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Perspective

Molecular Glues: A New Approach to Modulating GPCR Signaling Bias

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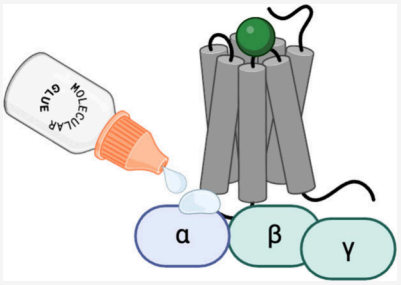
ABSTRACT: G-protein-coupled receptors (GPCRs) transmit an extracellular chemical/biological signal across the cell membrane, stimulating an array of intracellular signaling cascades. Canonically, these extracellular signaling molecules bind to the endogenous ligand pocket (orthosteric pocket), which stabilizes either an active or inactive conformational ensemble of the receptor. However, recent structural evidence indicates that small molecules can mediate the protein−protein interactions between the GPCR and their intracellular transducers. These small molecules are reminiscent of molecular glues and can be powerful tools for modulating GPCR signaling bias. In this Perspective, we will investigate the current structural information available on molecular glues and how they modulate GPCR signaling bias. We also examine the prospects of molecular glues and GPCR drug/probe design.



Widely regarded as the most functionally diverse and drugged receptor family, G-protein-coupled receptors (GPCRs) are integral membrane-bound proteins containing a bundle of seven transmembrane α-helixes (7TM) separated by both intracellular and extracellular loop regions.[1,2] They recognize various signaling molecules including photons, ions, neurotransmitters, hormones, lipids, peptides, and proteins.[1,3−6] These signaling molecules bind to the endogenous ligand pocket or the orthosteric pocket and either stabilize a set of active (i.e., full or partial agonists) or inactive (i.e., antagonists or inverse agonists) conformational states of the receptor.[6−8] Upon receptor stimulation, signal transduction primarily occurs through the recruitment and activation of heterotrimeric G-proteins and arrestins[7] (Figure 1). GPCRs also interact with various scaffolding proteins and G-protein-coupled receptor kinases (GRKs)[9,10] (Figure 1). When a drug modulates GPCR activity by binding to a site distinct from the orthosteric pocket, this is known as allosteric modulation.[8] Like orthosteric ligands, these allosteric modulators can be classified as positive (PAMs) (i.e., enhancing the activity of the ligand bound to orthosteric pocket) or negative (NAMs) (i.e., reducing the activity of the ligand in the orthosteric pocket).[8]

Recently, several groups reported small molecules bound to allosteric pockets that mediate the protein−protein interactions (PPIs) between the GPCR and either the G-alpha subunit or arrestin.[11−14] This mode of action, where a small molecule modulates PPI, is reminiscent of molecular glues. Molecular glues are often developed for targeted protein degradation (TPD), where a small molecule mediates PPIs between the target protein and other interacting partners to inactivate or degrade the target, directly.[15,16]

However, in the case of GPCRs, molecular glues work allosterically by facilitating PPIs between the receptor and transducer or can also mediate interactions between peptide-based ligands and the receptor.[16] In this Perspective we will give an overview of the current structural biology of molecular glues and how they regulate GPCR biased signaling.

STRUCTURAL AND THERAPEUTIC IMPLICATIONS FOR BIASED SIGNALING

GPCRs can promiscuously couple multiple transducers (both G-proteins and β-arrestins), activating numerous signaling pathways[17−19] (Figure 1). However, they are canonically classified by their preferred G-protein coupling pair − $G_{i/o}$, G_s, or $G_{q/11}$ (Figure 1). Signaling bias arises when a ligand stabilizes a distinct conformational state of the receptor, which is more favorable to coupling one transducer over another (i.e., it is preferential for G-protein over β-arrestin signaling)[18,20−22] (Figure 2A-B). In this sense, biased ligands act at a distance. A ligand stabilizes a specific network of interactions stemming from the orthosteric pocket, which ultimately modulates the

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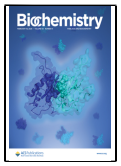

