Published in final edited form as:

Nat Rev Mol Cell Biol. 2009 December ; 10(12): 819–830. doi:10.1038/nrm2803.

Fine-tuning of GPCR activity by receptor-interacting proteins

Stefanie L. Ritter and Randy A. Hall

Department of Pharmacology, Emory University School of Medicine Atlanta, GA, USA, 30322

Preface

G protein-coupled receptors (GPCRs) mediate physiological responses to a large variety of different hormones, neurotransmitters, sensory stimuli and other ligands. The signaling and trafficking properties of GPCRs are often highly malleable depending on cellular context. Such fine-tuning of GPCR function can be attributed in many cases to receptor-interacting proteins that are differentially expressed in distinct cell types. In some cases, these GPCR-interacting partners directly mediate receptor signaling, whereas in other instances the interacting partners serve mainly as scaffolds to modulate G protein-mediated signaling. Furthermore, GPCR-interacting proteins can also exert profound regulation over receptor trafficking, localization and/or pharmacological properties.

Introduction

G protein-coupled receptors (GPCRs) comprise the largest family of transmembrane proteins in vertebrates and are the molecular targets for nearly half of the therapeutic drugs that are prescribed worldwide¹. The approximately 1000 members of this family can be divided into three main subfamilies (termed A, B, and C) based on sequence similarity, with all members exhibiting a conserved seven-transmembrane domain topology. The canonical view of how GPCRs regulate cellular physiology is that the binding of ligands (such as hormones, neurotransmitters or sensory stimuli) induces conformational changes in the receptors' transmembrane and intracellular domains, thereby allowing receptor interactions with heterotrimeric G proteins. Activated receptors act as guanine nucleotide exchange factors (GEFs) for the G α subunits, catalyzing the release of GDP and the binding of GTP to activate the G proteins. The activated G protein subunits (G α and G $\beta\gamma$) can then associate with downstream effectors to modulate various aspects of cellular physiology.

In addition to interacting with G proteins, agonist-bound GPCRs also associate with G protein-coupled receptor kinases (GRKs), leading to receptor phosphorylation. GRKs are a seven-member family of related kinases (GRK1-7) that have differential patterns of distribution across bodily tissues and distinct preferences for certain receptors²⁻⁴. However, a common property of GPCR phosphorylation by GRKs is decreased receptor interactions with G proteins and enhanced receptor interactions with arrestins, which are members of a family of four closely-related scaffold proteins. GPCR interactions with arrestins serve to further turn off receptor signaling through G proteins while simultaneously turning on certain other signaling pathways that are initiated via arrestin-mediated recruitment of signaling proteins to activated receptors². Furthermore, arrestins can directly link active receptors to clathrin-coated pits to facilitate receptor endocytosis, which is an important process controlling the desensitization and resensitization of GPCR activity⁵.

GPCR interactions with G proteins, GRKs and arrestins have been intensively studied for a large number of receptors and exhaustively reviewed elsewhere²⁻⁵. For this reason, these broadly-important interactions, which now represent a canonical model of GPCR regulation (Box 1), will not be further reviewed here. Similarly, the importance of homomeric and

heteromeric interactions between GPCRs has also been thoroughly reviewed elsewhere⁶⁻⁸ and will not be addressed here. The focus of this review will be on recent advances in the characterization of receptor-selective GPCR associations with various proteins outside of the four previously-mentioned families of general GPCR-interacting proteins (G proteins, GRKs, arrestins and other receptors). These receptor-selective partners can mediate GPCR signaling, organize receptor signaling through G proteins, direct GPCR trafficking, anchor GPCRs in particular subcellular areas and/or influence GPCR pharmacology. Since many of these receptor-selective partners exhibit limited patterns of tissue expression, these interactions can help to explain many examples of cell-specific fine-tuning of GPCR functional activity.

Main Text

Mediation of GPCR Signaling

For a GPCR-interacting protein to be considered as a mediator of GPCR signaling, it would seem a requirement that the protein's interaction with the receptor should be regulated by agonist stimulation, since agonist-induced changes are the essence of receptor-initiated signaling. For example, GPCR interactions with G proteins, GRKs and arrestins are all strongly enhanced by agonist stimulation¹⁻⁵. Beyond G proteins, GRKs and arrestins, certain other GPCR-interacting proteins have been shown to interact with specific receptors in an agonist-promoted fashion to mediate particular aspects of receptor signaling, and these examples will be considered here in this section. In contrast, a number of other GPCR-interacting proteins associate with receptors in an agonist-independent manner. Such proteins can potentially modulate G protein-mediated signaling, as described in the next section, but they should not themselves be considered as mediators of receptor signaling if their interactions with receptors are not influenced by agonist stimulation. If a GPCR can be considered as analogous in some ways to a gun, ligand-dependent interactors that mediate signaling are analogous to bullets, whereas ligand-independent interactors that modulate signaling are analogous to silencers and scopes, which influence gun function but do not directly mediate the effects of guns on targets.

Agonist-promoted interactors that mediate signaling—Several different GPCRs have been shown to initiate cellular signaling through agonist-promoted interactions with members of the Janus (Jak) family of non-receptor protein tyrosine kinases. For example, the agonist-dependent activation of the AT1 angiotensin receptor can recruit a complex of Jak2 and the tyrosine phosphatase SHP-2 to associate with the receptor's C-terminus, facilitating Jak2 phosphorylation and activation^{9, 10}. Activated Jak2 can then recruit and phosphorylate members of the STAT family of transcription factors, leading to STAT dissociation from Jak2 and translocation to the nucleus (Figure 1A). The agonist-promoted interaction between Jak2 and AT1 demonstrates an additional signaling avenue for AT1, beyond the receptor's well-established coupling to Gαq, and can help to explain certain effects of AT1 stimulation on cellular physiology that are not explained by G protein-mediated signaling¹¹. Interestingly, AT1 receptor coupling to Gαq can induce rises in intracellular calcium that further potentiate Jak/STAT signaling by the receptor^{12, 13}, providing an example of how G protein-dependent and G protein-independent signaling mechanisms can in some cases work together synergistically. Another GPCR that can interact with Jak2 is the platelet activating factor (PAF) receptor, which associates with a Tyk2/Jak2 complex in an agonist-regulated manner^{14, 15}. A mutant version of the PAF receptor that does not couple to G proteins but still interacts with the Tyk2/Jak2 complex is fully capable of activating downstream STATs, demonstrating the physiological importance of the receptor's recruitment of Jak2¹⁵.

GPCR interactions with proteins that possess PDZ domains can also in some cases mediate agonist-promoted GPCR signaling. PDZ domains, named for the first three proteins in which they were discovered (PSD-95, Discs-large and ZO-1), can mediate high-affinity interactions with specific motifs at the distal C-termini of target proteins¹⁶. For example, the PDZ protein

Na⁺/H⁺ exchange regulatory factor 1 (NHERF-1; also known as EBP50) has been shown to associate in an agonist-promoted fashion with the C-terminus of the β_2 -adrenergic receptor (β_2 -AR)^{17, 18}. The recruitment of NHERF-1 to the β_2 -AR disrupts the ability of NHERF-1 to inhibit the Na⁺/H⁺ exchanger type 3 (NHE3), providing a G protein-independent mechanism by which the β_2 -AR can activate Na⁺/H⁺ exchange in kidney cells¹⁷. The κ opioid receptor (κ OPR) is another GPCR that can regulate Na⁺/H⁺ exchange via agonist-induced interactions with NHERF-1^{19, 20}. Interestingly, the studies on κ OPR provide an example of how interaction with a protein such as NHERF-1 can confer cell-specific signaling to a given GPCR, as κ OPR stimulation robustly activates NHE3 activity in cell lines expressing high levels of NHERF-1 but not in other cell lines that lack significant NHERF-1 expression²⁰.

Agonist-disrupted interactors that mediate signaling—In addition to the aforementioned examples of agonist-promoted associations with receptor-interacting proteins mediating aspects of GPCR signaling, agonist-disrupted interactions between a GPCR and a cytoplasmic binding partner can also initiate cellular signaling. For example, it has been shown that agonist stimulation of the somatostatin receptor type 2 (SSTR2) disrupts its constitutive association with the phosphatidylinositol 3-kinase (PI3K) subunit p85 to negatively regulate PI3K signaling²¹ (Figure 1B). In the absence of agonist, association of p85 with the first intracellular loop of SSTR2 constitutively enhances PI3K activity to promote cell survival through the Akt pathway. However, following agonist stimulation of SSTR2, association of the receptor with p85 is disrupted, leading to decreased PI3K activity and sensitization of cells to stimuli that induce apoptosis²¹. Thus, even though the classical examples of proteins that mediate GPCR signaling (G proteins and arrestins) exhibit enhanced associations with receptors following agonist stimulation, it is evident that as long as a GPCR-interacting partner exhibits some type of change in its location and/or activity in response to agonist stimulation, this can be sufficient to lead to the initiation of cellular signaling.

Modulation of GPCR Signaling

Some GPCR-interacting partners increase the speed and efficiency of GPCR signaling by acting as scaffolds to tether downstream effectors in close proximity to the receptor. On the other hand, certain GPCR-interacting partners can decrease the intensity and/or time course of GPCR signaling by disrupting association with G proteins or, in some cases, recruiting negative regulators of GPCR signaling. By finely tuning the spatial and temporal resolution of signaling, certain GPCR interactors can dramatically affect the ability of GPCRs to transduce extracellular stimuli into changes in cellular physiology.

Interactors that enhance G protein-mediated signaling—The visual system of the fruit fly *Drosophila melanogaster* has long served as an important model system for studying fast and efficient G protein-mediated signaling. Visual signaling in *Drosophila* is mediated by light stimulation of rhodopsin, which couples to G α_q to activate phospholipase C β (PLC β), leading to the generation of the second messengers inositol triphosphate (IP₃) and diacylglycerol and the opening of transient receptor potential (TRP) calcium channels. The influx of calcium and production of diacylglycerol lead to activation of protein kinase C (PKC), which plays a key role in the termination of visual signaling. Interestingly, almost all of the components of the *Drosophila* visual signaling pathway are known to be tethered together by a large PDZ-domain containing scaffold protein known as InaD, which associates with rhodopsin, TRP channels, PLC β and PKC²²⁻²⁸. By tethering these downstream effectors in close proximity to rhodopsin, InaD creates an efficient signaling complex that dramatically increases the speed and amplitude of physiological responses to light stimulation²⁹.

