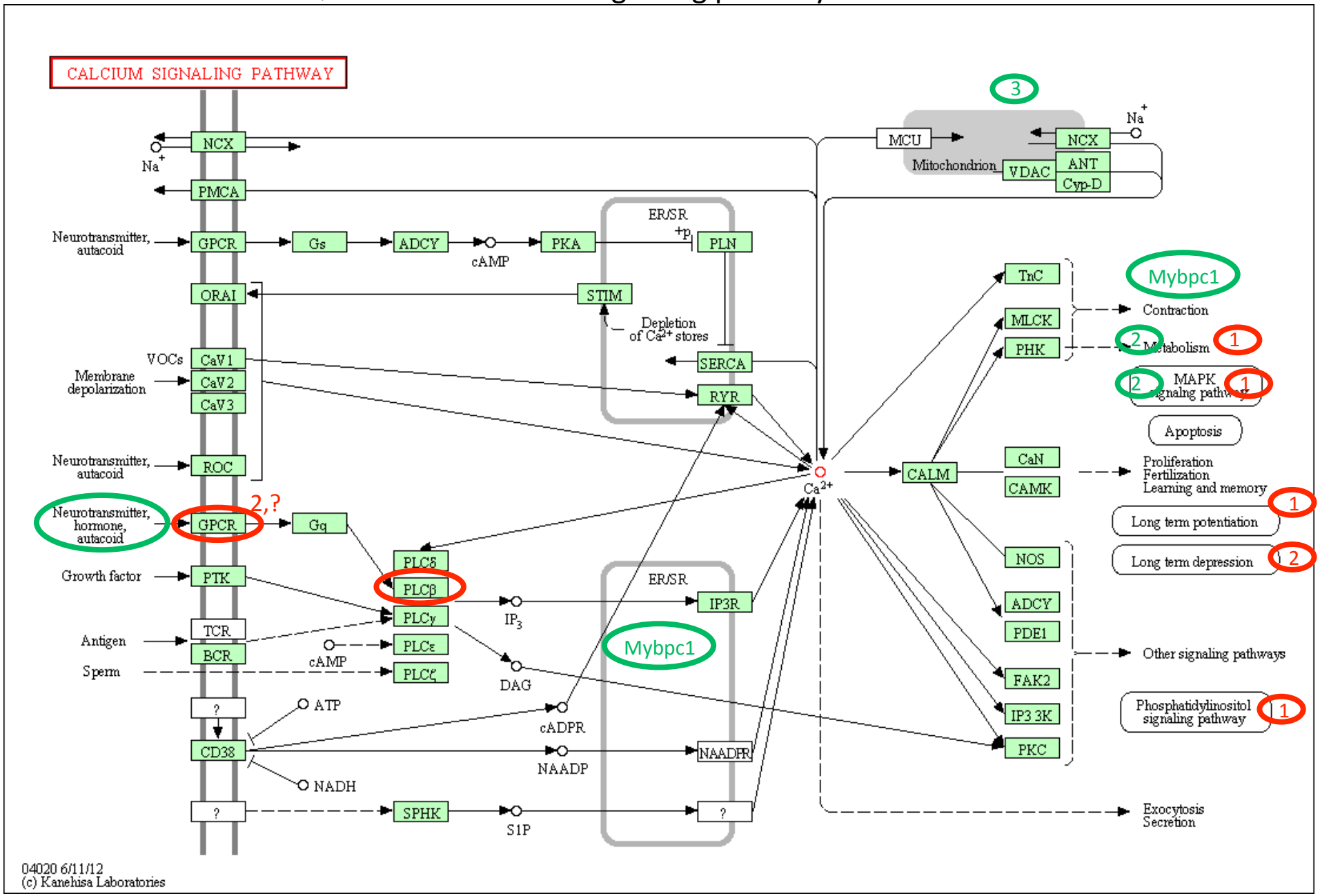
Lab meeting 8-17-12

- See spreadsheets

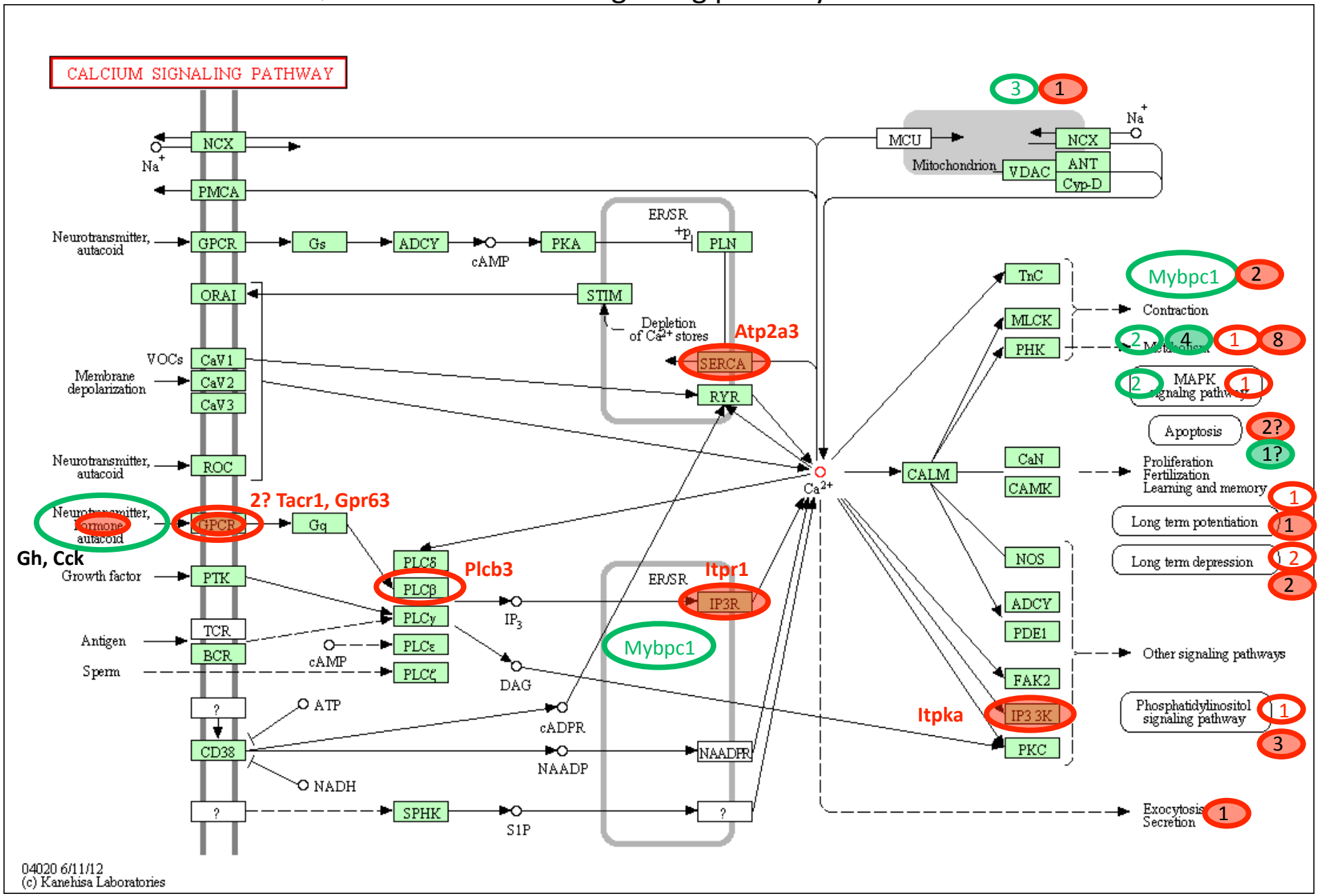
Comparison of RNA expression patterns in some well known pathways (Q127 vs WT)