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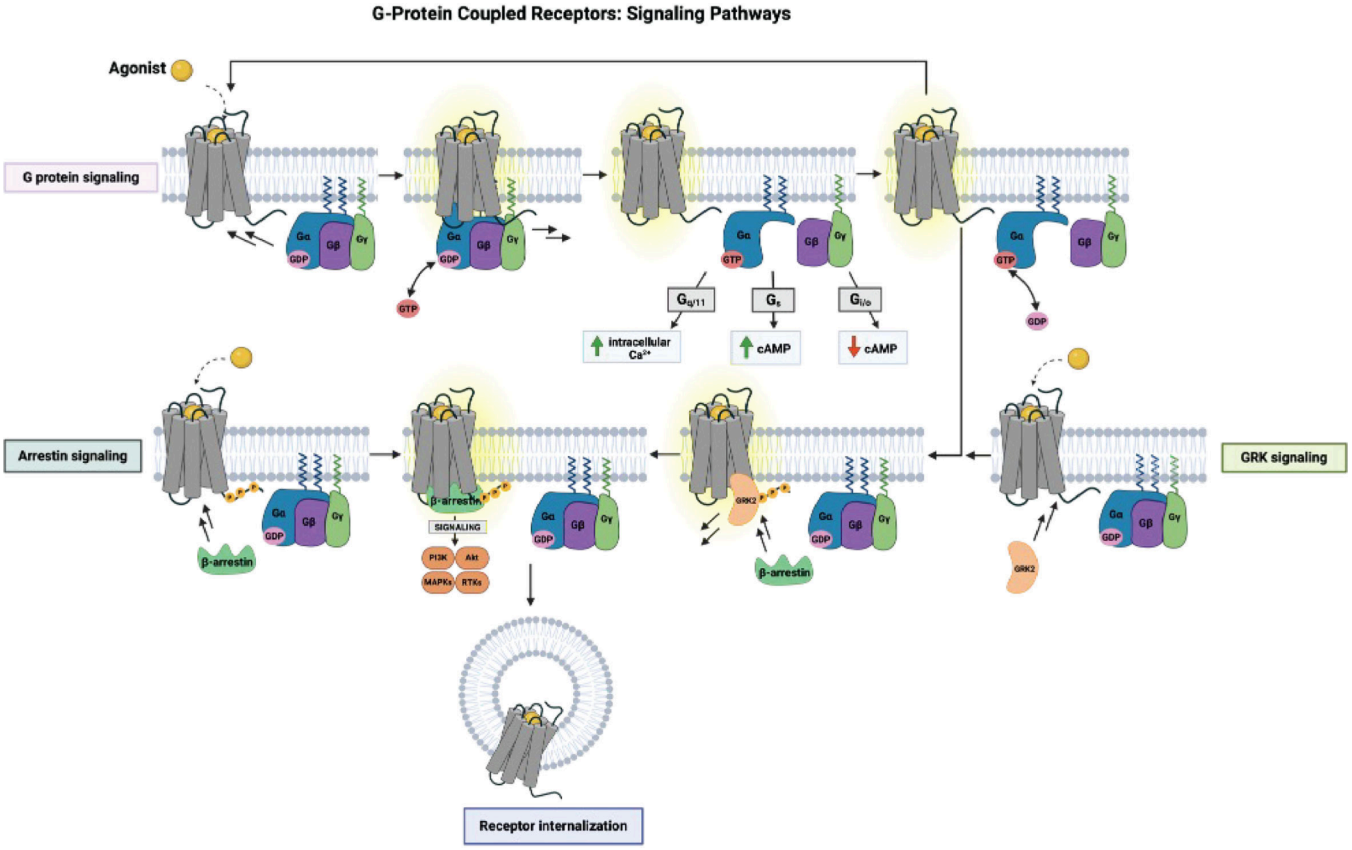

Figure 1. GPCR signaling pathways. Agonist induced signaling pathways for GPCRs. One being the typical G-protein mediated pathway which canonically signals through Gq/11, Gs, and Gi/o. Additionally once turnover has completed from the G-protein pathways, GRKs phosphorylate the receptor so then arrestin can couple. This can initiate internalization of the receptor and downstream signaling events from arrestin. Additionally, either pathway can be the preferred modality of signaling by a biased ligand.