Analogous to *Drosophila* InaD, some mammalian PDZ scaffolds have been found to interact with GPCRs to enhance the efficiency of receptor-stimulated G protein signaling. For example,

associations of NHERF-1 and/or the closely-related protein NHERF-2 with several different GPCRs, including the PTH1 parathyroid hormone receptor³⁰⁻³⁴, LPA₂ lysophosphatidic acid receptor³⁵, P2Y1 purinergic receptor³⁶ and mGluR5 metabotropic glutamate receptor³⁷ can enhance PLC-mediated signaling by these receptors. Unlike the aforementioned interactions of NHERF-1 with β_2 AR and κ OPR, which are regulated by agonist, the associations of the NHERF proteins with PTH1-R, LPA₂-R, P2Y1-R and mGluR5 are not altered by agonist stimulation and mainly seem to facilitate enhanced G protein-mediated signaling by the receptors. Interestingly, many known NHERF-binding partners (beyond GPCRs and NHE3) are downstream components of G α_q /PLC signaling pathways, including G α_q ³⁸, several TRP channels^{39,40}, various isoforms of PLC β ^{41,36}, PKC⁴² and protein kinase D (PKD)⁴³. The interaction of NHERF-2 with PTH1-R provides a particularly compelling example as to how receptor-interacting scaffolds can help to explain cell-specific fine-tuning of receptor signaling. PTH1-R signals mainly through adenylyl cyclase (AC) regulation in ROS 17/2.8 cells, which do not express detectable levels of NHERF proteins, but mainly through PLC regulation in ECV304 cells, which contain high levels of both NHERF-1 and NHERF-2³⁰. PTH1-R has long been enigmatic for seeming to signal primarily through AC regulation in some cell types but PLC regulation in other cells, and it now seems that such cell-specific differences in the receptor's signaling activity can be accounted for in many cases by differential cellular expression of the NHERF proteins⁴⁴ (Fig. 2A).

In addition to the examples of the NHERF proteins and InaD, a variety of other PDZ scaffolds have been shown to associate with specific GPCRs to enhance certain signaling pathways. For example, associations of the multi-PDZ protein Mupp1 with GABA_B receptors⁴⁵ and MT₁ melatonin receptors⁴⁶ result in markedly enhanced G α_i -mediated signaling following receptor stimulation. Similarly, interactions of the PDZ scaffold MAGI-3 with Frizzled-4⁴⁷ and the LPA₂ receptor⁴⁸ enhance receptor-mediated activation of ERK/mitogen-activated protein (MAP) kinase pathways. Conversely, associations of the LPA₂ receptor with two related PDZ scaffolds, PDZ-RhoGEF and LARG, do not result in enhanced downstream MAP kinase activation but rather potentiate LPA₂-induced stimulation of Rho signaling to modify cytoskeleton dynamics⁴⁹. Thus, the LPA₂ receptor can preferentially couple to downstream PLC activation³⁵, MAP kinase activation⁴⁸ or Rho activation⁴⁹ in a cell-specific manner depending upon which LPA₂-interacting PDZ scaffold is expressed in a particular cell type.

Other prominent examples of GPCR-associated proteins that enhance the efficiency of G protein-mediated signaling are the Homer proteins, which associate with mGluR1 and mGluR5⁵⁰⁻⁵³, and members of the A-kinase anchoring protein (AKAP) family, which interact with β -adrenergic receptors⁵⁴⁻⁵⁸. In addition to associating with mGluRs, the Homer proteins also interact with intracellular IP₃ receptors, thereby serving to link the IP₃ receptors, mGluRs and other components together to greatly increase the efficiency of mGluR-stimulated calcium signaling^{52, 59, 60}. The AKAPs were originally named because of their associations with protein kinase A (PKA), and indeed the interactions of AKAP79⁵⁵ and AKAP250^{54, 56, 57} with β_2 AR serve to tether PKA in the vicinity of the receptor and increase the efficiency of PKA-mediated phosphorylation of various substrates that are downstream of receptor activation, including the receptor itself. The consequences of the increased functional relationship between PKA and β_2 AR include enhanced efficiency of receptor resensitization^{54, 57} and more robust β_2 AR-mediated ERK signaling⁵⁵.

Interactors that reduce G protein-mediated signaling—In contrast to the examples described above of GPCR-interacting proteins that increase the efficiency of certain GPCR-stimulated signaling pathways, some proteins that associate with GPCRs can decrease the efficiency of G protein-mediated signaling. The arrestins are perhaps the most general example of this phenomenon. A more receptor-specific example is spinophilin, which has been shown to interact with the third intracellular loops of a handful of different GPCRs, including members

of the dopamine, adrenergic and muscarinic acetylcholine receptor families⁶¹⁻⁶⁵. Spinophilin also binds to several members of the regulators of G-protein signaling (RGS) family of proteins, which tightly regulate the intensity and time course of GPCR signaling by accelerating the inherent GTPase activity of activated G α subunits. Thus, spinophilin can tether RGS proteins in close proximity to receptors to attenuate receptor-stimulated G protein signaling^{63, 64}. Interestingly, RGS proteins can also sometimes associate directly with the intracellular regions of certain GPCRs to inhibit receptor signaling and exert cell-specific regulation of receptor activity⁶⁶⁻⁷¹. RGS proteins have been reviewed in detail elsewhere^{72, 73}, and will therefore not be discussed further here.

Calmodulin is another protein that can interact with a variety of different GPCRs to modulate receptor functional properties. Calmodulin is a widely-expressed calcium-binding protein that can associate in a calcium-sensitive manner with metabotropic glutamate⁷⁴⁻⁷⁷, serotonin⁷⁸⁻⁸⁰, dopamine^{81, 82} and other⁸³⁻⁸⁵ receptors. The functional effects of calmodulin interaction vary depending on the receptor, but perhaps the most commonly-reported effect of calmodulin interaction with GPCRs is an attenuation of G protein coupling^{79, 81, 83, 85}. Since stimulation of many GPCRs can result in downstream increases in cellular calcium levels, the calcium-dependent interactions of calmodulin with GPCRs can in some cases represent a form of feedback inhibition that restrains receptor-initiated G protein signaling. However, G protein-independent signaling pathways may actually be potentiated by receptor interactions with calmodulin, as for example calmodulin association with the serotonin 5-HT_{2C} receptor strongly promotes arrestin-mediated signaling (but not G protein-mediated signaling) by the receptor in both transfected cells and cultured neurons⁸⁰.

Two other proteins that can associate with a select number of GPCRs to tone down G protein-mediated signaling are periplakin and neurochondrin. Periplakin was first reported to associate with the C-terminus of the μ opioid receptor (μ OPR)⁸⁶ and the melanin-concentrating hormone receptor 1 (MCH_{1R})⁸⁷, while neurochondrin was found to interact with the same region of the MCH_{1R} C-terminus as periplakin⁸⁸. Both periplakin and neurochondrin have recently been shown to interact with a handful of other GPCRs in addition to μ OPR and MCH_{1R}⁸⁹. For all of the receptors that associate with periplakin and neurochondrin, the primary functional consequence is an attenuation of G protein-mediated signaling⁸⁶⁻⁸⁹ (Fig. 2B). Since periplakin and neurochondrin exhibit discrete patterns of distribution in the brain and other tissues⁸⁹, it seems likely that they contribute to the cell-context-dependent sculpting of receptor signaling for the various receptors with which they interact. Considered together with the other GPCR partners discussed in this section, the emerging theme is that receptor-interacting proteins can exert bidirectional effects on the efficiency of G protein-mediated signaling to impart cell-specific fine-tuning of GPCR activity.

Regulation of GPCR Trafficking

GPCRs are typically trafficked to the plasma membrane to achieve functional activity. Following agonist stimulation, most receptors internalize into endosomes and may then either be targeted for lysosomal degradation or recycled back to the plasma membrane. A number of GPCR-interacting proteins have been shown to exert dramatic effects on both the biosynthetic trafficking and post-endocytic sorting of particular receptors.

Interactors that regulate biosynthetic trafficking—GPCRs must be properly folded after translation and in most cases transported to the plasma membrane to achieve functional activity. A number of GPCR-interacting proteins can regulate the folding, biosynthetic trafficking and surface expression of receptors in a cell-specific and receptor-specific manner. A classic example is the *Drosophila melanogaster* protein Nina A, which associates with rhodopsin to enhance the receptor's folding and forward trafficking^{90, 91}. Similarly, Ran-

binding protein 2 is a vertebrate homolog of Nina A that associates with vertebrate opsins to enhance their biosynthetic trafficking⁹². Another protein that regulates rhodopsin trafficking is the dynein light chain component Tctex-1, which directly associates with vertebrate rhodopsin to promote the receptor's surface trafficking^{93, 94}. Other vertebrate proteins that have been found to act as chaperones to enhance GPCR surface expression in a receptor-specific manner include GEC1^{95, 96}, RACK1⁹⁷, DRIP78^{98, 99}, ATBP50¹⁰⁰, Usp4¹⁰¹, which are listed in Table 1. For each of these GPCR-interacting proteins, their expression level in a given cell type can strongly influence the level of functional receptor expression for the particular GPCRs with which they interact.