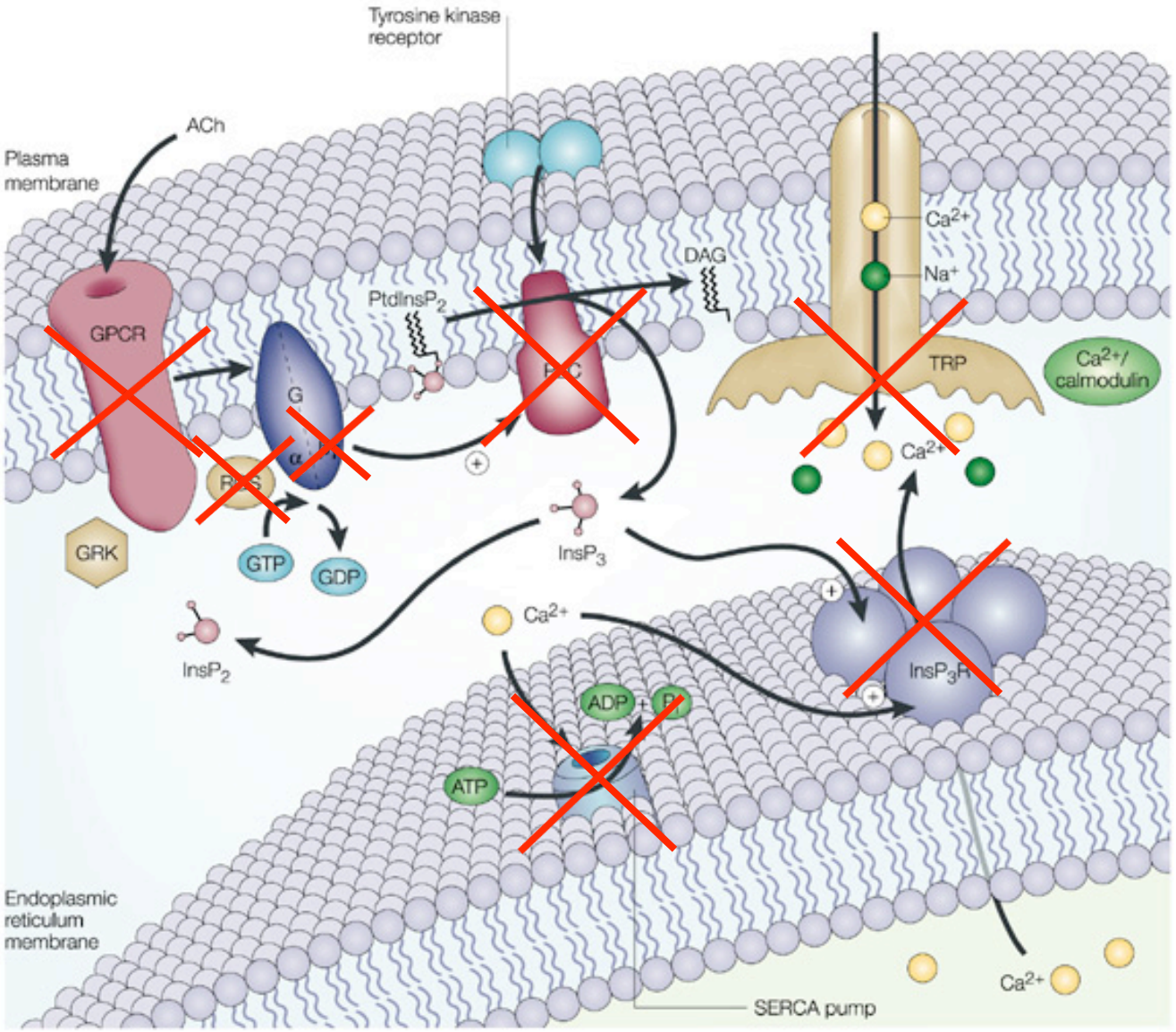
Key: A significant difference in expression is represented by a circle. Red signifies lower expression relative to WT, green signifies higher expression. Open circles represent 4 week-old comparisons, filled circles represent 8 week-old comparisons.

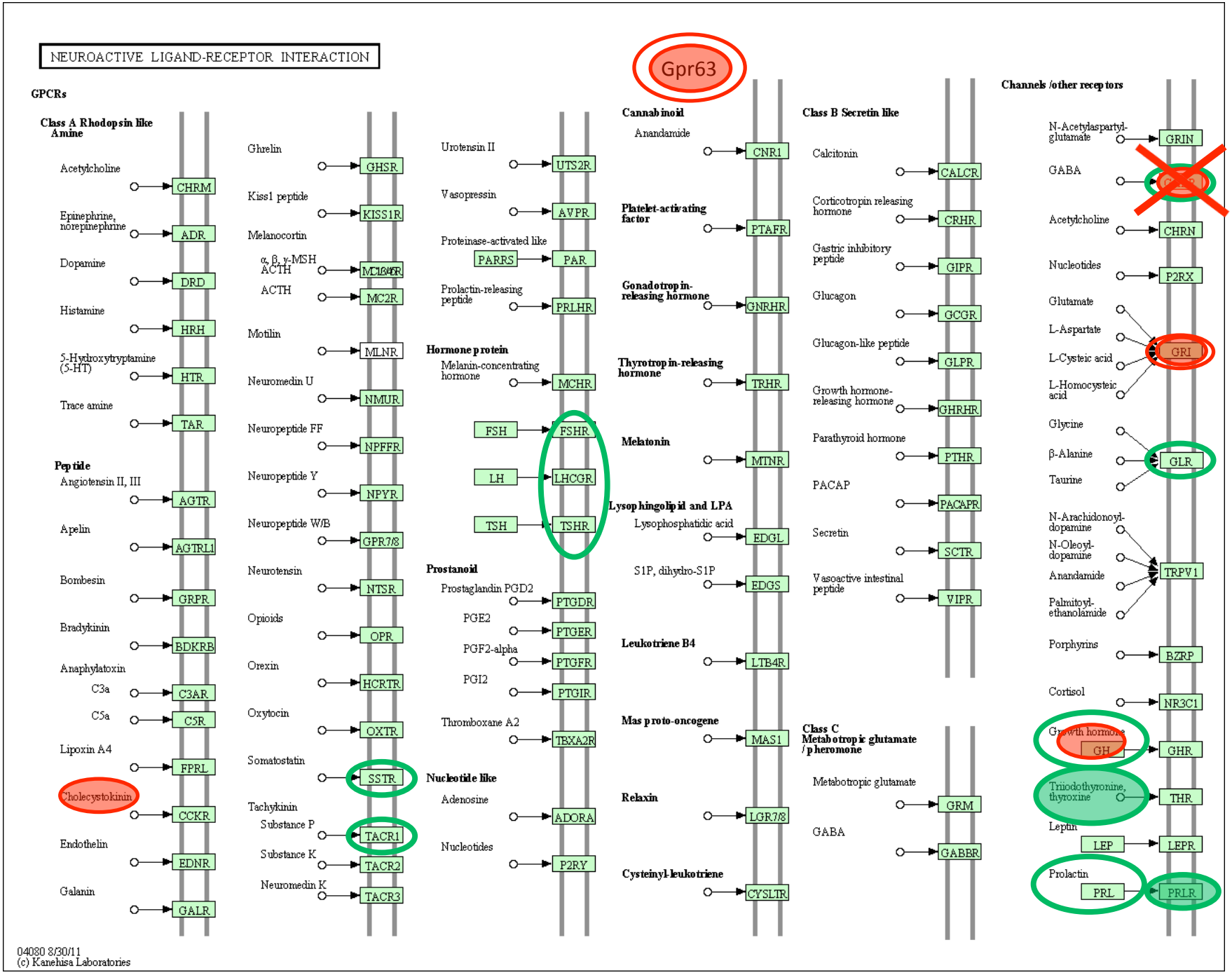
Q127 effects on Ca++ signaling pathway at 4 weeks

