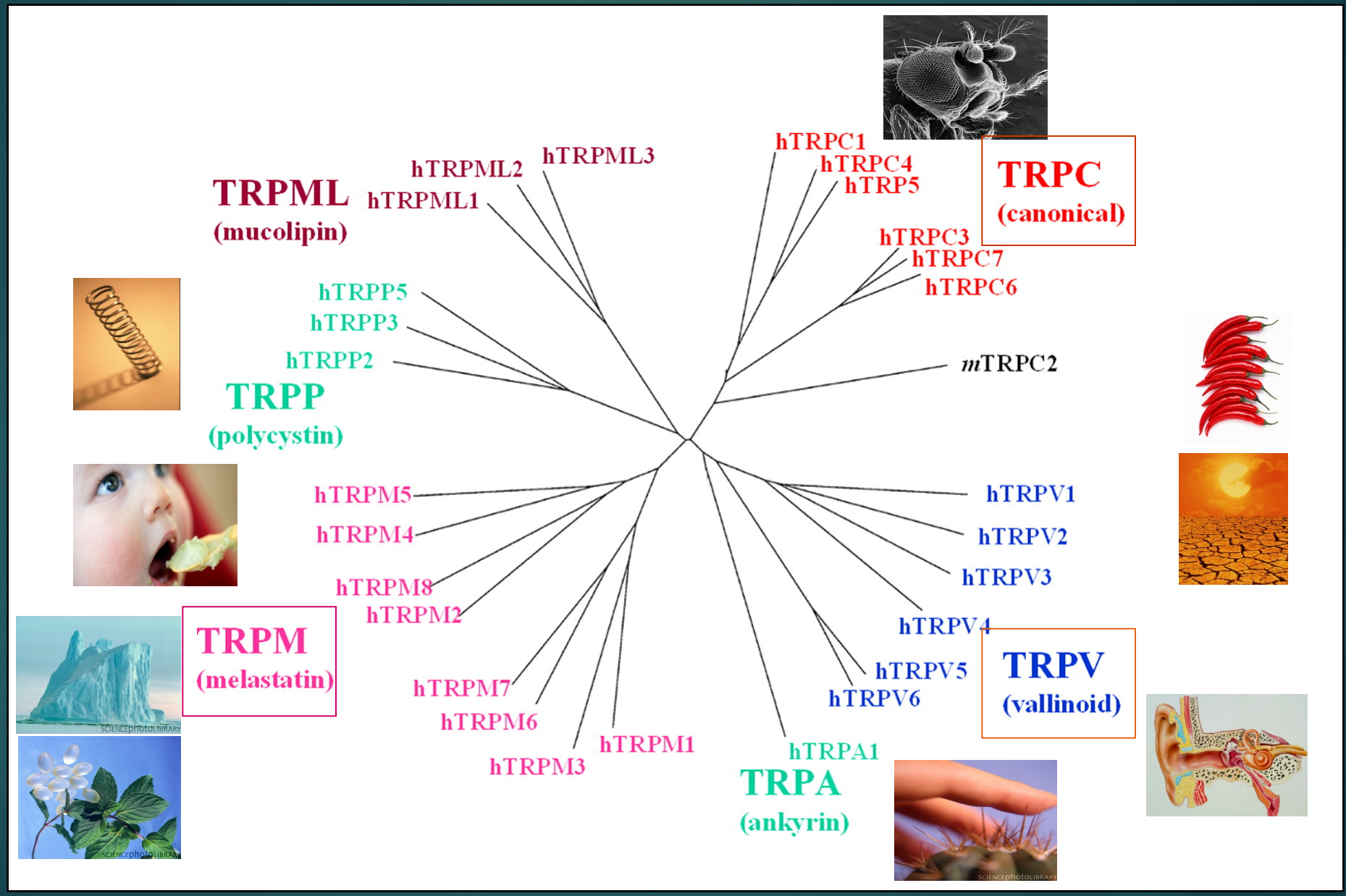
Montell C et al. *Neuron.* **2**, 1313-1323 (1989).