Olfactory receptors (ORs), the largest sub-family of GPCRs, have proven notoriously difficult to study in heterologous cells (e.g., cells other than olfactory sensory neurons) due to poor trafficking to the plasma membrane¹⁰². The severe trafficking deficits of ORs in heterologous cells suggest a key role for chaperone proteins in the cell-specific control of anterograde OR trafficking in olfactory sensory neurons. Indeed, the *Caenorhabditis elegans* protein ODR-4 is expressed exclusively in chemosensory neurons, where it regulates the forward trafficking of chemosensory receptors such as ODR-10¹⁰³. In vertebrates, two unrelated families of transmembrane proteins have been shown to associate with vertebrate ORs to enhance receptor trafficking to the plasma membrane: the receptor transporting protein (RTPs) and receptor expression enhancing protein (REEPs)^{104, 105}. Certain RTP and REEP isoforms are expressed exclusively in the olfactory epithelium, where they exert cell-specific enhancement of OR trafficking¹⁰⁴. However, other RTP and REEP isoforms are more widely expressed and can interact with other GPCRs, including T2R bitter taste receptors¹⁰⁶ and opioid receptors¹⁰⁷, to enhance receptor trafficking to the plasma membrane.

Analogous to the RTPs and REEPs, two other types of single-transmembrane proteins have been shown to control the biosynthetic trafficking of particular GPCRs. V2R vomeronasal receptors had proven very difficult to study in heterologous cells until the observation that these receptors interact with M10 MHC molecules, which associate with β 2-microglobulins to strongly promote the receptors' surface expression in heterologous cells¹⁰⁸. In a similar vein, the melanocortin 2 receptor (MC2R)-associated proteins, MRAP and MRAP2, can directly interact with MC2R and dramatically enhance the receptor's surface expression¹⁰⁹⁻¹¹³. The physiological importance of MC2R-MRAP interactions *in vivo* has been well-established by studies revealing that naturally-occurring MRAP mutations cause defects in MC2R trafficking and functionality, resulting in an inherited disease known as familial glucocorticoid deficiency type 2¹⁰⁹.

Interactors that influence post-endocytic trafficking—Most GPCRs undergo significant endocytosis from the plasma membrane in response to agonist stimulation. In some cases, the receptors are recycled back to the plasma membrane, but in other cases the receptors are targeted to lysosomes for degradation⁵. GPCR internalization is heavily influenced by two of the canonical families of GPCR-interacting proteins, the GRKs and arrestins². However, certain other GPCR-interacting proteins can also regulate endocytic trafficking of GPCRs in a more receptor-selective fashion. For example, the GPCR-associated sorting proteins (GASPs) comprise a family of 10 proteins, with the founding member GASP1 originally identified in a yeast two-hybrid screen for proteins interacting with the C-terminus of the δ opioid receptor (δ OPR)¹¹⁴. Association of δ OPR with GASP1 strongly promotes receptor trafficking to lysosomes following agonist-stimulated endocytosis¹¹⁴ (Figure 3A), and has similar effects on a handful of other GPCRs, including D2 dopamine¹¹⁵ and CB1 cannabinoid receptors^{116, 117}. Most of the GASP family members are preferentially expressed in the central nervous system (CNS)¹¹⁸, suggesting that they may act in a cell-specific manner to control the post-endocytic fate (recycling versus degradation) of certain CNS-enriched GPCRs.

Beyond the members of the GASP family, a handful of other GPCR-interacting proteins have also been shown to influence the post-endocytic sorting of particular receptors. For example, sorting nexin 1 (SNX1) directly associates with the protease-activated receptor 1 (PAR1) to promote PAR1 post-endocytic trafficking to lysosomes^{119, 120}. SNX1 can also bind to the C-termini of several other GPCRs¹²¹, but how this influences the trafficking of those receptors is still unknown. In contrast to the effects of GASPs and SNX1, which associate with GPCRs to decrease receptor recycling to the plasma membrane, GPCR interactions with NHERF-1 have been found to *promote* receptor recycling following endocytosis^{19, 122-124}. As mentioned earlier, β_2 AR and κ OPR interactions with NHERF-1 mediate certain aspects of receptor signaling, while NHERF interactions with other GPCRs can modulate signaling by G proteins. Interestingly, mutant versions of β_2 AR that cannot associate with NHERF-1 are targeted much more robustly than wild-type receptors to lysosomes after agonist stimulation¹²², revealing that their interaction with NHERF-1 promotes receptor recycling (Figure 3B). Association of κ OPR with NHERF-1 also promotes receptor recycling over targeting to lysosomes¹⁹, and interaction of PTH1-R with NHERF-1 increases the plasma membrane retention of the receptor¹²⁵. Moreover, transplantation of the NHERF-1 binding motif onto the C-termini of receptors that do not normally interact with NHERF-1, such as δ OPR, dramatically enhances the efficiency of receptor recycling back to the plasma membrane following endocytosis^{123, 124}. These findings provide an example of how certain GPCR-interacting proteins, such as NHERF-1, can act as signaling intermediates, regulators of G protein signaling, *and* regulators of receptor trafficking, analogous to the multiple roles played by arrestins in the signaling and trafficking of many GPCRs².

Control of GPCR Targeting

In addition to the effects of many of the aforementioned GPCR-interacting proteins on receptor signaling and trafficking, some of these interactions also control receptor anchoring to discrete regions of the plasma membrane. For example, Homer proteins not only enhance the efficiency of mGluR1/5-mediated calcium signaling (as described above), they also facilitate the clustering and anchoring of mGluR1/5 in post-synaptic dendritic spines¹²⁶⁻¹²⁸. Consequently, mGluR1/5 receptors can be selectively localized near the post-synaptic density, positioning them to respond to glutamate that is released into the synaptic cleft. Members of the Shank family of PDZ scaffolds interact with both Homer proteins and mGluRs to further strengthen the anchoring of mGluRs to post-synaptic regions¹²⁹⁻¹³¹. The Shank family of scaffolding proteins also induce clustering of the latrotoxin-binding GPCR latrophilin-1 (also known as CL1 and C1RL-1) in heterologous cells and co-localize with latrophilin-1 at synapses in native brain tissue^{132, 133}.

A variety of PDZ scaffolds beyond the Shank proteins have also been shown to regulate GPCR clustering and anchoring in receptor-specific ways. For example, interactions of the post-synaptic density protein PSD-95 with the serotonin 5-HT_{2A} receptor induce clustering of 5-HT_{2A} in heterologous cells¹³⁴ and facilitate targeting of 5-HT_{2A} to post-synaptic dendritic compartments in cultured cortical pyramidal neurons¹³⁵. PSD-95 also associates with the C-terminus of the β_1 -AR to facilitate receptor clustering with other components of the post-synaptic density such as NMDA receptors¹³⁶. For both 5-HT_{2A} and β_1 AR, the anchoring to the plasma membrane induced by association with PSD-95, which is known to be palmitoylated and therefore tightly membrane-associated¹³⁷, greatly reduces agonist-promoted internalization of the receptors^{134, 136, 138}. In contrast, PSD-95 association with the serotonin 5-HT_{2C} receptor actually facilitates agonist-dependent internalization, while another PDZ scaffold protein, MPP3, associates with 5-HT_{2C} to prevent agonist-dependent internalization and stabilize the receptor at the cell surface in primary cortical neurons¹³⁹. As for other examples, interaction of the mGluR7a C-terminus with the PDZ scaffold PICK1 results in specific clustering of the receptor at pre-synaptic sites^{140, 141}, the α_{1D} -adrenergic receptor

interacts with syntrophins to achieve linkage to the dystrophin-utrophin complex in smooth muscle cells^{142, 143}, and the SSTR3 somatostatin receptor interacts with the PDZ protein Mupp1 to be targeted to tight junctions in epithelial cells¹⁴⁴. These examples illustrate how certain GPCR-interacting partners can selectively target GPCRs to specialized cellular compartments to promote the receptors' physiological activity.

Regulation of Ligand Binding

The vast majority of GPCR-interacting proteins described above associate with intracellular regions of the receptors and therefore have no direct effects on the pharmacological properties of the receptors, which are typically determined by the receptors' extracellular and/or transmembrane domains. However, a handful of GPCR-interacting proteins have been shown to exert striking effects on the agonist selectivity of the receptors with which they interact. The most intensively-studied examples of GPCR-interacting partners that regulate receptor pharmacology are the receptor activity modifying proteins, RAMP1, -2 and -3¹⁴⁵. The RAMP proteins were initially identified in experiments designed to search for the receptor activated by calcitonin gene-related peptide (CGRP). Surprisingly, it was found that expression of a functional CGRP receptor required co-expression of an orphan GPCR, known as the calcitonin-receptor-like receptor (CRLR, also known as CL), with a receptor-interacting partner, the single-transmembrane protein RAMP1¹⁴⁶. When CRLR was co-expressed with RAMP2, this was shown to result in the formation of receptors activated not by CGRP but rather by a related peptide known as adrenomedullin¹⁴⁶. Subsequent work has revealed that CRLR can also interact with RAMP3 to form a distinct subtype of adrenomedullin receptor^{147, 148}. Moreover, expression of the calcitonin (CT) receptor with any of the RAMPs results in the formation of receptors with unique pharmacological properties, including preferential activation of some RAMP/CT receptor combinations by a distinct peptide known as amylin^{149, 150}. It is clear from work in this area that the pharmacological properties of the CT receptor and CRLR are heavily regulated in a cell-specific fashion depending on which of the RAMP proteins is expressed in a given cell type, and furthermore the RAMP proteins can also dramatically affect the surface expression levels of the CT receptor and CRLR¹⁴⁵. Thus, a comprehensive understanding of RAMP/GPCR interactions is essential for developing therapeutics that might target the various RAMP-interacting receptors¹⁴⁵.