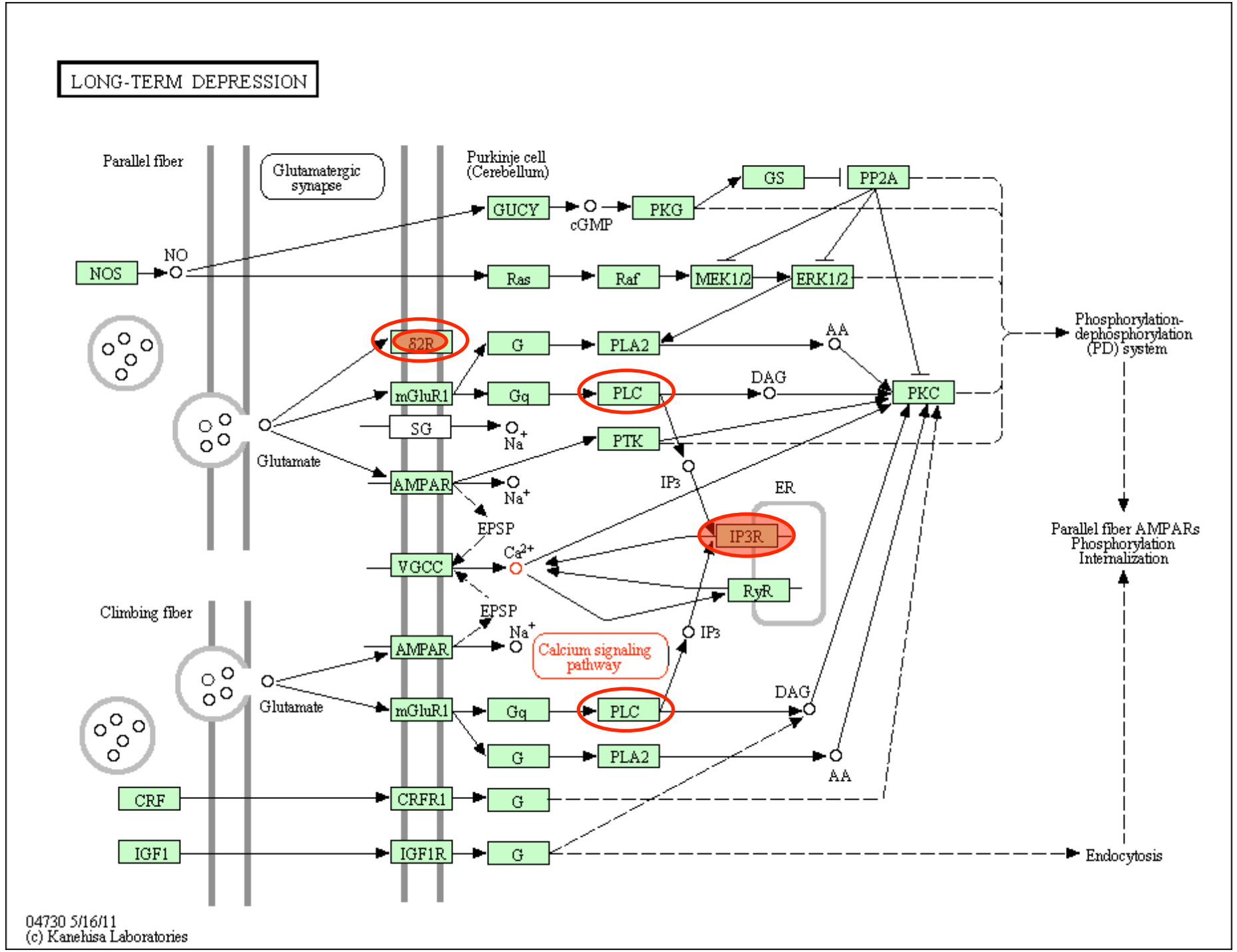


Q127 effects on Ca++ signaling pathway at 8 weeks




Tyrosine kinase receptor
ACh
Plasma membrane
GPCR
G
α
PtdInsP2
PLC
DAG
Ca2+
Na+
TRP
Ca2+/ calmodulin
Ca2+
GRK
GTP
GDP
InsP3
InsP2
Ca2+
InsP3R
ADP
ATP
Endoplasmic reticulum membrane
SERCA pump
Ca2+

