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R9AP and R7BP: traffic cops for the RGS7 family in phototransduction and neuronal GPCR signaling

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Abstract

RGS (regulator of G protein signaling) proteins have emerged as crucial regulators, effectors and integrators in G-protein-coupled receptor (GPCR) signaling networks. Many RGS proteins accelerate GTP hydrolysis by Gα subunits, thereby regulating G protein activity, whereas certain RGS proteins also transduce Gα signals to downstream targets. Particularly intriguing are members of the RGS7 (R7) family (RGS6, RGS7, RGS9 and RGS11), which heterodimerize with Gβ5. In *Caenorhabditis elegans*, R7-Gβ5 heterodimers regulate synaptic transmission, anesthetic action and behavior. In vertebrates, they regulate vision, postnatal development, working memory and the action of psychostimulants or morphine. Here we highlight R9AP and R7BP, a related pair of recently identified SNARE-like R7-family binding proteins, which regulate intracellular trafficking, expression and function of R7-Gβ5 heterodimers in retina and brain. Emerging understanding of R7BP and R9AP promises to provide new insights into neuronal GPCR signaling mechanisms relevant to the causes and treatment of neurological disorders.

Introduction

Synaptic transmission is modulated by scores of cell-surface neurotransmitter receptors that are coupled to intracellular heterotrimeric guanine nucleotide-binding proteins (G proteins) consisting of α, β and γ subunits (Box 1; reviewed in Ref. [1]). In addition to their roles in modulatory neurotransmitter signaling, G-protein-coupled receptors (GPCRs) mediate the actions of drugs of abuse or therapeutic agents used to treat various neurological or neuropsychiatric disorders, including chronic pain, depression, schizophrenia and bipolar disorder (reviewed in Ref. [2]). In standard models (Figure 1), agonist-bound GPCRs initiate signaling by catalyzing guanine nucleotide exchange on the Gα subunit of Gαβγ heterotrimers, resulting in the dissociation of GTP-bound Gα subunits from Gβγ dimers, which interact with various effector proteins that regulate transmitter release, postsynaptic response or other processes such as gene transcription and cell survival. Signal deactivation occurs when Gα subunits hydrolyze GTP to GDP, dissociate from effectors and reform Gαβγ heterotrimers.

The RGS (regulator of G protein signaling) protein superfamily has emerged recently as a key component of GPCR signaling pathways (Figure 1). Many RGS proteins are GTPase-activating proteins (GAPs) for Gα subunits, thereby regulating the kinetics, magnitude and fidelity of GPCR signaling (reviewed in Ref. [3]). The importance of RGS-mediated GAP activity is underscored by the pleiotropic phenotypes of knock-in mice expressing RGS-resistant Giα2

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[4–8]. Besides possessing GAP activity, several RGS proteins are effectors and integrators in signal transduction networks (Box 1; reviewed in Ref. [9]).

RGS proteins have been implicated in many disorders of the nervous system, including chronic pain, schizophrenia, anxiety and drug reward and withdrawal [10–22]. RGS proteins might contribute to pharmacogenetic variations within human populations [23–25], or provide new targets for pharmacotherapy of central nervous system disorders [26,27]. Small molecule inhibitors of RGS GAP activity, such as that recently identified for RGS4 [28], might have potential as 'adjuvants' that augment GPCR agonist potency, specificity or duration of action [27]. Targeting RGS protein expression, activity, posttranslational modification, degradation or subcellular localization, might provide additional means of modulating endogenous neurotransmitter or drug action.

In this article we introduce the neuronal RGS7 (R7) family, which has key roles in processes ranging from phototransduction to psychostimulant or morphine action. We highlight new insights into mechanisms that regulate the activity, intracellular trafficking and degradation of R7 proteins, and suggest how further studies of these mechanisms promise to provide deeper understanding of nervous system development, signaling and drug action.

The RGS7 family in *C. elegans* and vertebrates

The prototypical R7 proteins, EGL-10 and EAT-16, were discovered in the worm *C. elegans* by screening for mutations affecting the core machinery that controls synaptic transmission and behavior [29,30]. EGL-10 and EAT-16 share a common domain architecture (Figure 2a) consisting of a conserved N-terminal DEP (disheveled, Egl-10 and pleckstrin), DHEX/R7H (DEP helical extension, or RGS7 homology) and GGL (G protein gamma-like) domains, followed by a C-terminal RGS domain that is necessary and sufficient for GAP activity. Despite having similar domain structures, EGL-10 and EAT-16 have opposing biological functions. Loss of EGL-10 augments Goα signaling, which inhibits locomotion and egg laying [29]. By contrast, loss of EAT-16 augments Gqα signaling, which stimulates these behaviors [30]. EGL-10 and EAT-16 also have opposing effects on the actions of cholinesterase inhibitors and volatile anesthetics [31]. Surprisingly, although the RGS domains of EGL-10 and EAT-16 are required for function, they do not determine which G protein pathways are regulated. Instead, the DEP and DHEX domains of EGL-10 and EAT-16 determine G protein pathway specificity [32]. Whether the DEP and DHEX domains of EGL-10 and EAT-16 restrict the GAP activity of their appended RGS domains to Goα and Gqα, respectively, or determine pathway specificity by other mechanisms remains unknown.