intracellular conformations of the receptor.[20,23,24] Moreover, the opposite could also be true in which ligand independent transducer coupling to a receptor allosterically modulates orthosteric pocket conformations such that specific ligand binding is facilitated.[25,26] This concept is quite powerful in that there are existing compounds that can activate single receptor/transducer pairs, dramatically reducing unwanted side effects while precisely activating therapeutic signaling pathways.[27−29]

In theory, biased signaling is a drug hunter's dream, but it has proven challenging to implement. However, there are a handful of success stories in which a biased agonist has been implemented in a clinically relevant manner. One of these success stories is the Mu-opioid receptor G-protein biased agonist oliceridine (TPV130), which the FDA approved for human use in late 2020.[16] While efficacious in producing analgesic effects in patients having undergone bunion or abdominal surgery, the side effect profile is comparable to other approved opioids. A phase 2 clinical trial did find that patient oxygen saturation levels were higher compared to morphine, but the study was underpowered to measure respiratory depression as a clinical outcome.[30] For this reason, the FDA has restricted the use of oliceridine only to be consumed under medical supervision and not take-home prescriptions.

Another emerging success story of biased agonism comes from the 5-HT_{2C} G-protein biased agonist BMB-101. After completing Phase 1 trials with exceptional tolerability and a minimal side-effect profile, BMB-101 is currently undergoing Phase 2 trials for patients with Dravet's syndrome—an epileptic syndrome in which prolonged seizures begin within the first year of life.[31] Being a G-protein biased agonist, others have reported the potential for an improved therapeutic profile due to its minimal engagement with arrestin.[16] Arrestin signaling often leads to receptor internalization and desensitization of the drug (i.e., tolerance).[32] Additionally, we urge the reader to examine the following reviews/manuscripts for a more in-depth exploration of biased agonists in the clinic.[33−35]

Outside the clinic, many reports of biased agonists have been developed as tool compounds across many druggable GPCR targets. These targets include but are not limited to dopamine, opioid, serotonin, and many other receptors.[27,34−43] One interesting target in which signaling bias has played a central role in understanding the biological and potential therapeutic mechanisms is the 5-HT_{2A} receptor.[44−47] The 5-HT_{2A} receptor is the purported target of psychedelic drug actions and mainly couples to $G_{q/11}$ and β-arrestin2 pathways.[46] While there is minimal data for 5-HT_{2A} coupling to $G_{i/o}$ family, mainly G_z, sufficient evidence has yet to be reported in the literature of psychedelics' effects on these pathways.[48] However, it has become clear that coupling to $G_{q/11}$ is imperative to psychedelic drug actions and possibly their purported therapeutic effects.[49] Conversely, it has also been reported that β-arrestin 2 is essential in mediating LSD-induced head-twitch response—a proxy for hallucinogenic activity in mice.[50] Also, as previously mentioned, a G-protein biased ligand would theoretically not be prone to receptor

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Biochemistry 2025, 64, 749−759