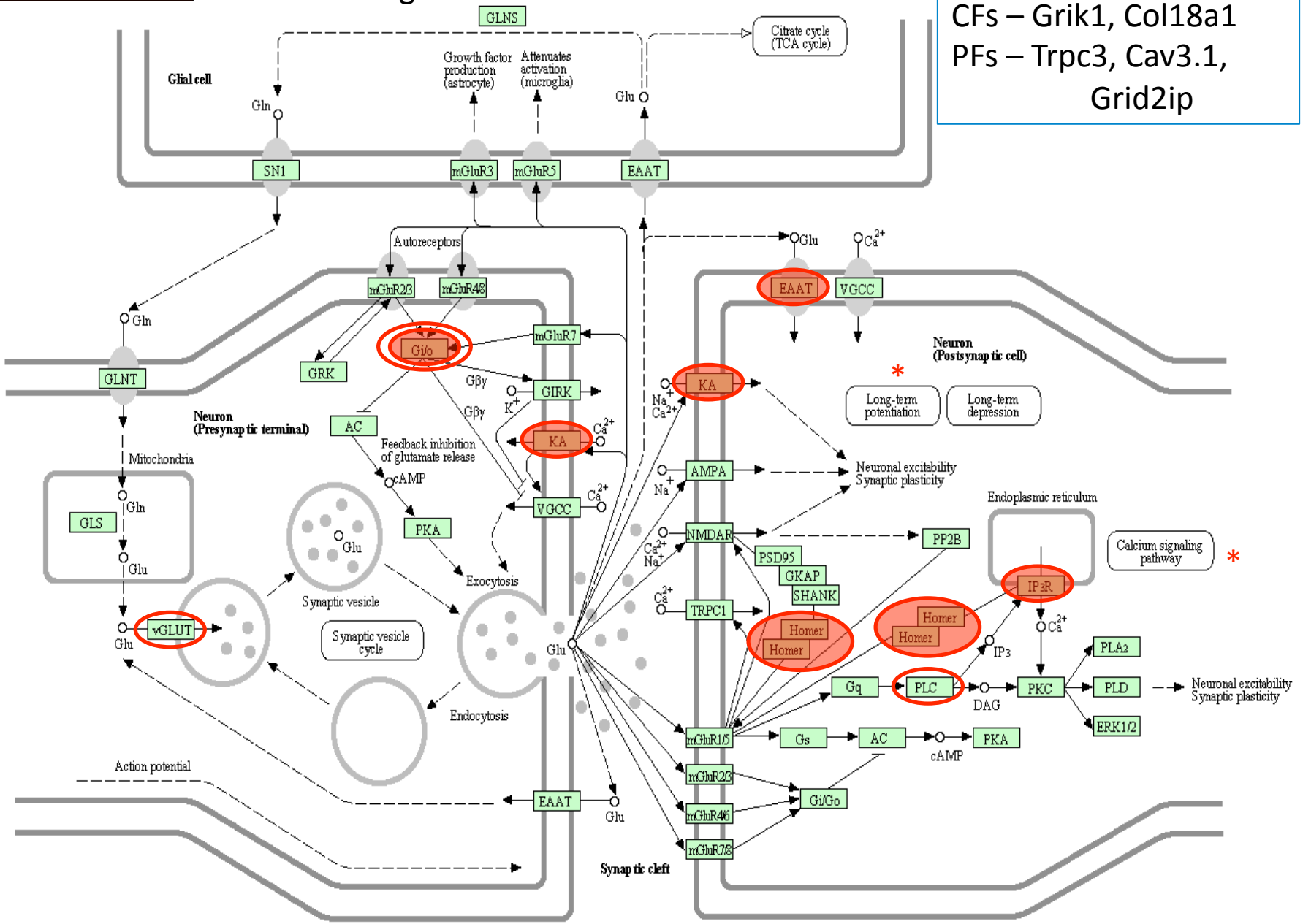

NEUROACTIVE LIGAND-RECEPTOR INTERACTION
Gpr63
GPCRs
Class A Rhodopsin like Amine
Acetylcholine
CHRM
Epinephrine, norepinephrine
ADR
Dopamine
DRD
Histamine
HRH
5-Hydroxytryptamine (5-HT)
HTR
Trace amine
TAR
Peptide
Angiotensin II, III
AGTR
Apelin
AGTRL1
Bombesin
GRPR
Bradykinin
BDKRB
Anaphylatoxin
C3a
C3AR
C5a
C5R
Lipoxin A4
FPRL
Cholecystokinin
CCKR
Endothelin
EDNR
Galanin
GALR
Ghrelin
GHSR
Kiss1 peptide
KISS1R
Melanocortin
α, β, γ-MSH ACTH
MC1/4R
ACTH
MC2R
Motilin
MLNR
Neuromedin U
NMUR
Neuropeptide FF
NPFFR
Neuropeptide Y
NPYR
Neuropeptide W/B
GPR7/8
Neurotensin
NTSR
Opioids
OPR
Orexin
HCRTR
Oxytocin
OXTR
Somatostatin
SSTR
Tachykinin
Substance P
TACR1
Substance K
TACR2
Neuromedin K
TACR3
Urotensin II
UTS2R
Vasopressin
AVPR
Proteinase-activated like
PARRS
PAR
Prolactin-releasing peptide
PRLHR
Hormone protein
Melanin-concentrating hormone
MCHR
FSH
FSHR
LH
LHCGR
TSH
TSHR
Prostanoid
Prostaglandin PGD2
PTGDR
PGE2
PTGER
PGF2-alpha
PTGFR
PGI2
PTGIR
Thromboxane A2
TBXA2R
Nucleotide like
Adenosine
ADORA
Nucleotides
P2RY
Cannabinoid
Anandamide
CNR1
Platelet-activating factor
PTAFR
Gonadotropin-releasing hormone
GNRHR
Thyrotropin-releasing hormone
TRHR
Melatonin
MTNR
Lysophingolipid and LPA
Lysophosphatidic acid
EDGL
S1P, dihydro-S1P
EDGS
Leukotriene B4
LTB4R
Mas proto-oncogene
MAS1
Relaxin
LGR7/8
Cysteinyl-leukotriene
CYSLTR
Class B Secretin like
Calcitonin
CALCR
Corticotropin releasing hormone
CRHR
Gastric inhibitory peptide
GIPR
Glucagon
GCGR
Glucagon-like peptide
GLPR
Growth hormone-releasing hormone
GHRHR
Parathyroid hormone
PTHR
PACAP
PACAPR
Secretin
SCTR
Vasoactive intestinal peptide
VIPR
Class C Metabotropic glutamate / pheromone
Metabotropic glutamate
GRM
GABA
GABBR
Channels /other receptors
N-Acetylaspartyl-glutamate
GRIN
GABA
Acetylcholine
CHRN
Nucleotides
P2RX
Glutamate
L-Aspartate
L-Cysteic acid
L-Homocysteic acid
GRI
Glycine
β-Alanine
Taurine
GLR
N-Arachidonoyl-dopamine
N-Oleoyl-dopamine
Anandamide
Palmitoyl-ethanolamide
TRPV1
Porphyrins
BZRP
Cortisol
NR3C1
Growth hormone
GH
GHR
Triiodothyronine, thyroxine
THR
Leptin
LEP
LEPR
Prolactin
PRL
PRLR
04080 8/30/11
(c) Kanehisa Laboratories