Conclusions and Perspectives

The signaling and trafficking of most GPCRs involves receptor interactions with G proteins, GRKs, arrestins and other receptors. In addition to these widespread canonical GPCR associations, there are also many other types of GPCR-interacting proteins that can interact with particular receptors to fine-tune receptor activity. GPCRs are important drug targets, and since it is often desirable to achieve cell-specificity of drug action in order to minimize side effects, it can be clinically useful to consider the ways in which GPCRs can be differentially modulated by therapeutics depending on cellular context. Along these lines, there has been increasing interest over the past few years in studies on "biased agonists" that can preferentially activate G protein-mediated versus arrestin-mediated GPCR signaling pathways^{151, 152}. It is worth keeping in mind that agonists are likely to be differentially-biased depending on the cellular context, which is determined largely by the set of GPCR-interacting proteins that regulate receptor signaling, trafficking and/or localization in a particular cell type. Thus, uncovering the full set of GPCR-interacting proteins for receptors of clinical interest can provide novel therapeutic insights by shedding light on the fundamental mechanisms controlling the fine-tuning of GPCR activity.

Box 1**Canonical mechanisms of GPCR signaling**

A) In the classical view of GPCR signaling, agonist binds to extracellular and/or transmembrane regions of the receptor, leading to interactions with heterotrimeric G proteins. The receptor acts as a guanine nucleotide exchange factor, catalyzing the exchange of GDP for GTP on the $G\alpha$ subunit and inducing dissociation of the $G\alpha$ and $G\beta\gamma$ subunits. Activated $GTP-G\alpha$ subunits, of which there are multiple sub-types including $G_{\alpha s}$, $G_{\alpha i}$, $G_{\alpha 12/13}$, and $G_{\alpha q}$, subsequently bind to and regulate the activity of effectors such as adenylyl cyclase, RhoGEF, or phospholipase C, thereby generating second messengers that modulate further downstream effectors. The $G\beta\gamma$ subunits can also bind to and regulate certain downstream effectors, such as ion channels and phospholipase C, following their liberation from the heterotrimeric complex.

B) G protein-mediated signaling by agonist-activated receptors can be terminated via receptor phosphorylation by G protein-coupled receptor kinases (GRKs) and concomitant receptor association with arrestins, which interact with clathrin (black shapes) and the clathrin adaptor AP2 (green circles) to drive receptor internalization into endosomes². This physiological process of receptor internalization regulates the functional process of receptor desensitization. However, in addition to attenuating G protein-mediated signaling and facilitating receptor internalization, recruitment of arrestins to activated GPCRs can also lead in many cases to the initiation of distinct arrestin-mediated signaling pathways. Following association with arrestins, GPCRs may then be trafficked to lysosomes, where they are ultimately degraded, or to recycling endosomes for recycling back to the cell surface in the functional process of resensitization. Interestingly, “biased” agonists have been recently characterized that specifically activate G protein-mediated signaling pathways over arrestin-mediated receptor signaling pathways or vice versa^{151, 152}. This new concept illustrates the importance of characterizing all GPCR downstream signaling pathways in order to more fully exploit the therapeutic potential of clinically-important receptors.

References

1. Bridges TM, Lindsley CW. G-protein-coupled receptors: from classical modes of modulation to allosteric mechanisms. *ACS Chem Biol* 2008;3:530–41. [PubMed: 18652471]
2. Reiter E, Lefkowitz RJ. GRKs and beta-arrestins: roles in receptor silencing, trafficking and signaling. *Trends Endocrinol Metab* 2006;17:159–65. [PubMed: 16595179]
3. Moore CA, Milano SK, Benovic JL. Regulation of receptor trafficking by GRKs and arrestins. *Annu Rev Physiol* 2007;69:451–82. [PubMed: 17037978]
4. Premont RT, Gainetdinov RR. Physiological roles of G protein-coupled receptor kinases and arrestins. *Annu Rev Physiol* 2007;69:511–34. [PubMed: 17305472]
5. Hanyaloglu AC, von Zastrow M. Regulation of GPCRs by endocytic membrane trafficking and its potential implications. *Annu Rev Pharmacol Toxicol* 2008;48:537–68. [PubMed: 18184106]
6. Bulenger S, Marullo S, Bouvier M. Emerging role of homo- and heterodimerization in G-protein-coupled receptor biosynthesis and maturation. *Trends Pharmacol Sci* 2005;26:131–7. [PubMed: 15749158]
7. Prinster SC, Hague C, Hall RA. Heterodimerization of g protein-coupled receptors: specificity and functional significance. *Pharmacol Rev* 2005;57:289–98. [PubMed: 16109836]
8. Milligan G. G protein-coupled receptor hetero-dimerization: contribution to pharmacology and function. *Br J Pharmacol*. 2009
9. Marrero MB, Venema VJ, Ju H, Eaton DC, Venema RC. Regulation of angiotensin II-induced JAK2 tyrosine phosphorylation: roles of SHP-1 and SHP-2. *Am J Physiol* 1998;275:C1216–23. [PubMed: 9814969]
10. Godeny MD, et al. The N-terminal SH2 domain of the tyrosine phosphatase, SHP-2, is essential for Jak2-dependent signaling via the angiotensin II type AT1 receptor. *Cell Signal* 2007;19:600–9. [PubMed: 17027227]
11. Aplin M, Christensen GL, Hansen JL. Pharmacologic perspectives of functional selectivity by the angiotensin II type 1 receptor. *Trends Cardiovasc Med* 2008;18:305–12. [PubMed: 19345318]
12. Frank GD, et al. Requirement of Ca^{2+} and PKCdelta for Janus kinase 2 activation by angiotensin II: involvement of PYK2. *Mol Endocrinol* 2002;16:367–77. [PubMed: 11818507]
13. Hunyady L, Catt KJ. Pleiotropic AT1 receptor signaling pathways mediating physiological and pathogenic actions of angiotensin II. *Mol Endocrinol* 2006;20:953–70. [PubMed: 16141358]

14. Lukashova V, Asselin C, Krolewski JJ, Rola-Pleszczynski M, Stankova J. G-protein-independent activation of Tyk2 by the platelet-activating factor receptor. *J Biol Chem* 2001;276:24113–21. [PubMed: 11309383]
15. Lukashova V, Chen Z, Duhe RJ, Rola-Pleszczynski M, Stankova J. Janus kinase 2 activation by the platelet-activating factor receptor (PAFR): roles of Tyk2 and PAFR C terminus. *J Immunol* 2003;171:3794–800. [PubMed: 14500680]
16. Dev KK. Making protein interactions druggable: targeting PDZ domains. *Nat Rev Drug Discov* 2004;3:1047–56. [PubMed: 15573103]
17. Hall RA, et al. The beta2-adrenergic receptor interacts with the Na⁺/H⁺-exchanger regulatory factor to control Na⁺/H⁺ exchange. *Nature* 1998;392:626–30. [PubMed: 9560162]
18. Hall RA, et al. A C-terminal motif found in the beta2-adrenergic receptor, P2Y1 receptor and cystic fibrosis transmembrane conductance regulator determines binding to the Na⁺/H⁺ exchanger regulatory factor family of PDZ proteins. *Proc Natl Acad Sci USA* 1998;95:8496–501. [PubMed: 9671706]
19. Li JG, Chen C, Liu-Chen LY. Ezrin-radixin-moesin-binding phosphoprotein-50/Na⁺/H⁺ exchanger regulatory factor (EBP50/NHERF) blocks U50,488H-induced down-regulation of the human kappa opioid receptor by enhancing its recycling rate. *J Biol Chem* 2002;277:27545–52. [PubMed: 12004055]
20. Huang P, et al. kappa Opioid receptor interacts with Na⁽⁺⁾/H⁽⁺⁾-exchanger regulatory factor-1/Ezrin-radixin-moesin-binding phosphoprotein-50 (NHERF-1/EBP50) to stimulate Na⁽⁺⁾/H⁽⁺⁾ exchange independent of G(i)/G(o) proteins. *J Biol Chem* 2004;279:25002–9. [PubMed: 15070904]
21. Bousquet C, et al. Direct binding of p85 to sst2 somatostatin receptor reveals a novel mechanism for inhibiting PI3K pathway. *EMBO J* 2006;25:3943–54. [PubMed: 16917505]
22. Huber A, et al. The transient receptor potential protein (Trp), a putative store-operated Ca²⁺ channel essential for phosphoinositide-mediated photoreception, forms a signaling complex with NorpA, InaC and InaD. *EMBO J* 1996;15:7036–45. [PubMed: 9003779]
23. Shieh BH, Zhu MY. Regulation of the TRP Ca²⁺ channel by INAD in *Drosophila* photoreceptors. *Neuron* 1996;16:991–8. [PubMed: 8630257]
24. Chevesich J, Kreuz AJ, Montell C. Requirement for the PDZ domain protein, INAD, for localization of the TRP store-operated channel to a signaling complex. *Neuron* 1997;18:95–105. [PubMed: 9010208]
25. Tsunoda S, et al. A multivalent PDZ-domain protein assembles signalling complexes in a G-protein-coupled cascade. *Nature* 1997;388:243–9. [PubMed: 9230432]
26. Xu XZ, Choudhury A, Li X, Montell C. Coordination of an array of signaling proteins through homo- and heteromeric interactions between PDZ domains and target proteins. *J Cell Biol* 1998;142:545–55. [PubMed: 9679151]
27. Popescu DC, Ham AJ, Shieh BH. Scaffolding protein INAD regulates deactivation of vision by promoting phosphorylation of transient receptor potential by eye protein kinase C in *Drosophila*. *J Neurosci* 2006;26:8570–7. [PubMed: 16914683]
28. Peng L, Popescu DC, Wang N, Shieh BH. Anchoring TRP to the INAD macromolecular complex requires the last 14 residues in its carboxyl terminus. *J Neurochem* 2008;104:1526–35. [PubMed: 18036153]
29. Scott K, Zuker CS. Assembly of the *Drosophila* phototransduction cascade into a signalling complex shapes elementary responses. *Nature* 1998;395:805–8. [PubMed: 9796815]
30. Mahon MJ, Donowitz M, Yun CC, Segre GV. Na⁽⁺⁾/H⁽⁺⁾ exchanger regulatory factor 2 directs parathyroid hormone 1 receptor signalling. *Nature* 2002;417:858–61. [PubMed: 12075354]
31. Sneddon WB, et al. Activation-independent parathyroid hormone receptor internalization is regulated by NHERF1 (EBP50). *J Biol Chem* 2003;278:43787–96. [PubMed: 12920119]
32. Mahon MJ, Segre GV. Stimulation by parathyroid hormone of a NHERF-1-assembled complex consisting of the parathyroid hormone I receptor, phospholipase C β , and actin increases intracellular calcium in opossum kidney cells. *J Biol Chem* 2004;279:23550–8. [PubMed: 15037630]
33. Wheeler D, et al. Regulation of parathyroid hormone type 1 receptor dynamics, traffic, and signaling by the Na⁺/H⁺ exchanger regulatory factor-1 in rat osteosarcoma ROS 17/2.8 cells. *Mol Endocrinol* 2008;22:1163–70. [PubMed: 18202147]