Box 1

G-protein-coupled receptor signaling

The human genome encodes >600 G-protein-coupled receptors (GPCRs) – or 7 transmembrane domain receptors (7TMRs) – that are activated by a diverse array of agonists and stimuli, including catecholamines, amino acids, nucleotides, peptides, hormones, ions, lipid mediators, olfactants and light. Receptor-coupled G proteins are heterotrimers consisting of Gα, Gβ and Gγ subunits, each encoded by a multigene family. Activated GPCRs stimulate exchange GDP for GTP on Gα subunits. GTP-loaded Gα subunits and Gβγ heterodimers subsequently trigger intracellular responses by interacting with downstream effectors.

G protein classes and representative effectors:

$G\alpha_s$-class: $G\alpha_s$, $G\alpha_{sXL}$, $G\alpha_{olf}$

Effectors: adenylyl cyclase, *Axin*

$G\alpha_{i/o}$-class: $G\alpha_{i1-3}$, $G\alpha_o$, $G\alpha_t$, $G\alpha_z$, $G\alpha_{gust}$

Effectors: adenylyl cyclase

$G\alpha_{q/11}$-class: $G\alpha_q$, $G\alpha_{11}$, $G\alpha_{14}$, $G\alpha_{15/16}$

Effectors: phospholipase Cβs, Lbc, G-protein-coupled receptor kinases (*GRKs*)

$G\alpha_{12/13}$-class: $G\alpha_{12}$, $G\alpha_{13}$

Effectors: p115-RhoGEF, LARG, PDZ-RhoGEF, AKAP-Lbc

Gβγ dimers: $G\beta_{1-5}$, $G\gamma_{(1-4,5,7-10,12-14)}$ $G\gamma_{olf}$, $G\gamma_{cone}$

Effectors: adenylyl cyclase, phosphatidylinositol 3-kinaseγ, phospholipase Cβs, GRKs, G protein-regulated inward rectifying K^+ channels (GIRKs), voltage-regulated Ca^{2+} channels (CaV2) (Effectors containing an RGS domain are indicated in *italics*.)

Because EGL-10 and EAT-16 have critical roles in the *C. elegans* nervous system, proteins with similar structures could have important functions in vertebrates. Indeed, vertebrates possess four genes (*RGS6, RGS7, RGS9* and *RGS11*) encoding EGL-10- and EAT-16-like proteins, which comprise the RGS7 (R7) family (reviewed in Ref. [9]). Similar to EGL-10 and EAT-16, the collective R7 family is expressed predominantly and widely in the vertebrate nervous system. Furthermore, the GGL domains of vertebrate R7 proteins, EGL-10 and EAT-16, all bind specifically to a diverged Gβ family member (Gβ5 in vertebrates and GPB-2 in *C. elegans*), forming obligate heterodimers that structurally resemble classical Gβγ complexes (Figure 2b) [33]. However, whereas the Gβ subunit of classical Gβγ heterodimers binds Gα subunits, the Gβ5 subunit of R7-Gβ5 complexes does not, apparently because it is occluded by the DEP and DHEX domains (Figure 2b) [33]. Instead, Gα subunits interact with the RGS domain of R7-Gβ5 heterodimers.

Vertebrate R7 proteins possess several activities expected to impact signaling by scores of modulatory GPCRs, including those having prominent roles in drug action and addiction. Each R7-Gβ5 heterodimer has GAP activity *in vitro* specific for Gi/o-class α subunits [34], which mediate the inhibitory action of GABA(B), D2-type dopamine, mu-opioid and other GPCRs. R7-Gβ5 complexes can also regulate signaling by non-GAP mechanisms. Indeed, R7-Gβ5 complexes containing RGS6 or RGS7 can inhibit the ability of classical Gβγ dimers to activate phospholipase Cβ2 *in vitro* [35], albeit by unknown mechanisms. Furthermore, heterologously expressed RGS7-Gβ5 complexes attenuate Gq signaling by m3 muscarinic receptors by a non-GAP mechanism that remains to be elucidated in detail [36,37].

R7 isoforms are likely to have diverse biological functions because they differ strikingly in their regional expression patterns in brain and retina (Table 1) [38–41]. RGS9–1 is a retinal photoreceptor-specific splice variant that accelerates deactivation of transducin α subunits in rods and cones [42–46]. This mechanism is essential for normal visual perception, as indicated by human RGS9 mutants who adapt slowly to abrupt changes in light intensity, experience difficulty seeing moving, low-contrast objects, and exhibit faster cone responses at low light levels [47–49]. By contrast, mRNA encoding the RGS9–2 splice variant is densely expressed in striatum [50,51], which has critical roles in movement, reward and executive function. Indeed, RGS9–2 regulates psychostimulant and morphine action [20,21], and inhibits D2-like dopamine receptor-mediated modulation of Cav2.2 Ca^{2+} channels and NMDA-evoked currents in striatal neurons [52,53]. RGS7 mRNA is widely and densely expressed in many brain regions, with highest expression in cerebellar granule cells [38]. By contrast, RGS6 and RGS11 transcripts are expressed at low density in restricted patterns [38]. In brain, RGS6 is expressed mainly in olfactory bulb, scattered striatal cells, medial habenula, reticular thalamic, subthalamic, and pontine nuclei, whereas RGS11 is expressed in the subfornical organ, which