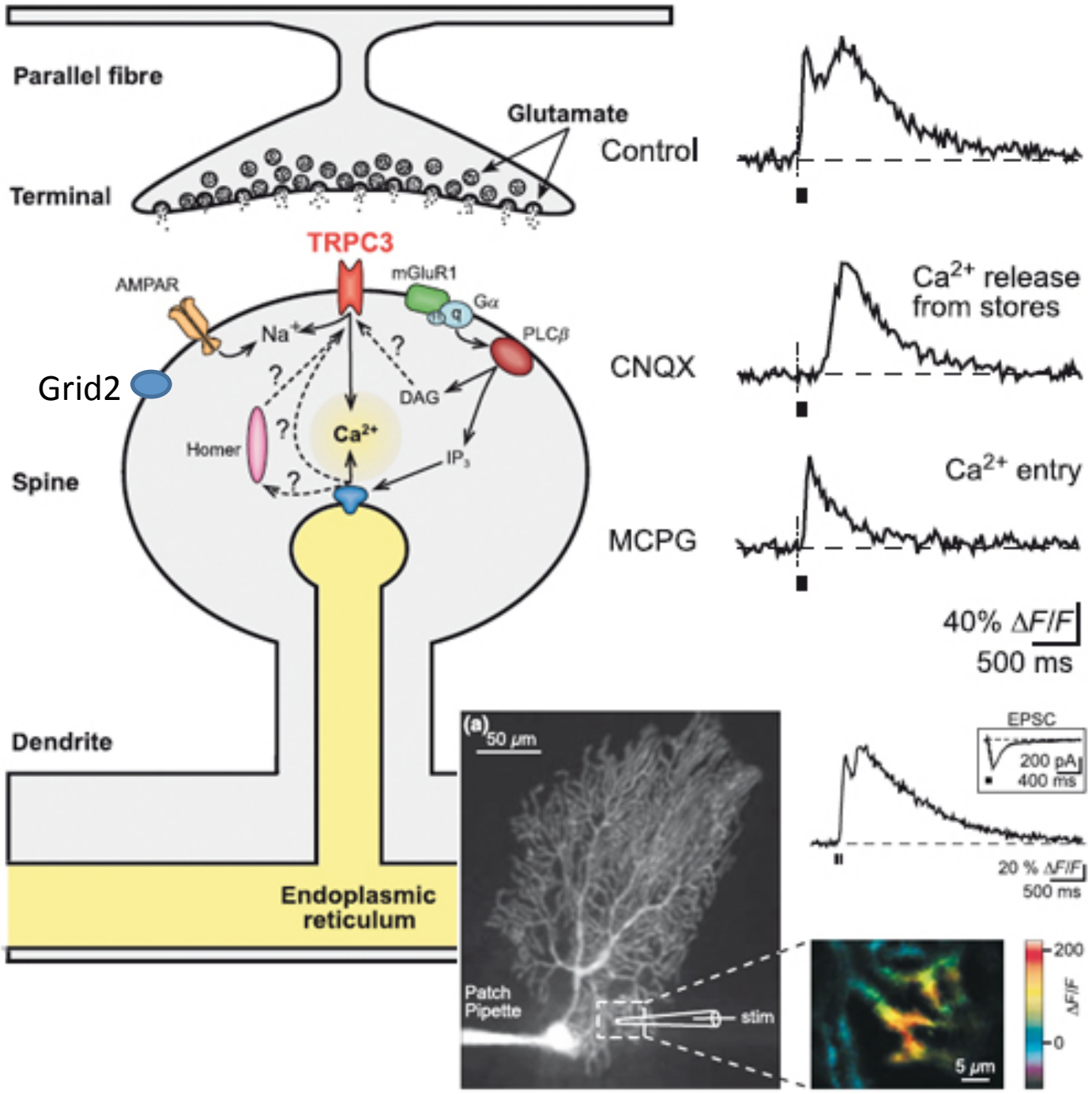

LONG-TERM DEPRESSION
Parallel fiber
Glutamatergic synapse
Purkinje cell (Cerebellum)
GUCY
cGMP
PKG
GS
PP2A
NOS
NO
Ras
Raf
MEK1/2
ERK1/2
Phosphorylation-dephosphorylation (PD) system
52R
G
PLA2
AA
mGluR1
Gq
PLC
DAG
PKC
SG
Na+
PTK
Glutamate
AMPAR
Na+
IP3
ER
EPSP
IP3R
Ca2+
VGCC
RyR
Parallel fiber AMPARs Phosphorylation Internalization
Climbing fiber
EPSP
Na+
AMPAR
Calcium signaling pathway
IP3
Glutamate
mGluR1
Gq
PLC
DAG
G
PLA2
AA
CRF
CRFR1
G
IGF1
IGF1R
G
Endocytosis
04730 5/16/11
(c) Kanehisa Laboratories