34. Wang B, Yang Y, Abou-Samra AB, Friedman PA. NHERF1 regulates parathyroid hormone receptor desensitization: interference with beta-arrestin binding. *Mol Pharmacol* 2009;75:1189–97. [PubMed: 19188335]
35. Oh YS, et al. NHERF2 specifically interacts with LPA2 receptor and defines the specificity and efficiency of receptor-mediated phospholipase C-beta3 activation. *Mol Cell Biol* 2004;24:5069–79. [PubMed: 15143197]
36. Fam SR, et al. P2Y1 receptor signaling is controlled by interaction with the PDZ scaffold NHERF-2. *Proc Natl Acad Sci USA* 2005;102:8042–7. [PubMed: 15901899]
37. Paquet M, et al. The PDZ scaffold NHERF-2 interacts with mGluR5 and regulates receptor activity. *J Biol Chem* 2006;281:29949–61. [PubMed: 16891310]
38. Rochdi MD, et al. Regulation of GTP-binding protein alpha q (Galpha q) signaling by the ezrin-radixin-moesin-binding phosphoprotein-50 (EBP50). *J Biol Chem* 2002;277:40751–9. [PubMed: 12193606]
39. Tang Y, et al. Association of mammalian trp4 and phospholipase C isozymes with a PDZ domain-containing protein, NHERF. *J Biol Chem* 2000;275:37559–64. [PubMed: 10980202]
40. Lee-Kwon W, Wade JB, Zhang Z, Pallone TL, Weinman EJ. Expression of TRPC4 channel protein that interacts with NHERF-2 in rat descending vasa recta. *Am J Physiol Cell Physiol* 2005;288:C942–9. [PubMed: 15590898]
41. Hwang JI, et al. Regulation of phospholipase C-beta 3 activity by Na⁺/H⁺ exchanger regulatory factor 2. *J Biol Chem* 2000;275:16632–7. [PubMed: 10748023]
42. Lee-Kwon W, et al. Ca²⁺-dependent inhibition of NHE3 requires PKC alpha which binds to E3KARP to decrease surface NHE3 containing plasma membrane complexes. *Am J Physiol Cell Physiol* 2003;285:C1527–36. [PubMed: 12954600]
43. Kunkel MT, Garcia EL, Kajimoto T, Hall RA, Newton AC. The protein scaffold NHERF-1 controls the amplitude and duration of localized protein kinase D activity. *J Biol Chem*. 2009 in press.
44. Weinman EJ, Hall RA, Friedman PA, Liu-Chen LY, Shenolikar S. The association of NHERF adaptor proteins with G protein-coupled receptors and receptor tyrosine kinases. *Annu Rev Physiol* 2006;68:491–505. [PubMed: 16460281]
45. Balasubramanian S, Fam SR, Hall RA. GABAB receptor association with the PDZ scaffold Mupp1 alters receptor stability and function. *J Biol Chem* 2007;282:4162–71. [PubMed: 17145756]
46. Guillaume JL, et al. The PDZ protein mupp1 promotes Gi coupling and signaling of the Mt1 melatonin receptor. *J Biol Chem* 2008;283:16762–71. [PubMed: 18378672]
47. Yao R, Natsume Y, Noda T. MAGI-3 is involved in the regulation of the JNK signaling pathway as a scaffold protein for frizzled and Ltap. *Oncogene* 2004;23:6023–30. [PubMed: 15195140]
48. Zhang H, Wang D, Sun H, Hall RA, Yun CC. MAGI-3 regulates LPA-induced activation of Erk and RhoA. *Cell Signal* 2007;19:261–8. [PubMed: 16904289]
49. Yamada T, Ohoka Y, Kogo M, Inagaki S. Physical and functional interactions of the lysophosphatidic acid receptors with PDZ domain-containing Rho guanine nucleotide exchange factors (RhoGEFs). *J Biol Chem* 2005;280:19358–63. [PubMed: 15755723]
50. Brakeman PR, et al. Homer: a protein that selectively binds metabotropic glutamate receptors. *Nature* 1997;386:284–8. [PubMed: 9069287]
51. Xiao B, et al. Homer regulates the association of group 1 metabotropic glutamate receptors with multivalent complexes of homer-related, synaptic proteins. *Neuron* 1998;21:707–16. [PubMed: 9808458]
52. Tu JC, et al. Homer binds a novel proline-rich motif and links group 1 metabotropic glutamate receptors with IP3 receptors. *Neuron* 1998;21:717–26. [PubMed: 9808459]
53. Kato A, et al. Novel members of the Ves1/Homer family of PDZ proteins that bind metabotropic glutamate receptors. *J Biol Chem* 1998;273:23969–75. [PubMed: 9727012]
54. Shih M, Lin F, Scott JD, Wang HY, Malbon CC. Dynamic complexes of beta2-adrenergic receptors with protein kinases and phosphatases and the role of gravin. *J Biol Chem* 1999;274:1588–95. [PubMed: 9880537]
55. Fraser ID, et al. Assembly of an A kinase-anchoring protein-beta(2)-adrenergic receptor complex facilitates receptor phosphorylation and signaling. *Curr Biol* 2000;10:409–12. [PubMed: 10753752]

56. Fan G, Shumay E, Wang H, Malbon CC. The scaffold protein gravin (cAMP-dependent protein kinase-anchoring protein 250) binds the beta 2-adrenergic receptor via the receptor cytoplasmic Arg-329 to Leu-413 domain and provides a mobile scaffold during desensitization. *J Biol Chem* 2001;276:24005–14. [PubMed: 11309381]
57. Tao J, Wang HY, Malbon CC. Protein kinase A regulates AKAP250 (gravin) scaffold binding to the beta2-adrenergic receptor. *EMBO J* 2003;22:6419–29. [PubMed: 14657015]
58. Gardner LA, Tavalin SJ, Goehring AS, Scott JD, Bahouth SW. AKAP79-mediated targeting of the cyclic AMP-dependent protein kinase to the beta1-adrenergic receptor promotes recycling and functional resensitization of the receptor. *J Biol Chem* 2006;281:33537–53. [PubMed: 16940053]
59. Kammermeier PJ, Worley PF. Homer 1a uncouples metabotropic glutamate receptor 5 from postsynaptic effectors. *Proc Natl Acad Sci USA* 2007;104:6055–60. [PubMed: 17389377]
60. Kammermeier PJ. Endogenous homer proteins regulate metabotropic glutamate receptor signaling in neurons. *J Neurosci* 2008;28:8560–7. [PubMed: 18716215]
61. Smith FD, Oxford GS, Milgram SL. Association of the D2 dopamine receptor third cytoplasmic loop with spinophilin, a protein phosphatase-1-interacting protein. *J Biol Chem* 1999;274:19894–900. [PubMed: 10391935]
62. Richman JG, et al. Agonist-regulated Interaction between alpha2-adrenergic receptors and spinophilin. *J Biol Chem* 2001;276:15003–8. [PubMed: 11154706]
63. Wang X, et al. Spinophilin regulates Ca²⁺ signalling by binding the N-terminal domain of RGS2 and the third intracellular loop of G-protein-coupled receptors. *Nat Cell Biol* 2005;7:405–11. [PubMed: 15793568]
64. Wang X, et al. Spinophilin/neurabin reciprocally regulate signaling intensity by G protein-coupled receptors. *EMBO J* 2007;26:2768–76. [PubMed: 17464283]
65. Wang Q, et al. Spinophilin blocks arrestin actions in vitro and in vivo at G protein-coupled receptors. *Science* 2004;304:1940–4. [PubMed: 15218143]
66. Snow BE, et al. GTPase activating specificity of RGS12 and binding specificity of an alternatively spliced PDZ (PSD-95/Dlg/ZO-1) domain. *J Biol Chem* 1998;273:17749–55. [PubMed: 9651375]
67. Bernstein LS, et al. RGS2 binds directly and selectively to the M1 muscarinic acetylcholine receptor third intracellular loop to modulate Gq/11alpha signaling. *J Biol Chem* 2004;279:21248–56. [PubMed: 14976183]
68. Hague C, et al. Selective inhibition of alpha1A-adrenergic receptor signaling by RGS2 association with the receptor third intracellular loop. *J Biol Chem* 2005;280:27289–95. [PubMed: 15917235]
69. Georgoussi Z, et al. Selective interactions between G protein subunits and RGS4 with the C-terminal domains of the mu- and delta-opioid receptors regulate opioid receptor signaling. *Cell Signal* 2006;18:771–82. [PubMed: 16120478]
70. Itoh M, Nagatomo K, Kubo Y, Saitoh O. Alternative splicing of RGS8 gene changes the binding property to the M1 muscarinic receptor to confer receptor type-specific Gq regulation. *J Neurochem* 2006;99:1505–16. [PubMed: 17064349]
71. Miyamoto-Matsubara M, Saitoh O, Maruyama K, Aizaki Y, Saito Y. Regulation of melanin-concentrating hormone receptor 1 signaling by RGS8 with the receptor third intracellular loop. *Cell Signal* 2008;20:2084–94. [PubMed: 18760349]
72. Neitzel KL, Hepler JR. Cellular mechanisms that determine selective RGS protein regulation of G protein-coupled receptor signaling. *Semin Cell Dev Biol* 2006;17:383–9. [PubMed: 16647283]
73. Xie GX, Palmer PP. How regulators of G protein signaling achieve selective regulation. *J Mol Biol* 2007;366:349–65. [PubMed: 17173929]
74. Minakami R, Jinnai N, Sugiyama H. Phosphorylation and calmodulin binding of the metabotropic glutamate receptor subtype 5 (mGluR5) are antagonistic in vitro. *J Biol Chem* 1997;272:20291–8. [PubMed: 9242710]
75. Lee JH, et al. Calmodulin dynamically regulates the trafficking of the metabotropic glutamate receptor mGluR5. *Proc Natl Acad Sci USA* 2008;105:12575–80. [PubMed: 18715999]
76. Nakajima Y, Yamamoto T, Nakayama T, Nakanishi S. A relationship between protein kinase C phosphorylation and calmodulin binding to the metabotropic glutamate receptor subtype 7. *J Biol Chem* 1999;274:27573–7. [PubMed: 10488094]