controls thirst and fluid balance, and the locus coeruleus, which influences stress, anxiety and addiction. In retina, RGS6, RGS7 and RGS11 proteins are differentially expressed in various nonphotoreceptor layers [39–41], suggesting that they regulate distinct aspects of postphotoreceptor signaling and visual processing. Despite such information, the physiological functions of RGS6, RGS7 or RGS11 in brain and retina remain unknown because studies of the relevant knockout mice have yet to be reported. However, the collective R7 family is important for postnatal development and survival, as well as postsynaptic morphogenesis of retinal ON bipolar neurons, as indicated by phenotypes of Gβ5-knockout mice in which all R7 isoforms are proteolytically degraded [40,54].

R9AP: a retina-specific membrane-trafficking protein for RGS9–1-Gβ5 complexes

Although RGS9–1-Gβ5 complexes associate with photoreceptor disk membranes where their substrate, transducin α subunits, localizes, they lack transmembrane domains or obvious lipid modification motifs, and they are largely soluble when heterologously expressed. However, disk membranes possess saturable, protein binding sites for RGS9–1-Gβ5 heterodimers [55]. Wensel and colleagues therefore identified a disk membrane ‘receptor’ for RGS9–1-Gβ5 heterodimers by showing that RGS9–1 purified from photoreceptor membranes forms a stoichiometric complex with Gβ5, transducin α, and a novel 25 kDa protein termed R9AP (RGS9 anchor protein) [56]. Recombinant R9AP stimulates the GAP activity of RGS9–1-Gβ5 complexes ~4–30-fold by membrane targeting and allosteric mechanisms [57–59]. However, native disk membranes stimulate RGS9–1-Gβ5 GAP activity 70-fold [55], suggesting that GAP regulation is not yet completely understood.

Intriguingly, R9AP is distantly related to syntaxins [57,60], a family of membrane-associated SNARE (soluble N-ethylmaleimide-sensitive factor attachment protein receptor) proteins that mediate vesicle docking/fusion in the secretory pathway (reviewed in Ref. [61]). R9AP contains an N-terminal region predicted to form a trihelical core, a heptad repeat similar to a R-SNARE motif, and a C-terminal transmembrane domain (Figure 2c) [56,57,60]. Indeed, over-expressed R9AP inhibits depolarization-evoked exocytosis in PC12 cells [60], an observation that warrants further investigation.

R9AP protein expression has been documented thus far only in mammalian retina. Here, R9AP localizes to photoreceptor disc membranes (Figure 3a) [56], and possibly also neuronal processes of the outer plexiform layer [56], which receive photoreceptor inputs. In chicken hatchlings, however, R9AP mRNA is more widely expressed, including in retinal photoreceptors, several brain regions, cochlear hair cells, a subpopulation of medium-sized dorsal root ganglion neurons, as well as lung and liver [60]. Whether this broader expression pattern reflects species or developmental differences is unknown.

In accord with its mammalian expression pattern and biochemical function, R9AP is critical for phototransduction regulation and visual perception. Mutations inactivating R9AP in mice or humans produce phototransduction and vision defects like those affecting RGS9 [47,62]. Unexpectedly, however, the absence of R9AP eliminates expression of RGS9–1 protein in photoreceptors [62], apparently by proteolytic degradation. Conversely, overexpression of R9AP in photoreceptors augments RGS9–1 protein expression and augments phototransduction deactivation kinetics [63], indicating that R9AP sets RGS9–1 levels that rate limit this process. Disk membrane targeting of RGS9–1 is required for normal phototransduction deactivation, as indicated by studies of an RGS9–1 mutant lacking its DEP domain [57].

The specialized architecture of photoreceptors requires mechanisms that target rhodopsin, transducin, RGS9–1-Gβ5-R9AP complexes and other components of the visual signal transduction cascade to disk membranes (reviewed in Ref. [64]). R9AP might possess disk trafficking signals because it is properly targeted in the absence of RGS9–1-Gβ5 complexes [57]. Therefore, rather than simply anchoring RGS9–1-Gβ5 to membranes, R9AP might 'steer' them to photoreceptor disk membranes. Perhaps the SNARE-like character of R9AP is relevant for such putative trafficking activity.

R7BP: a universal, regulated membrane-trafficking protein for R7-Gβ5 complexes

Proteins besides R9AP must also be responsible for localizing R7-Gβ5 complexes because mammalian R9AP protein is expressed only in photoreceptors whereas Gβ5 and the collective R7 family are expressed throughout the nervous system. Accordingly, mass spectrometry was used to identify proteins tightly associated with RGS9–2 purified from brain membrane extracts [65]. In another study, bioinformatic techniques were employed to search for genes encoding R9AP-like proteins [66]. Both approaches identified R7BP (RGS7-family binding protein), apparently the sole R9AP-like protein encoded in vertebrate genomes. Like R9AP, R7BP is distantly related to syntaxins, containing a putative N-terminal trihelical domain followed by a heptad repeat region (Figure 2c) [65,66].