GLUTAMATERGIC SYNAPSE

From Climbing Fibers & Parallel fibers

Genes down-regulated:
CFs – Grik1, Col18a1
PFs – Trpc3, Cav3.1, Grid2ip

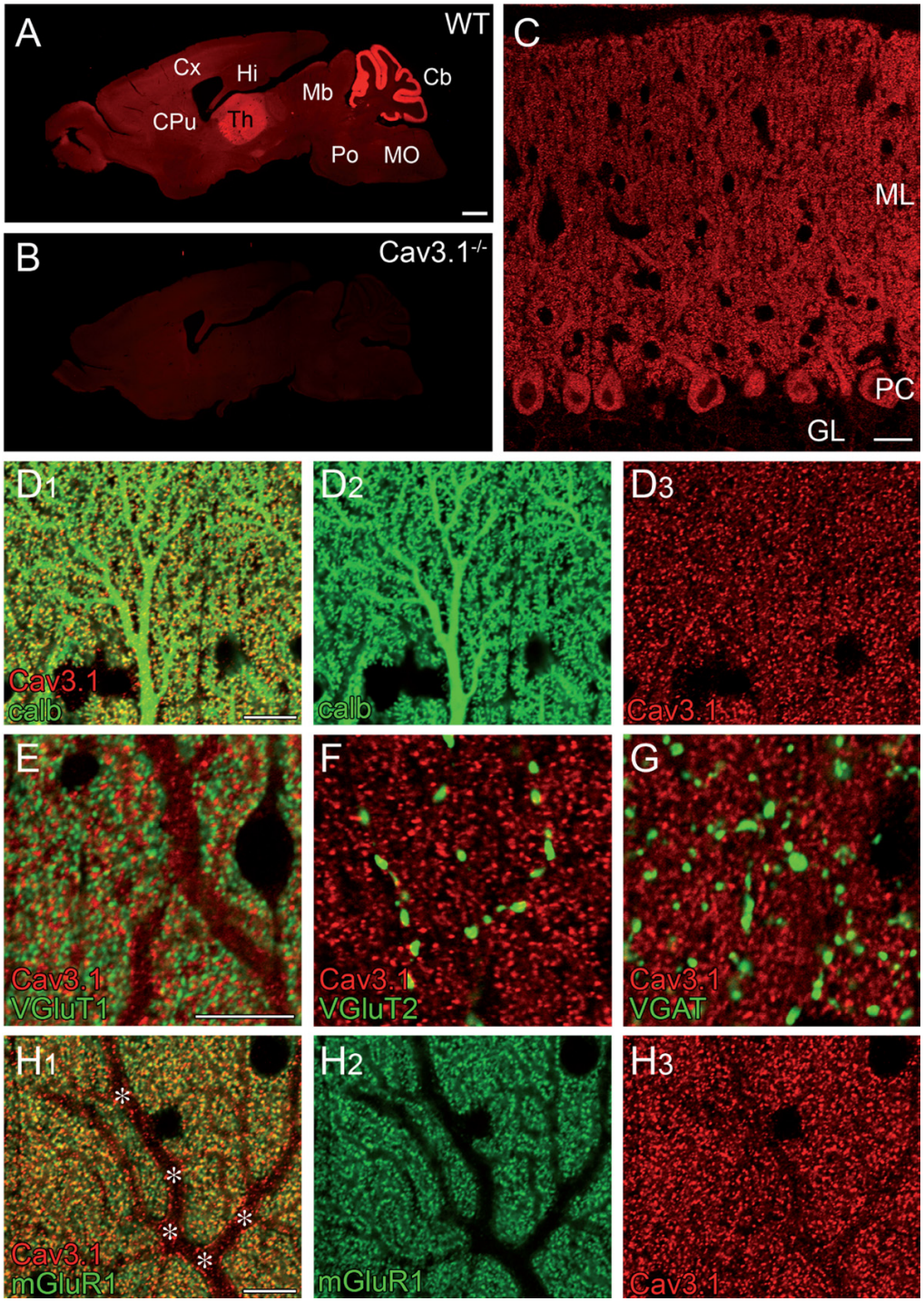



Parallel fibre
Glutamate
Terminal
TRPC3
AMPAR
mGluR1
Gα
q
PLCβ
Na+
?
Grid2
DAG
Homer
Ca2+
IP3
Spine
Dendrite
Endoplasmic reticulum
Control
CNQX
MCPG
Ca2+ release from stores
Ca2+ entry
40% ΔF/F
500 ms
EPSC
200 pA
400 ms
20 % ΔF/F
500 ms
(a)
50 μm
Patch Pipette
stim
5 μm
200
ΔF/F
0

Acta Physiol (Oxf). 2008 Oct 28. [Epub ahead of print]

Mechanisms of metabotropic glutamate receptor-mediated synaptic signaling in cerebellar Purkinje cells.

Hartmann J, Konnerth A.



Cav3.1 slow type are thought to set pacemaker rhythm. Parallel fiber stimulation activates mGluR1-potentiated T-type Ca++ transients

Functional Coupling between mGluR1 and Cav3.1 T-Type Calcium Channels Contributes to Parallel Fiber-Induced Fast Calcium Signaling within Purkinje Cell Dendritic Spines

Michael E. Hildebrand,1* Philippe Isope,2* Taisuke Miyazaki,6 Toshitaka Nakaya,6 Esperanza Garcia,1 Anne Feltz,2 Toni Schneider,5 Ju¨rgen Hescheler,5 Masanobu Kano,3 Kenji Sakimura,4 Masahiko Watanabe,6 Ste´phane Dieudonne´,2 and Terrance P. Snutch1

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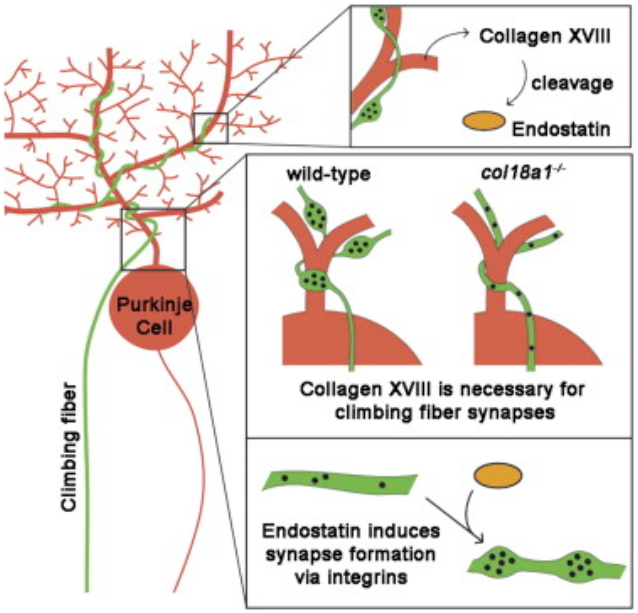
951-8585, Japan, 5Institute of Neurophysiology and Center for Molecular Medicine Cologne, University



Target-Derived Matricryptins Organize Cerebellar Synapse Formation through α3β1 Integrins

Jianmin Su[1], Renee S. Stenbjorn[1], Karen Gorse[1], Kaiwen Su[1], Kurt F. Hauser[2], Sylvie Ricard-Blum[3], Taina Pihlajaniemi[4, 5, 6] and Michael A. Fox[1],

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5 Biocenter Oulu, University of Oulu, Oulu 90014, Finland

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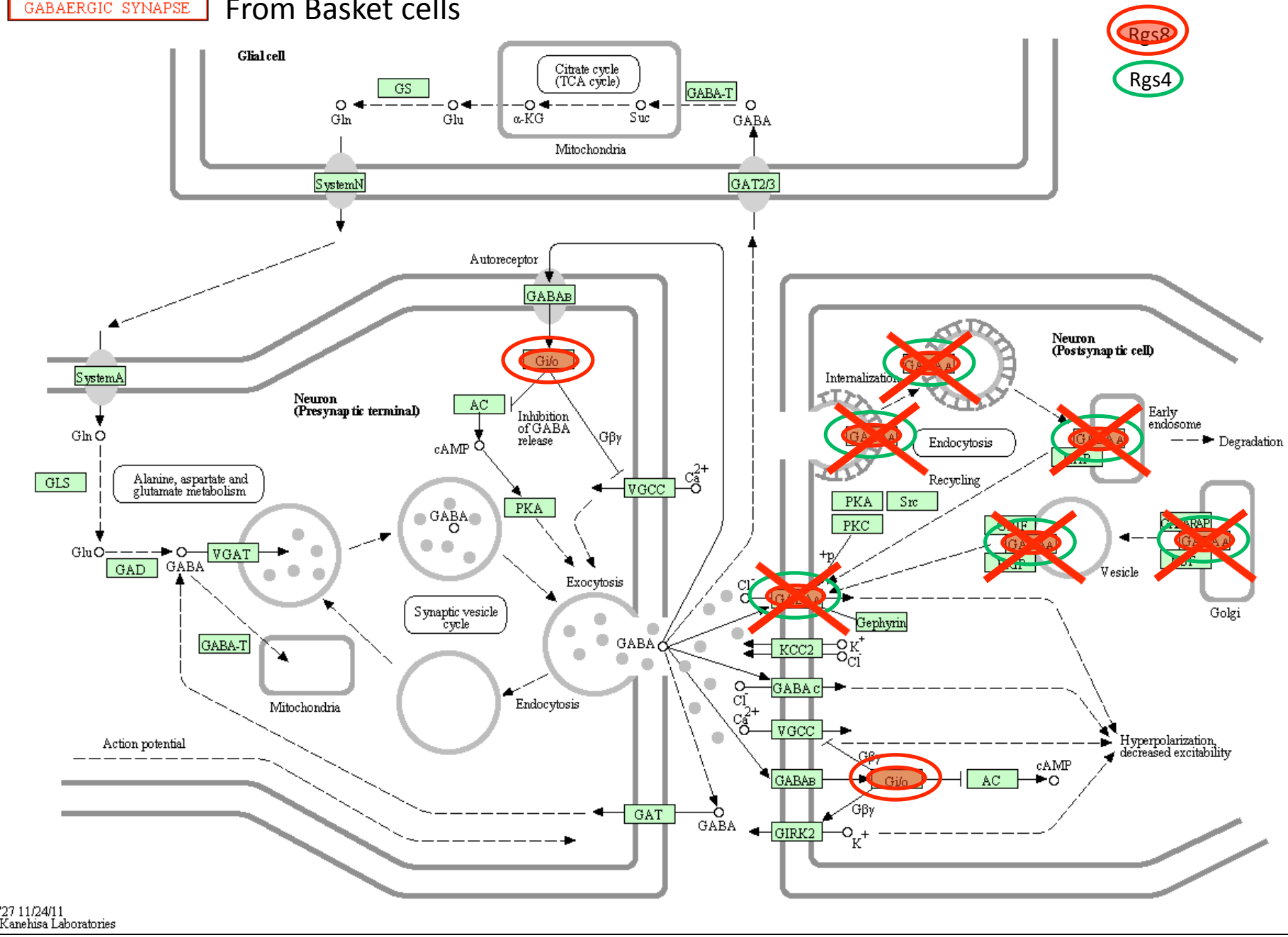
GABAERGIC SYNAPSE