77. Sorensen SD, Macek TA, Cai Z, Saugstad JA, Conn PJ. Dissociation of protein kinase-mediated regulation of metabotropic glutamate receptor 7 (mGluR7) interactions with calmodulin and regulation of mGluR7 function. *Mol Pharmacol* 2002;61:1303–12. [PubMed: 12021391]
78. Turner JH, Gelasco AK, Raymond JR. Calmodulin interacts with the third intracellular loop of the serotonin 5-hydroxytryptamine1A receptor at two distinct sites: putative role in receptor phosphorylation by protein kinase C. *J Biol Chem* 2004;279:17027–37. [PubMed: 14752100]
79. Turner JH, Raymond JR. Interaction of calmodulin with the serotonin 5-hydroxytryptamine2A receptor. A putative regulator of G protein coupling and receptor phosphorylation by protein kinase C. *J Biol Chem* 2005;280:30741–50. [PubMed: 15970592]
80. Labasque M, Reiter E, Becamel C, Bockaert J, Marin P. Physical interaction of calmodulin with the 5-hydroxytryptamine2C receptor C-terminus is essential for G protein-independent, arrestin-dependent receptor signaling. *Mol Biol Cell* 2008;19:4640–50. [PubMed: 18768750]
81. Bofill-Cardona E, et al. Binding of calmodulin to the D2-dopamine receptor reduces receptor signaling by arresting the G protein activation switch. *J Biol Chem* 2000;275:32672–80. [PubMed: 10926927]
82. Liu Y, Buck DC, Macey TA, Lan H, Neve KA. Evidence that calmodulin binding to the dopamine D2 receptor enhances receptor signaling. *J Recept Signal Transduct Res* 2007;27:47–65. [PubMed: 17365509]
83. Wang D, Sadee W, Quillan JM. Calmodulin binding to G protein-coupling domain of opioid receptors. *J Biol Chem* 1999;274:22081–8. [PubMed: 10419536]
84. Nickols HH, Shah VN, Chazin WJ, Limbird LE. Calmodulin interacts with the V2 vasopressin receptor: elimination of binding to the C terminus also eliminates arginine vasopressin-stimulated elevation of intracellular calcium. *J Biol Chem* 2004;279:46969–80. [PubMed: 15319442]
85. Mahon MJ, Shimada M. Calmodulin interacts with the cytoplasmic tails of the parathyroid hormone 1 receptor and a sub-set of class b G-protein coupled receptors. *FEBS Lett* 2005;579:803–7. [PubMed: 15670850]
86. Feng GJ, et al. Selective interactions between helix VIII of the human mu-opioid receptors and the C terminus of periplakin disrupt G protein activation. *J Biol Chem* 2003;278:33400–7. [PubMed: 12810704]
87. Murdoch H, et al. Periplakin interferes with G protein activation by the melanin-concentrating hormone receptor-1 by binding to the proximal segment of the receptor C-terminal tail. *J Biol Chem* 2005;280:8208–20. [PubMed: 15590649]
88. Francke F, et al. Interaction of neurochondrin with the melanin-concentrating hormone receptor 1 interferes with G protein-coupled signal transduction but not agonist-mediated internalization. *J Biol Chem* 2006;281:32496–507. [PubMed: 16945926]
89. Ward RJ, Jenkins L, Milligan G. Selectivity and functional consequences of interactions of family A G protein-coupled receptors with neurochondrin and periplakin. *J Neurochem* 2009;109:182–92. [PubMed: 19166508]
90. Shieh BH, Stamnes MA, Seavello S, Harris GL, Zuker CS. The *ninaA* gene required for visual transduction in *Drosophila* encodes a homologue of cyclosporin A-binding protein. *Nature* 1989;338:67–70. [PubMed: 2493138]
91. Baker EK, Colley NJ, Zuker CS. The cyclophilin homolog NinaA functions as a chaperone, forming a stable complex in vivo with its protein target rhodopsin. *EMBO J* 1994;13:4886–95. [PubMed: 7957056]
92. Ferreira PA, Nakayama TA, Pak WL, Travis GH. Cyclophilin-related protein RanBP2 acts as chaperone for red/green opsin. *Nature* 1996;383:637–40. [PubMed: 8857542]
93. Tai AW, Chuang JZ, Bode C, Wolfrum U, Sung CH. Rhodopsin's carboxy-terminal cytoplasmic tail acts as a membrane receptor for cytoplasmic dynein by binding to the dynein light chain Tctex-1. *Cell* 1999;97:877–87. [PubMed: 10399916]
94. Yeh TY, Peretti D, Chuang JZ, Rodriguez-Boulant E, Sung CH. Regulatory dissociation of Tctex-1 light chain from dynein complex is essential for the apical delivery of rhodopsin. *Traffic* 2006;7:1495–502. [PubMed: 16956385]
95. Chen C, et al. GEC1 interacts with the kappa opioid receptor and enhances expression of the receptor. *J Biol Chem* 2006;281:7983–93. [PubMed: 16431922]

96. Chen Y, et al. GEC1-kappa opioid receptor binding involves hydrophobic interactions: GEC1 has chaperone-like effect. *J Biol Chem* 2009;284:1673–85. [PubMed: 19001416]
97. Parent A, Laroche G, Hamelin E, Parent JL. RACK1 regulates the cell surface expression of the G protein-coupled receptor for thromboxane A(2). *Traffic* 2008;9:394–407. [PubMed: 18088317]
98. Bermak JC, Li M, Bullock C, Zhou QY. Regulation of transport of the dopamine D1 receptor by a new membrane-associated ER protein. *Nat Cell Biol* 2001;3:492–8. [PubMed: 11331877]
99. Leclerc PC, et al. A polyaromatic caveolin-binding-like motif in the cytoplasmic tail of the type 1 receptor for angiotensin II plays an important role in receptor trafficking and signaling. *Endocrinology* 2002;143:4702–10. [PubMed: 12446598]
100. Wruck CJ, et al. Regulation of transport of the angiotensin AT2 receptor by a novel membrane-associated Golgi protein. *Arterioscler Thromb Vasc Biol* 2005;25:57–64. [PubMed: 15539617]
101. Milojevic T, et al. The ubiquitin-specific protease Usp4 regulates the cell surface level of the A2A receptor. *Mol Pharmacol* 2006;69:1083–94. [PubMed: 16339847]
102. Bush CF, Hall RA. Olfactory receptor trafficking to the plasma membrane. *Cell Mol Life Sci* 2008;65:2289–95. [PubMed: 18373063]
103. Dwyer ND, Troemel ER, Sengupta P, Bargmann CI. Odorant receptor localization to olfactory cilia is mediated by ODR-4, a novel membrane-associated protein. *Cell* 1998;93:455–66. [PubMed: 9590179]
104. Saito H, Kubota M, Roberts RW, Chi Q, Matsunami H. RTP family members induce functional expression of mammalian odorant receptors. *Cell* 2004;119:679–91. [PubMed: 15550249]
105. Zhuang H, Matsunami H. Synergism of accessory factors in functional expression of mammalian odorant receptors. *J Biol Chem* 2007;282:15284–93. [PubMed: 17387175]
106. Behrens M, et al. Members of RTP and REEP gene families influence functional bitter taste receptor expression. *J Biol Chem* 2006;281:20650–9. [PubMed: 16720576]
107. Decaillot FM, Rozenfeld R, Gupta A, Devi LA. Cell surface targeting of mu-delta opioid receptor heterodimers by RTP4. *Proc Natl Acad Sci USA* 2008;105:16045–50. [PubMed: 18836069]
108. Loconto J, et al. Functional expression of murine V2R pheromone receptors involves selective association with the M10 and M1 families of MHC class Ib molecules. *Cell* 2003;112:607–18. [PubMed: 12628182]
109. Metherell LA, et al. Mutations in MRAP, encoding a new interacting partner of the ACTH receptor, cause familial glucocorticoid deficiency type 2. *Nat Genet* 2005;37:166–70. [PubMed: 15654338]
110. Roy S, Rached M, Gallo-Payet N. Differential regulation of the human adrenocorticotropin receptor [melanocortin-2 receptor (MC2R)] by human MC2R accessory protein isoforms alpha and beta in isogenic human embryonic kidney 293 cells. *Mol Endocrinol* 2007;21:1656–69. [PubMed: 17456795]
111. Sebag JA, Hinkle PM. Melanocortin-2 receptor accessory protein MRAP forms antiparallel homodimers. *Proc Natl Acad Sci USA* 2007;104:20244–9. [PubMed: 18077336]
112. Hinkle PM, Sebag JA. Structure and function of the melanocortin2 receptor accessory protein (MRAP). *Mol Cell Endocrinol* 2009;300:25–31. [PubMed: 19028547]
113. Sebag JA, Hinkle PM. Regions of melanocortin 2 (MC2) receptor accessory protein necessary for dual topology and MC2 receptor trafficking and signaling. *J Biol Chem* 2009;284:610–8. [PubMed: 18981183]
114. Whistler JL, et al. Modulation of postendocytic sorting of G protein-coupled receptors. *Science* 2002;297:615–20. [PubMed: 12142540]
115. Bartlett SE, et al. Dopamine responsiveness is regulated by targeted sorting of D2 receptors. *Proc Natl Acad Sci USA* 2005;102:11521–6. [PubMed: 16049099]
116. Tappe-Theodor A, et al. A molecular basis of analgesic tolerance to cannabinoids. *J Neurosci* 2007;27:4165–77. [PubMed: 17428994]
117. Martini L, et al. Ligand-induced down-regulation of the cannabinoid 1 receptor is mediated by the G-protein-coupled receptor-associated sorting protein GASP1. *FASEB J* 2007;21:802–11. [PubMed: 17197383]
118. Simonin F, Karcher P, Boeuf JJ, Matifas A, Kieffer BL. Identification of a novel family of G protein-coupled receptor associated sorting proteins. *J Neurochem* 2004;89:766–75. [PubMed: 15086532]