However, R7BP and R9AP differ strikingly in several ways, suggesting why vertebrates express two proteins of this class. First, R7BP and R9AP expression patterns in mammals are nonoverlapping (Table 1). Whereas R9AP protein is expressed highly in photoreceptors, R7BP protein is undetectable in photoreceptors but is highly expressed in the inner retina [41]. Moreover, whereas in mammals R9AP protein is undetectable in nonretinal tissues, R7BP is highly expressed in many brain regions and spinal cord [65–67], similar to the aggregate R7 family. Second, whereas R9AP binds only to heterodimers containing Gβ5 and RGS9 or RGS11, R7BP is a universal binding partner for all R7-Gβ5 complexes [65,66], targeting them to the plasma membrane [66,68]. Third, rather than possessing a transmembrane domain like R9AP, R7BP is modified by the fatty acid palmitate attached in thioester linkage to two conserved cysteine residues near its C terminus (Figure 2c), which targets the protein to the plasma membrane [66,68].

Because palmitoylation, unlike other lipid modifications of proteins, is reversible and can be regulated (reviewed in Ref. [69]), R7BP potentially is a regulated membrane anchor protein for the R7 family (Figure 3b). Indeed, blockade of R7BP palmitoylation in cultured cells eliminates plasma membrane targeting and causes R7-Gβ5-R7BP complexes to accumulate in the nucleus [66,68]. Nuclear localization and palmitoylation of R7BP require an evolutionarily conserved polybasic region (PBR) near its C-terminus [66,68]. Nuclear import receptors (importins) and palmitoyltransferases therefore might compete for the PBR to shunt R7BP to the nucleus or plasma membrane. When palmitoylated and targeted to the plasma membrane of cultured cells, R7BP can be depalmitoylated and transported into the nucleus if repalmitoylation does not occur [66]. Once in the nucleus, unpalmitoylated R7BP undergoes nuclear-cytoplasmic shuttling [70], potentially allowing it to be repalmitoylated and targeted to the plasma membrane. Therefore, R7BP palmitoylation and subcellular trafficking are dynamic, as expected if they regulate function of R7-Gβ5-R7BP complexes.

Although apparently surprising, such findings are consistent with evidence indicating that RGS7, RGS9–2 and Gβ5 in brain slices or extracts localize to the cytoplasm, nucleus and plasma membrane [71–74]. However, immunofluorescence and immunoelectron microscopy of brain have failed to detect R7BP within the nucleus [67,75], perhaps because of limited sensitivity. Nevertheless, a relatively small pool R7BP is associated with the nuclear fraction

of brain subcellular extracts [68]. Taken together, such evidence indicates that the vast majority of R7BP in brain is palmitoylated and membrane targeted. However, R7BP could undergo constitutive or signal-induced palmitate cycling such that at steady state only a small fraction of the R7BP pool is unpalmitoylated and nuclear. Addressing such hypotheses and dissecting the functions of nuclear R7-Gβ5-R7BP complexes remain important goals.

Equally surprising, R7BP can augment or inhibit R7-Gβ5 function. When expressed in *Xenopus* oocytes, RGS7-Gβ5 complexes accelerate the gating kinetics GIRK channels by Gi/o-coupled m2 muscarinic receptors. In this system, R7BP augments RGS7-Gβ5 activity by a membrane-targeting mechanism [70]. Similarly, in striatal cholinergic interneurons, R7BP apparently is required for RGS9–2-Gβ5 complexes to attenuate D2 dopamine receptor modulation of Cav2.2 Ca^{2+} channels, because intra-cellular perfusion of a recombinant protein consisting of the DEP and DHEX domains of RGS9 facilitates D2 receptor signaling [52], possibly by competing for a limited pool of R7BP. By contrast, R7BP expression in COS-7 cells inhibits the ability of RGS7-Gβ5 dimers to attenuate Ca^{2+} signaling by Gq-coupled m3 muscarinic receptors [76]. Thus, the R7BP-free pool of RGS7-Gβ5 complexes in brain extracts could inhibit Gq signaling whereas the R7BP-bound pool could not.