From Basket cells



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Molecular Brain Research 132 (2004) 128–145

RESEARCH

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Research report

Identification of candidate Purkinje cell-specific markers by gene expression profiling in wild-type and *pcd*^{3J} mice

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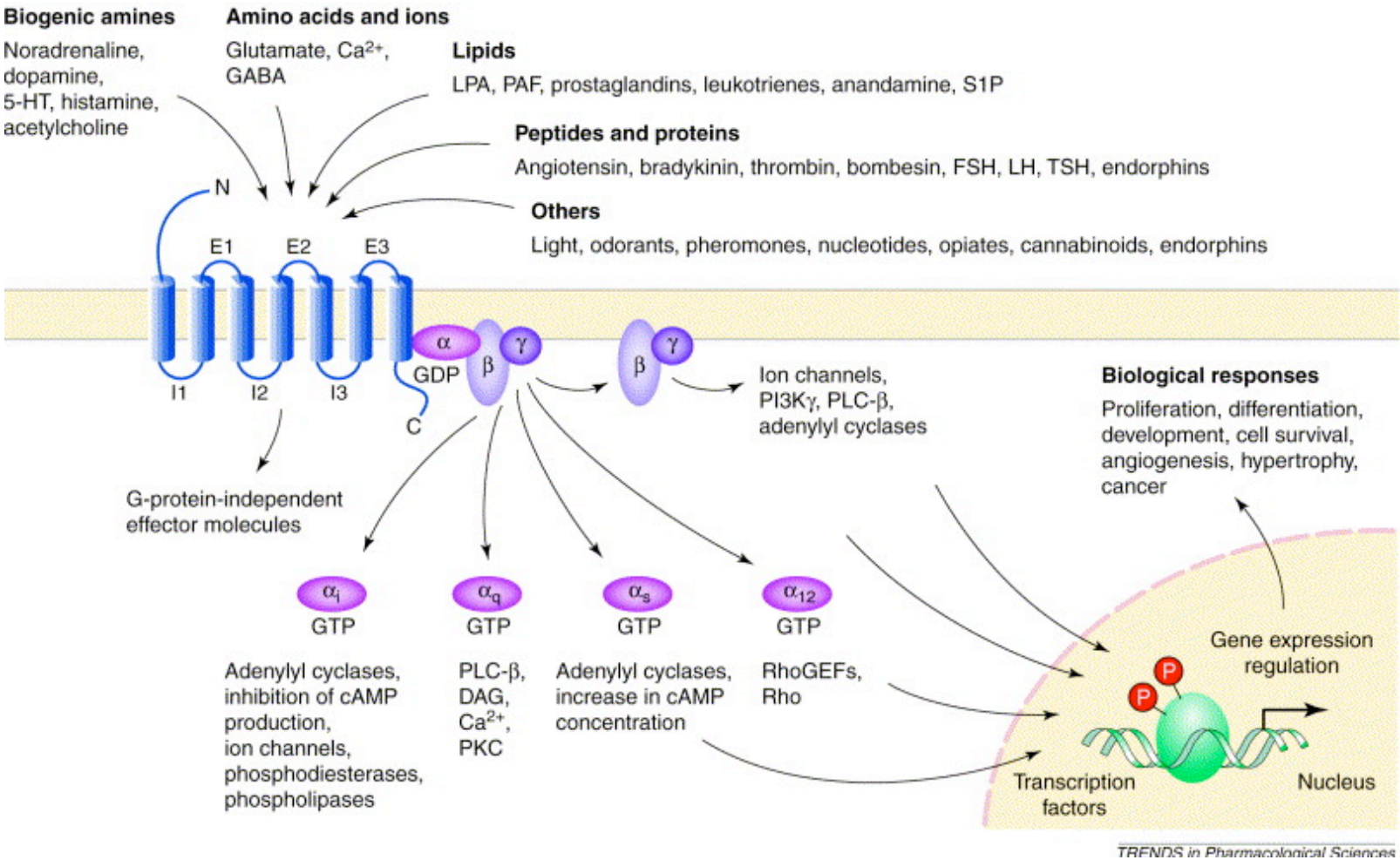
Accepted 7 October 2004

Abstract

The identification of mRNAs that have restricted expression patterns in the brain represents powerful tools with which to characterize and manipulate the nervous system. Here, we describe a strategy using microarray technology (Affymetrix Mouse Genome 430 2.0 Arrays) to identify mRNA transcripts that are candidate markers of cerebellar Purkinje neurons. Initially, gene expression profiles were compared between cerebella of 4-month-old Purkinje cell degeneration (*pcd*^{3J}) mice, in which most Purkinje cells had already degenerated and wild-type littermates with a normal complement of Purkinje neurons. Of 14,563 probe sets expressed in wild-type cerebellum, 797 showed a significant ($p<0.0001$) reduction in *pcd*^{3J} mice. These probes could represent transcripts with varying levels of specificity for Purkinje cells as well as transcripts in other cell types that decline as a secondary consequence of Purkinje cell loss. Ranking of the probe signals revealed that well-known Purkinje cell-specific transcripts such as calbindin and L7/pcp2 clustered in a group that was <33% of wild-type levels. Therefore, to identify potentially new Purkinje cell-specific transcripts that cluster with the known markers, more stringent selection criteria were applied (<33% of wild-type signal and $p<0.0001$). With these criteria, 55 independent transcripts were identified of which 33 were annotated genes and 22 were ESTs and RIKEN cDNAs. A literature search revealed that 25 of the 33 annotated genes were expressed in Purkinje cells, with no data being available on the other 8. Thus, the additional 8 annotated and 22 un-annotated genes are clustered with many genes expressed in Purkinje cells making them candidate markers. To confirm the microarray data, eight representative annotated genes were selected including five reported to be in Purkinje neurons and three for which no data was available. Semi-quantitative RT-PCR demonstrated reduced expression of all eight transcripts in cerebella from *pcd*^{3J} mice. The promoters of genes expressed selectively in subsets of neurons can be used to direct heterologous gene expression in transgenic mice and the more restricted the expression pattern the greater their utility. Therefore, microarray analysis was used to assess expression levels of all 55 transcripts in cerebral cortex, striatum, substantia nigra and ventral tegmental area. This permitted the identification of a set of genes whose promoters might have utility for selectively targeting gene expression to cerebellar Purkinje cells.