TRP family composition in worms, flies, mice and humans

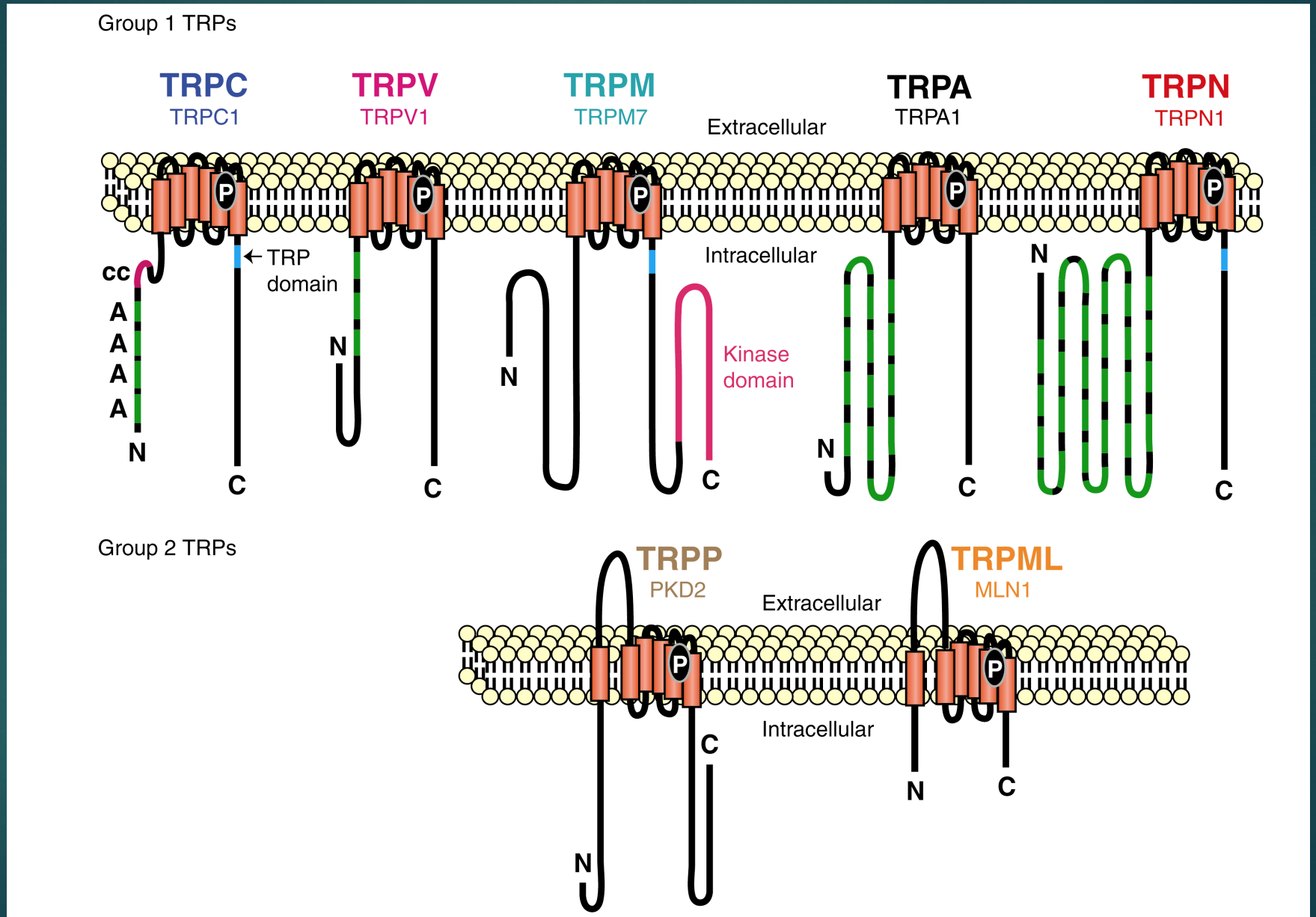
Subfamily	Worms	Flies	Mice	Humans
TRPC	3	3	7	6[1]
TRPV	5	2	6	6
TRPM	4	1	8	8
TRPA	2	4	1	1
TRPN	1	1	0	0
TRPP[2]	1	1[3]	3	3
TRPML	1	1	3	3
Total	17	13	28	27

C. Montell, Sci. STKE 2005, re3 (2005)

The TRP channels and 5 senses



Different topologies of TRP channels



The quartenary structure of TRP channels allows homo- or heteromeric configurations