119. Wang Y, Zhou Y, Szabo K, Haft CR, Trejo J. Down-regulation of protease-activated receptor-1 is regulated by sorting nexin 1. *Mol Biol Cell* 2002;13:1965–76. [PubMed: 12058063]
120. Gullapalli A, Wolfe BL, Griffin CT, Magnuson T, Trejo J. An essential role for SNX1 in lysosomal sorting of protease-activated receptor-1: evidence for retromer-, Hrs-, and Tsg101-independent functions of sorting nexins. *Mol Biol Cell* 2006;17:1228–38. [PubMed: 16407403]
121. Heydorn A, et al. A library of 7TM receptor C-terminal tails. Interactions with the proposed post-endocytic sorting proteins ERM-binding phosphoprotein 50 (EBP50), N-ethylmaleimide-sensitive factor (NSF), sorting nexin 1 (SNX1), and G protein-coupled receptor-associated sorting protein (GASP). *J Biol Chem* 2004;279:54291–303. [PubMed: 15452121]
122. Cao TT, Deacon HW, Reczek D, Bretscher A, von Zastrow M. A kinase-regulated PDZ-domain interaction controls endocytic sorting of the beta2-adrenergic receptor. *Nature* 1999;401:286–90. [PubMed: 10499588]
123. Gage RM, Kim KA, Cao TT, von Zastrow M. A transplantable sorting signal that is sufficient to mediate rapid recycling of G protein-coupled receptors. *J Biol Chem* 2001;276:44712–20. [PubMed: 11560936]
124. Gage RM, Matveeva EA, Whiteheart SW, von Zastrow M. Type I PDZ ligands are sufficient to promote rapid recycling of G Protein-coupled receptors independent of binding to N-ethylmaleimide-sensitive factor. *J Biol Chem* 2005;280:3305–13. [PubMed: 15548537]
125. Wang B, Bisello A, Yang Y, Romero GG, Friedman PA. NHERF1 regulates parathyroid hormone receptor membrane retention without affecting recycling. *J Biol Chem* 2007;282:36214–22. [PubMed: 17884816]
126. Ango F, et al. Dendritic and axonal targeting of type 5 metabotropic glutamate receptor is regulated by homer1 proteins and neuronal excitation. *J Neurosci* 2000;20:8710–6. [PubMed: 11102477]
127. Das SS, Banker GA. The role of protein interaction motifs in regulating the polarity and clustering of the metabotropic glutamate receptor mGluR1a. *J Neurosci* 2006;26:8115–25. [PubMed: 16885225]
128. Serge A, Fourgeaud L, Hemar A, Choquet D. Receptor activation and homer differentially control the lateral mobility of metabotropic glutamate receptor 5 in the neuronal membrane. *J Neurosci* 2002;22:3910–20. [PubMed: 12019310]
129. Tu JC, et al. Coupling of mGluR/Homer and PSD-95 complexes by the Shank family of postsynaptic density proteins. *Neuron* 1999;23:583–92. [PubMed: 10433269]
130. Sala C, et al. Regulation of dendritic spine morphology and synaptic function by Shank and Homer. *Neuron* 2001;31:115–30. [PubMed: 11498055]
131. Hayashi MK, et al. The postsynaptic density proteins Homer and Shank form a polymeric network structure. *Cell* 2009;137:159–71. [PubMed: 19345194]
132. Tobaben S, Sudhof TC, Stahl B. The G protein-coupled receptor CL1 interacts directly with proteins of the Shank family. *J Biol Chem* 2000;275:36204–10. [PubMed: 10958799]
133. Kreienkamp HJ, Zitzer H, Gundelfinger ED, Richter D, Bockers TM. The calcium-independent receptor for alpha-latrotoxin from human and rodent brains interacts with members of the ProSAP/SSTRIP/Shank family of multidomain proteins. *J Biol Chem* 2000;275:32387–90. [PubMed: 10964907]
134. Xia Z, Gray JA, Compton-Toth BA, Roth BL. A direct interaction of PSD-95 with 5-HT2A serotonin receptors regulates receptor trafficking and signal transduction. *J Biol Chem* 2003;278:21901–8. [PubMed: 12682061]
135. Xia Z, Hufeisen SJ, Gray JA, Roth BL. The PDZ-binding domain is essential for the dendritic targeting of 5-HT2A serotonin receptors in cortical pyramidal neurons in vitro. *Neuroscience* 2003;122:907–20. [PubMed: 14643760]
136. Hu LA, et al. beta 1-adrenergic receptor association with PSD-95. Inhibition of receptor internalization and facilitation of beta 1-adrenergic receptor interaction with N-methyl-D-aspartate receptors. *J Biol Chem* 2000;275:38659–66. [PubMed: 10995758]
137. Topinka JR, Bredt DS. N-terminal palmitoylation of PSD-95 regulates association with cell membranes and interaction with K⁺ channel Kv1.4. *Neuron* 1998;20:125–34. [PubMed: 9459448]

138. Xu J, et al. beta 1-adrenergic receptor association with the synaptic scaffolding protein membrane-associated guanylate kinase inverted-2 (MAGI-2). Differential regulation of receptor internalization by MAGI-2 and PSD-95. *J Biol Chem* 2001;276:41310–7. [PubMed: 11526121]
139. Gavarini S, et al. Opposite effects of PSD-95 and MPP3 PDZ proteins on serotonin 5-hydroxytryptamine_{2C} receptor desensitization and membrane stability. *Mol Biol Cell* 2006;17:4619–31. [PubMed: 16914526]
140. Boudin H, et al. Presynaptic clustering of mGluR7a requires the PICK1 PDZ domain binding site. *Neuron* 2000;28:485–97. [PubMed: 11144358]
141. Boudin H, Craig AM. Molecular determinants for PICK1 synaptic aggregation and mGluR7a receptor coclustering: role of the PDZ, coiled-coil, and acidic domains. *J Biol Chem* 2001;276:30270–6. [PubMed: 11375398]
142. Chen Z, Hague C, Hall RA, Minneman KP. Syntrophins regulate alpha_{1D}-adrenergic receptors through a PDZ domain-mediated interaction. *J Biol Chem* 2006;281:12414–20. [PubMed: 16533813]
143. Lyssand JS, et al. Blood pressure is regulated by an alpha_{1D}-adrenergic receptor/dystrophin signalosome. *J Biol Chem* 2008;283:18792–800. [PubMed: 18468998]
144. Liew CW, et al. Interaction of the human somatostatin receptor 3 with the multiple PDZ domain protein MUPP1 enables somatostatin to control permeability of epithelial tight junctions. *FEBS Lett* 2009;583:49–54. [PubMed: 19071123]
145. Hay DL, Poyner DR, Sexton PM. GPCR modulation by RAMPs. *Pharmacol Ther* 2006;109:173–97. [PubMed: 16111761]
146. McLatchie LM, et al. RAMPs regulate the transport and ligand specificity of the calcitonin-receptor-like receptor. *Nature* 1998;393:333–9. [PubMed: 9620797]
147. Fraser NJ, et al. The amino terminus of receptor activity modifying proteins is a critical determinant of glycosylation state and ligand binding of calcitonin receptor-like receptor. *Mol Pharmacol* 1999;55:1054–9. [PubMed: 10347248]
148. Hilairt S, Foord SM, Marshall FH, Bouvier M. Protein-protein interaction and not glycosylation determines the binding selectivity of heterodimers between the calcitonin receptor-like receptor and the receptor activity-modifying proteins. *J Biol Chem* 2001;276:29575–81. [PubMed: 11387328]
149. Christopoulos G, et al. Multiple amylin receptors arise from receptor activity-modifying protein interaction with the calcitonin receptor gene product. *Mol Pharmacol* 1999;56:235–42. [PubMed: 10385705]
150. Tilakaratne N, Christopoulos G, Zump E, Foord SM, Sexton PM. Amylin receptor phenotypes derived from human calcitonin receptor/RAMP coexpression exhibit pharmacological differences dependent on receptor isoform and host cell environment. *J Pharmacol Exp Ther* 2000;294:61–72. [PubMed: 10871296]
151. Urban JD, et al. Functional selectivity and classical concepts of quantitative pharmacology. *J Pharmacol Exp Ther* 2007;320:1–13. [PubMed: 16803859]
152. Violin JD, Lefkowitz RJ. Beta-arrestin-biased ligands at seven-transmembrane receptors. *Trends Pharmacol Sci* 2007;28:416–22. [PubMed: 17644195]
153. Ango F, et al. Agonist-independent activation of metabotropic glutamate receptors by the intracellular protein Homer. *Nature* 2001;411:962–5. [PubMed: 11418862]
154. Ango F, et al. Homer-dependent cell surface expression of metabotropic glutamate receptor type 5 in neurons. *Mol Cell Neurosci* 2002;20:323–9. [PubMed: 12093163]
155. Becamel C, et al. Interaction of serotonin 5-hydroxytryptamine type 2C receptors with PDZ10 of the multi-PDZ domain protein MUPP1. *J Biol Chem* 2001;276:12974–82. [PubMed: 11150294]
156. Zhang CS, et al. Knock-in mice lacking the PDZ-ligand motif of mGluR7a show impaired PKC-dependent autoinhibition of glutamate release, spatial working memory deficits, and increased susceptibility to pentylenetetrazol. *J Neurosci* 2008;28:8604–14. [PubMed: 18716219]

Figure 1.