R7BP expression, localization and function

Studies of R7BP expression and localization *in vivo* provide further insight into the functions of R7-Gβ5-R7BP complexes. R7BP is an unstable protein that requires R7-Gβ5 complexes for expression [67,75], indicating they are its obligate binding partners. Expression of R7 proteins, Gβ5 and R7BP is strikingly upregulated during the first 2 weeks of mouse postnatal life [67, 75], when synapses mature structurally and functionally pre- and postsynaptically to increase the speed and fidelity of synaptic transmission [77]. This interval of postnatal rodent brain development is equivalent to the human brain ‘growth spurt’, which occurs from the third trimester through the first few years of childhood. Developmental upregulation of R7-Gβ5-R7BP complexes therefore might provide GAP or other activities required for efficient modulatory action of GPCRs as the nervous system matures. Consistent with this hypothesis, immunoelectron microscopy of adult striatum indicates that R7BP localizes with RGS9–2 at postsynaptic densities of excitatory synapses, dendritic shafts and the somatic plasma membrane [75]. RGS9–2-Gβ5-R7BP complexes therefore are poised to regulate postsynaptic modulatory GPCR signaling. R7BP is also detected in unmyelinated axons, but rarely in myelinated axons or nerve terminals [67,75], suggesting that it has less prominent roles in controlling presynaptic GPCR signaling.

Studies of mice lacking or overexpressing R7BP have been reported recently [75], indicating that this protein profoundly affects RGS9–2 expression and localization. In the absence of R7BP, striatal RGS9–2 is delocalized from synaptic membranes, ubiquitinated and degraded by lysosomal cysteine proteases. Conversely, R7BP overexpression post-transcriptionally increases RGS9–2 protein levels. Expression levels of other R7 proteins are insensitive to R7BP deletion or overexpression. Whether RGS6, RGS7 and RGS11 are mislocalized in brain of R7BP-knockout mice remains to be studied. However, this possibility seems likely because RGS7-Gβ5 complexes expressed endogenously in a neuronal cell line are cytoplasmic in the absence of R7BP but plasma membrane targeted in its presence [67].

Physiological implications and future prospects

Whereas normal photoreceptor signaling and vision require R9AP, the physiological functions of R7BP remain to be determined. However, R7BP is poised to control many aspects of nervous system function owing to its wide expression in the adult nervous system, developmental upregulation, synaptic localization, ability to localize each R7 isoform, and potential to be

regulated by reversible palmitoylation. Present understanding indicates that R7BP functions coordinately with RGS9–2, which controls cocaine potency or efficacy, morphine dependence and withdrawal, motor coordination and working memory [20,21,78]. R7BP is also likely to be important for proper localization and function of other R7-Gβ5 complexes. However, R7-Gβ5 heterodimers might have functions beyond those requiring R7BP, because mice lacking all R7-Gβ5 complexes (i.e. Gβ5 knockouts) exhibit a postnatal developmental delay and a partially penetrant preweaning lethality phenotype [54], whereas R7BP-knockout mice apparently do not [75].

In conclusion, by providing new insight into mechanisms linked to synaptic development, transmission and plasticity, future investigations of R7-Gβ5-R7BP complexes promise to advance understanding into the causes or treatment of neurological disorders involving GPCR signaling. Major questions to be answered include: Which neurotransmitter and drug systems are regulated by each R7 isoform? How do R7-Gβ5-R7BP complexes control pre- or postsynaptic signaling? Do R7-Gβ5-R7BP complexes signal within the nucleus to mediate or regulate neurotransmitter or drug action? Does developmental upregulation of R7-Gβ5-R7BP complexes impact synaptic maturation or signaling? Is drug potency, efficacy, specificity, reward, tolerance or withdrawal modulated by altering R7BP palmitoylation or expression? Do mutations or polymorphisms affecting R7-Gβ5-R7BP complexes contribute to human neurological disorders or to the pharmacogenetics of drug action or side effects? Exciting and potentially unanticipated answers to such questions undoubtedly lie ahead.

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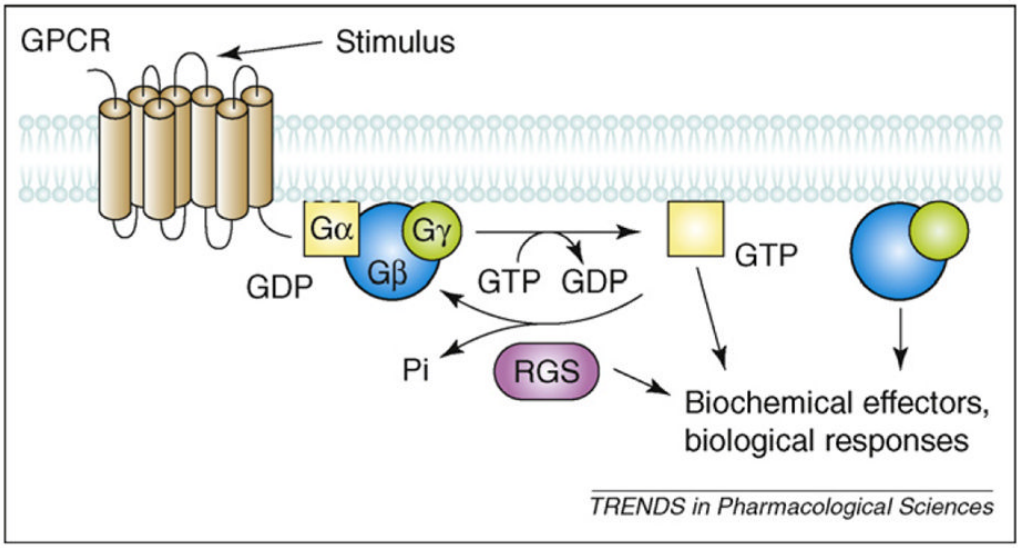

Figure 1.
RGS action in GPCR signaling networks. Many, but not all, proteins containing an RGS domain bind to various classes of activated GTP-bound Gα subunits, accelerating GTP hydrolysis and consequent G protein cycling between active and inactive states. The RGS domains of certain proteins bind specific classes of Gα subunits to transduce signals to downstream signaling effectors or regulators.