See common genes spreadsheet

- **Regulator of G-protein signaling 8** is a protein that in humans is encoded by the *RGS8* gene.[1][2]
- This gene is a member of the regulator of G protein signaling (RGS) family and encodes a protein with a single RGS domain. Regulator of G protein signaling (RGS) proteins are regulatory and structural components of G protein-coupled receptor complexes. They accelerate transit through the cycle of GTP binding and hydrolysis to GDP, thereby terminating signal transduction, but paradoxically, also accelerate receptor-stimulated activation.[2]
- Regulator of G protein signaling (RGS) family members are regulatory molecules that act as GTPase activating proteins (GAPs) for G alpha subunits of heterotrimeric G proteins. RGS proteins are able to deactivate G protein subunits of the Gi alpha, Go alpha and Gq alpha subtypes. They drive G proteins into their inactive GDP-bound forms. Regulator of G protein signaling 4 belongs to this family. All RGS proteins share a conserved 120-amino acid sequence termed the RGS domain. Regulator of G protein signaling 4 protein is 37% identical to RGS1 and 97% identical to rat Rgs4. This protein negatively regulates signaling upstream or at the level of the heterotrimeric G protein and is localized in the cytoplasm.[1]


Biogenic amines
Noradrenaline, dopamine, 5-HT, histamine, acetylcholine
Amino acids and ions
Glutamate, Ca2+, GABA
Lipids
LPA, PAF, prostaglandins, leukotrienes, anandamine, S1P
Peptides and proteins
Angiotensin, bradykinin, thrombin, bombesin, FSH, LH, TSH, endorphins
Others
Light, odorants, pheromones, nucleotides, opiates, cannabinoids, endorphins
N
E1
E2
E3
I1
I2
I3
C
α
β
γ
GDP
Ion channels, PI3Kγ, PLC-β, adenylyl cyclases
Biological responses
Proliferation, differentiation, development, cell survival, angiogenesis, hypertrophy, cancer
G-protein-independent effector molecules
αi GTP
αq GTP
αs GTP
α12 GTP
Adenylyl cyclases, inhibition of cAMP production, ion channels, phosphodiesterases, phospholipases
PLC-β, DAG, Ca2+, PKC
Adenylyl cyclases, increase in cAMP concentration
RhoGEFs, Rho
Gene expression regulation
P
P
Transcription factors
Nucleus
TRENDS in Pharmacological Sciences

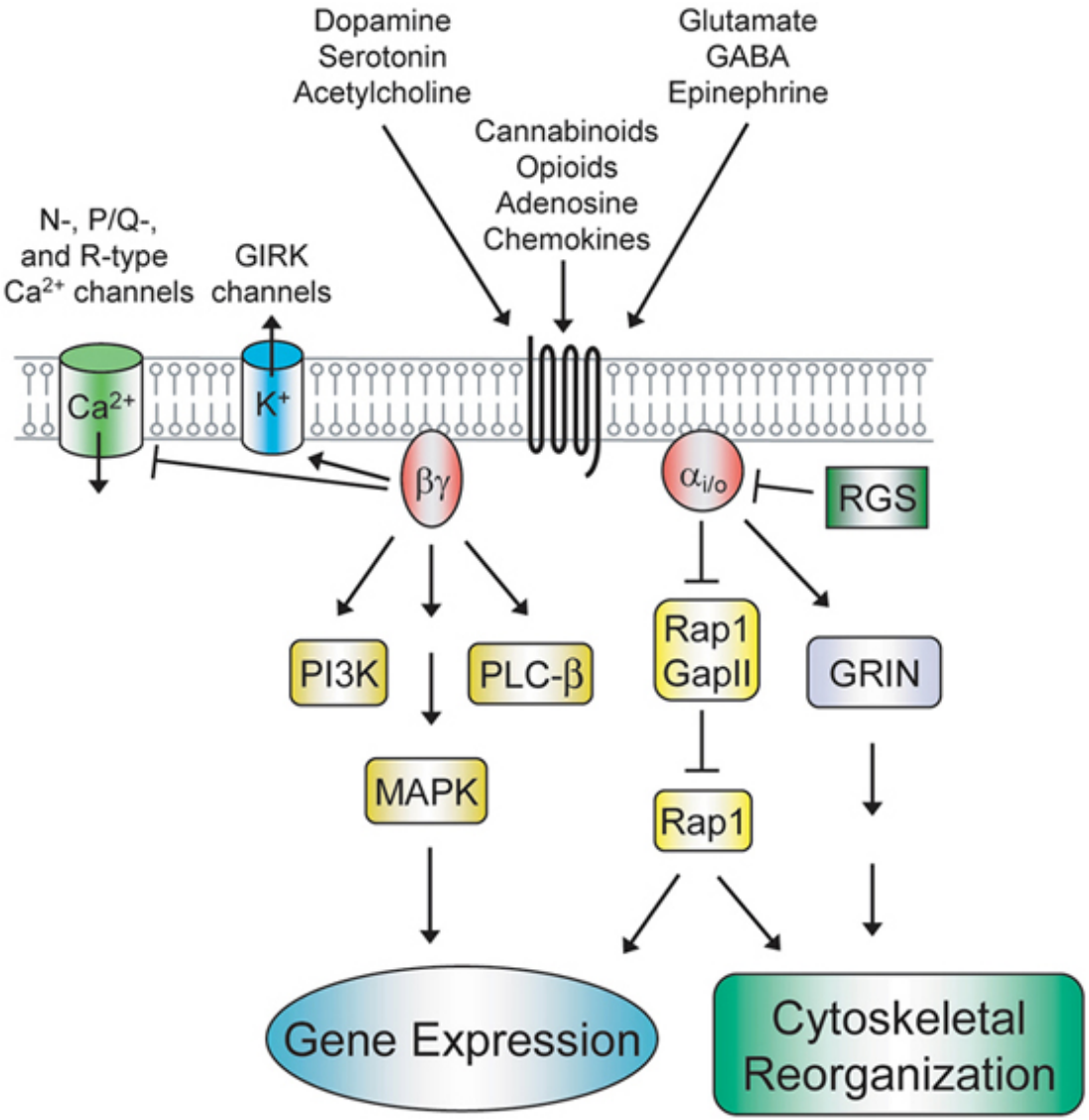


Figure 2. Effector pathways activated by $G_{i/o}$ signaling. Signals from a wide array of hormones,neurotransmitters, and chemokines are transduced into intracellular responses by $G_{i/o}$-coupled receptors. Depicted are pathways that are stimulated by Gα and Gβγ and lead to changes in gene expression and cytoskeletal reorganization. See text for further details. GIRK, G-protein-coupled inward rectifying potassium channels.