GPCR signaling can be mediated by receptor-interacting proteins. A) Certain GPCR-interacting proteins can act as mediators of agonist-induced receptor signaling, independent of G protein- or arrestin-mediated signaling pathways. An example of this phenomenon is the interaction of the non-receptor tyrosine kinase Jak2 with the angiotensin AT1 receptor. Association of Jak2 with the phosphatase SHP-2 and stimulation of AT1 with angiotensin II together promote Jak2 association with the AT1 receptor and initiation of Jak2-dependent signaling. Activated Jak2 can phosphorylate members of the STAT family of transcription factors, leading to STAT dimerization, translocation into the nucleus, and regulation of genes controlling cell growth. B) In other cases, the agonist-dependent dissociation of an interacting protein from a GPCR can alter the activity of an intracellular signaling pathway. For example, the interaction between the p85 regulatory subunit of phosphatidylinositol 3-kinase (PI3K) and the somatostatin type II receptor (SSTR2) is disrupted by agonist stimulation, leading to reduced PI3K-mediated Akt signaling and suppression of cell survival pathways.

Figure 2.

GPCR-interacting proteins can modulate G protein-mediated signaling. In the example shown in panel A, interaction between the NHERF scaffold proteins and the parathyroid hormone receptor (PTH1R) leads to a preferential enhancement of the downstream Gαq-mediated signaling by PTH1R. Through its various protein-protein interaction domains, NHERF not only binds to PTH1R, but also tethers multiple downstream signaling effectors in close proximity to the PTH1R, thereby creating an efficient complex for Gαq-mediated signaling. However, when PTH1R is in a different cell type or separate cellular compartment in which NHERF proteins are absent, PTH1R instead preferentially signals through Gαs to activate adenylyl cyclase. A contrasting example is shown in panel B. Periplakin can associate with the μ opioid receptor (μOPR) to attenuate G protein-mediated signaling. However, in cell types or cellular compartments where periplakin is not found, μOPR ligands can more robustly activate the receptor to stimulate signaling by G proteins.

Figure 3.

GPCR-interacting proteins can regulate the post-endocytic trafficking of GPCRs. Following agonist-induced receptor endocytosis, some GPCRs are targeted for proteolytic and/or lysosomal degradation, while other GPCRs rapidly recycle back to the plasma membrane. As shown in panel A, the interaction between the GPCR-interacting protein GASP1 and the δ -opioid receptor (δ OPR) promotes the endocytic targeting of agonist-internalized δ -opioid receptors to lysosomes, where the receptors are degraded. In contrast, as shown in panel B, the interaction between the GPCR-interacting protein NHERF-1 and the β 2-adrenergic receptor (β 2-AR) promotes the rapid recycling of receptors following agonist-promoted internalization.

Interactor*	Associated GPCR	Location	Significance of Interaction	Ref. #
AKAP79	β 2-AR	CT	Tethers PKA near receptor	55, 58
AKAP250	β 2-AR	CT	Tethers PKA near receptor	54, 56, 57
ATBP50	AT2R	CT	Enhances receptor surface expression	100
Calmodulin	5-HT1A	i3L	Competes with PKC for receptor phosphorylation	78
Calmodulin	5-HT2A	i2L, CT	Impairs G protein-coupling	79
Calmodulin	5-HT2C	CT	Promotes arrestin-dependent ERK activation	80
Calmodulin	D2R	i3L	Modulates G protein signaling	81, 82
Calmodulin	mGluR5	CT	Promotes receptor surface expression	74, 75
Calmodulin	mGluR7	CT	Regulates receptor phosphorylation	76, 77
Calmodulin	PTH1R	CT	Inhibits receptor activity	85
Calmodulin	V2R	CT	Enhances receptor-induced calcium signaling	84
Calmodulin	μ OPR	i3L	Inhibits G protein-coupling	83
DRIP78	AT1R	CT	Promotes receptor surface expression	99
DRIP78	D1R	CT	Promotes receptor surface expression	98
GASP1	CB1R	CT	Targets receptor to lysosomes for degradation	116, 117
GASP1	D2R	CT	Targets receptor to lysosomes for degradation	115
GASP1	δ OPR	CT	Targets receptor to lysosomes for degradation	114
GEC1	EP3R	CT	Enhances receptor surface expression	96
GEC1	κ OPR	CT	Enhances receptor surface expression	95, 96
Homer	mGluR1	CT	Regulates receptor signaling and localization	51, 52, 127, 153
Homer	mGluR5	CT	Regulates receptor signaling and localization	51, 52, 126, 128, 153, 154
InaD	Rhodopsin	CT	Enhances speed and efficiency of receptor signaling	22-29
Jak2	AT1	CT	Promotes Jak/STAT signaling	9, 10, 12, 13
Jak2	PAF	CT	Promotes Jak/STAT signaling	14, 15
LARG	LPA2	CT	Facilitates receptor-mediated activation of Rho	49
M10 MHC	V2R		Enhances receptor surface expression	108
MAGI-3	Frizzled-4	CT	Potentiates receptor-mediated ERK/MAPK activation	47
MAGI-3	LPA ₂	CT	Potentiates receptor-mediated ERK/MAPK activation	48
MPP3	5-HT2C	CT	Inhibits agonist-induced 5-HT2C internalization	139
MRAP	MC2R	NT, TM	Promotes forward trafficking of receptor	109-113
MRAPII	MC2R	NT, TM	Promotes forward trafficking of receptor	110, 113
Mupp1	5-HT2C	CT	Induces receptor clustering	155

Interactor*	Associated GPCR	Location	Significance of Interaction	Ref. #
Mupp1	GABA _B	CT	Increases receptor stability	45
Mupp1	hSSTR3	CT	Targets receptor to epithelial tight junctions	144
Mupp1	MT ₁	CT	Enhances receptor-mediated G _{ai} signaling	46
Neurochondrin	MCH1R	CT	Disrupts G protein-mediated signaling	88
NHERF-1	PTH1R	CT	Enhances G _{aq} -mediated receptor signaling	31-34
NHERF-1	β ₂ -AR	CT	Mediates activation of Na ⁺ /H ⁺ exchange, promotes receptor recycling	17, 18, 122
NHERF-1	κOPR	CT	Mediates activation of Na ⁺ /H ⁺ exchange, promotes receptor recycling	19, 20
NHERF-2	LPA ₂	CT	Enhances G _{aq} -mediated receptor signaling	35
NHERF-2	mGluR5	CT	Prolongs receptor-mediated calcium signaling	37
NHERF-2	P2Y ₁	CT	Prolongs receptor-mediated calcium signaling	36
NHERF-2	PTH1R	CT	Enhances G _{aq} -mediated receptor signaling	30
Nina A	Rhodopsin		Promotes receptor biogenesis and trafficking	90, 91
ODR-4	ODR-10		Facilitates receptor folding and surface expression	103
p85	SSTR2	i1L, CT	Mediates survival signaling by receptor	21
PDZ-RhoGEF	LPA ₂	CT	Facilitates receptor-mediated activation of Rho	49
Periplakin	MCH1R	CT	Impairs G protein-mediated signaling	87
Periplakin	μOPR	CT	Impairs G protein-mediated signaling	86
PICK1	mGluR7a	CT	Facilitates receptor clustering in presynaptic active zones	140, 141, 156
PSD-95	5HT _{2A}	CT	Impairs receptor internalization and directs localization	134, 135
PSD-95	β ₁ -AR	CT	Attenuates agonist-promoted receptor internalization	136, 138
RACK1	TXA ₂ R	i1L, CT	Promotes forward trafficking of receptor	97
RAMP1	CRLR	NT, TM	Forms functional CGRP receptors	146
RAMP1	CT	NT, TM	Forms functional amylin receptors	149, 150
RAMP2	CRLR	NT, TM	Forms functional adrenomedullin receptors	146
RAMP3	CRLR	NT, TM	Forms functional adrenomedullin receptors	147, 148
RAMP3	CT	NT, TM	Forms functional amylin receptors	149, 150
RnBP2	Opsin		Promotes forward trafficking of receptor	92
REEP	OR		Promotes forward trafficking of receptors	104
RTP	OR		Promotes forward trafficking of receptors	104, 105
RTP/REEP	T2R		Promotes surface expression of receptors	106
RTP4	μ-δ OPR	CT	Promotes heterodimer surface expression	107
Shank	CL1	CT	Promotes receptor clustering	132

Interactor*	Associated GPCR	Location	Significance of Interaction	Ref. #
Shank	mGluR1	CT	Anchors receptor in mature dendritic spines	129
Shank	mGluR5	CT	Anchors receptor in mature dendritic spines	129
SNX1	PAR1	CT	Facilitates agonist-promoted receptor degradation	119, 120
Spinophilin	D2R	i3L	Reduces G protein and arrestin-mediated signaling	61
Spinophilin	mAChR	i3L	Reduces receptor-mediated calcium signaling	64
Spinophilin	α 2-AR	i3L	Reduces receptor-mediated calcium signaling	62-65
Syntrophin	α 1D-AR	CT	Enhances receptor stability	142, 143
Tctex-1	Rhodopsin	CT	Promotes apical delivery of receptor in polarized cells	93, 94
Usp4	A2AR	CT	Enhances receptor surface expression	101

* A comprehensive list of interactors and their associated GPCRs. Interactors are organized alphabetically, followed by associated GPCR, location of interactor association on the receptor, functional significance of interaction, and references. Blanks in the table are because location of interaction is unknown. Not included on this list are GPCR interactions with G proteins, GRKs, arrestins, RGS proteins, or other receptors. Abbreviations used in the location column: CT, carboxyl-terminus; i3L, third intracellular loop; i2L, second intracellular loop; NT, amino-terminus; TM, transmembrane; i1L, first intracellular loop.