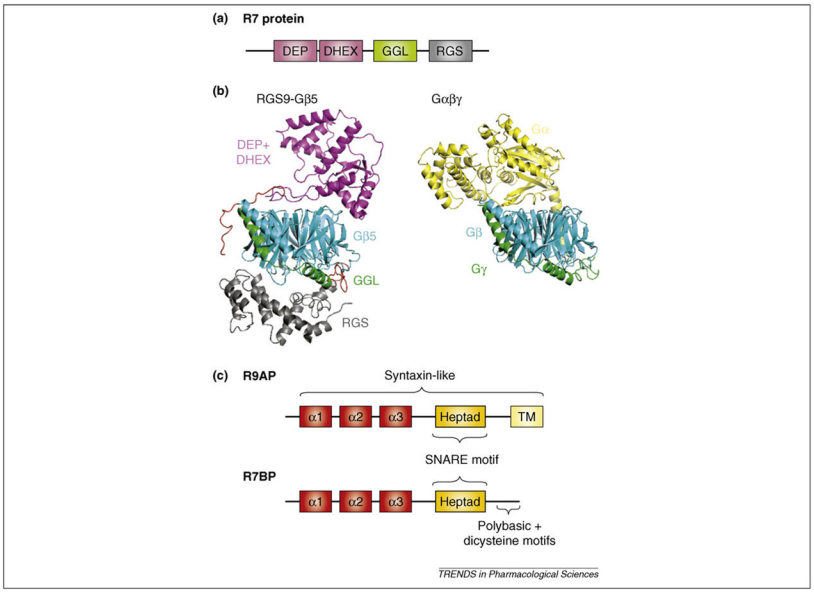

Figure 2.

RGS7-family RGS proteins, Gβ5 and their membrane anchors R9AP and R7BP. **(a)** Domain organization of RGS7-family (R7) proteins. All R7 proteins contain three conserved regions: an N-terminal region consisting of DEP and DHEX domains that bind R9AP and R7BP, a central GGL domain that binds Gβ5, and a C-terminal RGS domain that possesses GAP activity *in vitro* for Gi/oα subunits. **(b)** Structural comparison of RGS9-Gβ5 heterodimers and classical Gαβγ heterotrimers. The RGS9-Gβ5 crystal structure [33] indicates that Gβ5 (cyan) and the GGL domain (green) of RGS9 interact like classical Gβγ subunits, and that the DEP and DHEX domains (magenta) of RGS9 occlude the surface of Gβ5 analogous to that in a typical Gβγ dimer that binds Gα (yellow). **(c)** Domain organization of R9AP and R7BP. R9AP and R7BP are syntaxin-like proteins containing an N-terminal trihelical region (red), a heptad/SNARE motif (orange) and a C-terminal membrane-targeting domain, which in R9AP is a transmembrane segment (yellow) and in R7BP is a palmitoylation signal consisting of a polybasic region and a dicysteine motif. The polybasic region also functions as a nuclear localization sequence when R7BP is unpalmitoylated.

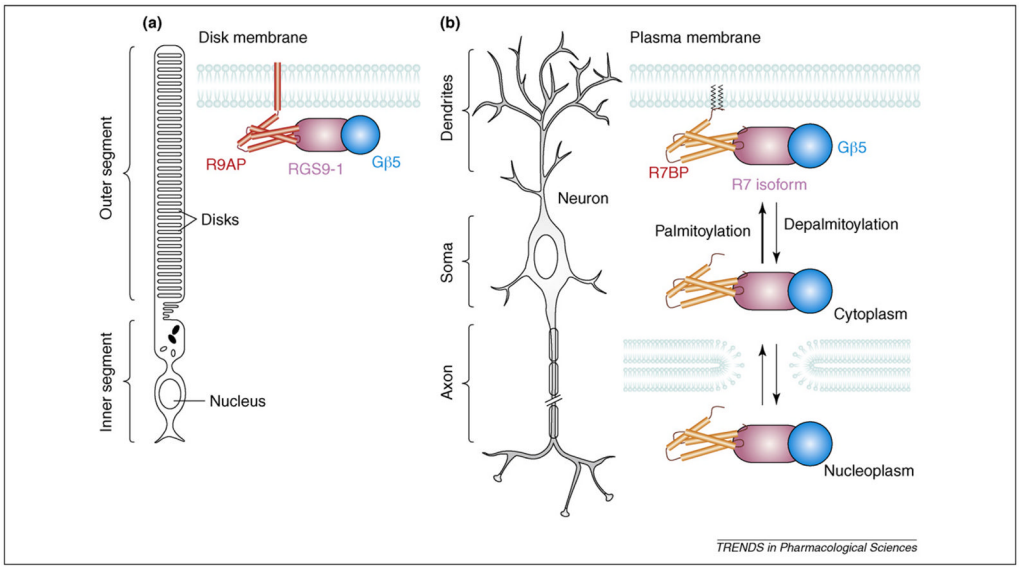

Figure 3.

Intracellular trafficking of photoreceptor and neuronal R7-Gβ5 complexes by R9AP and R7BP. **(a)** Trafficking by R9AP. R9AP localizes RGS9–1 (*purple*) and Gβ5 (*cyan*) heterodimers to the cytoplasmic surface of disk membranes in photoreceptor outer segments where they act as GAPs for transducin α subunits to control light adaptation and visual perception. R9AP is shown as a model based on its sequence similarity to syntaxins of known structure. **(b)** Trafficking by R7BP. R7BP is a palmitoylation-dependent plasma membrane anchor for any R7-Gβ5 (purple and cyan, respectively) heterodimer in neurons. In cultured cells, when R7BP is depalmitoylated it releases from the plasma membrane and shuttles with its bound R7-Gβ5 cargo in and out of the nucleus. Whether this occurs *in vivo* and serves to regulate or transduce signals remains unknown. R7BP is shown as a model based on its sequence similarity to syntaxins of known structure.

Table 1

Regional expression of the R7 family, R7BP, R9AP and Gβ5 in brain and retina

Region	RGS6	RGS7	RGS9	RGS11	R7BP	R9AP	Gβ5
Brain							
—Neocortex							
——Cortical layer I	$-^{r}$ [38]	$-^{r}$ [38], $+++^{p}$ [79]	$-^{r}$ [38], $+^{p}$ [80]	$-^{r}$ [38]	$++++^{p}$ [67]	n.r.	$++^{p}$ [79]
——Cortical layer II–III	$+^{r}$ [38]	$+++^{r}$ [38], $++^{p}$ [79]	$-^{r}$ [38], $+^{p}$ [80]	$-^{r}$ [38]	$++++^{p}$ [67]	n.r.	$+++^{p}$ [79]
——Cortical layer IV	$+^{r}$ [38]	$+^{r}$ [38], $+^{p}$ [79]	$-^{r}$ [38], $+^{p}$ [80]	$-^{r}$ [38]	$+++^{p}$ [67]	n.r.	$+^{p}$ [79]
——Cortical layer V–VI	$+^{r}$ [38]	$+^{r}$ [38], $++^{p}$ [79]	$+^{r}$ [38], $+++^{p}$ [80]	$-^{r}$ [38]	$++++^{p}$ [67]	n.r.	$+++^{p}$ [79]
——Piriform cortex	$+^{r}$ [38]	$+++++^{r}$ [38], $-^{p}$ [79]	$+^{r}$ [38], $+++^{p}$ [80]	$-^{r}$ [38]	n.r.	n.r.	n.r.
—Hippocampus							
——CA1–3 pyramidal cells	$-^{r}$ [38]	$+++^{r}$ [38], $-^{p}$ [79]	$-^{r}$ [38]	$+^{r}$ [38]	$++++^{p}$ [67]	n.r.	$++^{p}$ [79]
——Dentate gyrus granule cells	$-^{r}$ [38]	$+++^{r}$ [38], $-^{p}$ [79]	$+^{r}$ [38]	$+^{r}$ [38]	$++++^{p}$ [67]	n.r.	$++^{p}$ [79]
—Striatum							
——Caudate-putamen	$-^{r}$ [38]	$+^{r}$ [38], $++^{p}$ [79]	$+++++^{r}$ [38],$+++^{p}$ [80]	$-^{r}$ [38]	$++++^{p}$ [67]	n.r.	$+++^{p}$ [79]
——Nucleus accumbens	$-^{r}$ [38]	$+^{r}$ [38], $++^{p}$ [79]	$+++++^{r}$ [38], $++^{p}$ [80]	$-^{r}$ [38]	n.r.	n.r.	$++^{p}$ [79]
—Amygdala							
——Central amygdala	$-^{r}$ [38]	$+^{r}$ [38], $+^{p}$ [79]	$+^{r}$ [38]	$-^{r}$ [38]	n.r.	n.r.	n.r.
——Lateral amygdala	$-^{r}$ [38]	$+++^{r}$ [38], $+^{p}$ [79]	$-^{r}$ [38]	$-^{r}$ [38]	n.r.	n.r.	n.r.
——Medial amygdala	$-^{r}$ [38]	$+++^{r}$ [38], $+^{p}$ [79]	$++^{r}$ [38]	$-^{r}$ [38]	n.r.	n.r.	n.r.
——Medial habenula	$+++++^{r}$ [38]	$+^{r}$ [38], $+++^{p}$ [79]	$+^{r}$ [38]	$-^{r}$ [38]	n.r.	n.r.	$++^{p}$ [79]
Hypothalamus							
—Ventromedial nucleus	$-^{r}$ [38]	$+++^{r}$ [38], $+^{p}$ [79]	$+++^{r}$ [38], $+^{p}$ [80]	$-^{r}$ [38]	n.r.	n.r.	$++^{p}$ [79]
—Arcuate nucleus	$-^{r}$ [38]	$++^{r}$ [38], $+^{p}$ [79]	$++++^{r}$ [38], $+^{p}$ [80]	$-^{r}$ [38]	n.r.	n.r.	$+++^{p}$ [79]
—Subfornical organ	n.r.	n.r.	n.r.	$+++^{r}$ [38]	n.r.	n.r.	n.r.
—Thalamus							
—Midline/intralaminar nucleus	$-^{r}$ [38]	$+++^{r}$ [38], $++^{p}$ [79]	$-^{r}$ [38]	$-^{r}$ [38]	$+^{p}$ [67]	n.r.	$+^{p}$ [79]
—Reticular nucleus	$++++^{r}$ [38]	$+^{r}$ [38], $++^{p}$ [79]	$-^{r}$ [38]	$-^{r}$ [38]	n.r.	n.r.	$++^{p}$ [79]
—Subthalamic nucleus	$+++++^{r}$ [38]	$++++^{r}$ [38], $++^{p}$ [79]	$-^{r}$ [38]	$-^{r}$ [38]	$++++^{p}$ [67]	n.r.	$+++^{p}$ [79]
Brainstem							
—Substantia nigra	$++^{r}$ [38]	$++^{r}$ [38], $++^{p}$ [79]	$-^{r}$ [38], $++^{p}$ [80]	$-^{r}$ [38]	$+++^{p}$ [67]	n.r.	$++^{p}$ [79]
—Pontine nucleus	$+++++^{r}$ [38]	$+++^{r}$ [38], $++++^{p}$ [79]	$-^{r}$ [38]	$+^{r}$ [38]	n.r.	n.r.	$++^{p}$ [79]
—Dorsal raphe nucleus	$+^{r}$ [38]	$++^{r}$ [38], $+^{p}$ [79]	$+^{r}$ [38], $+^{p}$ [80]	$+^{r}$ [38]	n.r.	n.r.	$++^{p}$ [79]
—Locus coeruleus	$+++^{r}$ [38]	$+++++^{r}$ [38]	$+^{r}$ [38], $+++^{p}$ [80]	$+++^{r}$ [38]	n.r.	n.r.	n.r.
Cerebellum							
—Purkinje cell layer	$+^{r}$ [38]	$-^{r}$ [38], $-^{p}$ [79]	$-^{r}$ [38]	$-^{r}$ [38]	$+++++^{p}$[67]	n.r.	$+++^{p}$ [79]
—Granule cell layer	$-^{r}$ [38]	$++++^{r}$ [38], $+++^{p}$ [79]	$-^{r}$ [38]	$-^{r}$ [38]	$++++^{p}$ [67]	n.r.	$+^{p}$ [79]
—Olfactory tubercle	$-^{r}$ [38]	$+^{r}$ [38]	$+++++^{r}$ [38], $++++^{p}$ [80]	0^{r}[38]	n.r.	n.r.	n.r.
Retina							
—Photoreceptor outer segment	$++^{p}$ [41]	$-^{p}$ [41]	$++++^{p}$ [41]	$+++^{p}$[41]	$-^{p}$ [41]	$++++^{p}$[41]	$++++^{p}$ [40]
—Photoreceptor inner segment	$+++^{p}$ [41]	$-^{p}$ [41]	$++^{p}$ [41]	$+++^{p}$[41]	$-^{p}$ [41]	$-^{p}$ [41]	$++^{p}$ [40]
—Outer nuclear layer	$++^{p}$ [41]	$-^{p}$ [41]	$+^{p}$ [41]	$++^{p}$ [41]	$-^{p}$ [41]	$-^{p}$ [41]	$+^{p}$ [40]
—Outer plexiform layer	$++^{p}$ [41]	$+^{p}$ [41]	$-^{p}$ [41]	$++^{p}$ [41]	$++++^{p}$ [41]	$-^{p}$ [41]	$+++^{p}$ [40]
—Inner nuclear layer	$+^{p}$ [41]	$+^{p}$ [41]	$-^{p}$ [41]	$++^{p}$ [41]	$+^{p}$ [41]	$-^{p}$ [41]	$+^{p}$ [40]
—Inner plexiform layer	$++^{p}$ [41]	$++++^{p}$ [41]	$-^{p}$ [41]	$+^{p}$ [41]	$++++^{p}$ [41]	$-^{p}$ [41]	$+++^{p}$ [40]
—Ganglion cell layer	$-^{p}$ [41]	$-^{p}$ [41]	$-^{p}$ [41]	$-^{p}$ [41]	$+++^{p}$ [41]	$-^{p}$ [41]	$-^{p}$ [40]

Labeling densities: very dense, +++++; dense, ++++; moderately dense, +++; light, ++; faint, +; not detectable, −.

Abbreviations: ^{r}, mRNA expression; ^{p}, protein expression; n.r., not reported.

References cited are in brackets.