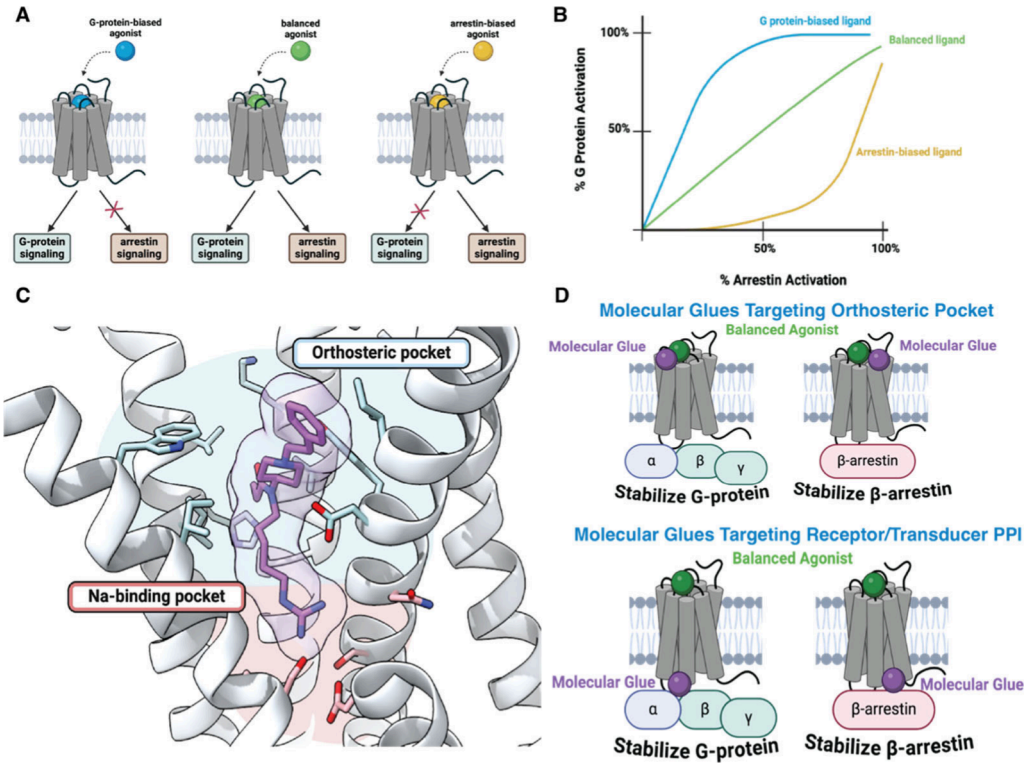

Figure 2. GPCR signaling bias and interactions with molecular glues. A. Illustration of signaling bias and how different ligands can stabilize different transducer coupled states. **B.** An example of a bias plot, which can be used to quantify signaling bias. Here the G-protein activation is plotted on the *y*-axis and the Arrestin activation is plotted on the *x*-axis with the line of unity exemplifying a perfectly balanced agonist (i.e., activates G-protein and arrestin pathways equally). **C.** An illustration of the C5-guano fentanyl bitopic targeting the Mu-opioid receptor (PDB: 7U2L) showing the engagement of the orthosteric pocket and the Na-binding site. **D.** Example of the two different ways that molecular glues can influence GPCR signaling outcomes. The top one shows the molecular glue mediating interactions between a ligand (a protein/peptide) and the receptor near the orthosteric pocket. The bottom one shows the molecular glue facilitating the PPI between the receptor and selected transducer imparting signaling bias.

internalization and rapid tolerance increases, which is a phenomenon often reported with classical psychedelics.[51] A recent study revealed several novel 5-HT_{2A} G-protein biased agonists via a large-scale *in silico* docking campaign in which three of them were also found to be selective − Q2118, Z7757, and Z2504.[37] Furthermore, the cryoEM structure of Z7757 bound to the active state complex of 5-HT_{2A} was solved, showing remarkable similarity to the predicted binding pose.

Another method to overcome the hurdle of creating biased ligands has been to simultaneously target the orthosteric pocket and Na^+ site with a bitopic ligand.[52] The Na^+ site in class A GPCRs has been extensively shown to act as a switch between inactive and active states.[52−56] At physiological concentrations, this site is occupied with Na^+, stabilizing the inactive state of the receptor. Additionally, the Na^+ site acts as an efficacy switch, potentially modulating bias between G-protein or arrestin.[54] Recently, several bitopic ligands were shown to target the orthoseric pocket and Na^+ site of the Mu-opioid receptor[52] (Figure 2C). Importantly, these bitopic ligands were found to impede arrestin recruitment compared to the orthosteric pharmacophore while maintaining the nanomolar potency for the G-protein pathways. Both structural validation via cryoEM and extensive pharmacological characterization supported these studies, which acts as a proof of concept that functionally selective ligands can be designed with fewer side effects.[52]

CHALLENGES IN STRUCTURE-GUIDED GPCR DRUG DESIGN

The advances in computational methods and structural biology have revolutionized structure-guided drug design for many therapeutically relevant GPCRs. Utilizing the vast amount of structural data now available in the PDB, researchers have gotten very good at predicting and designing molecules that can bind to a receptor.[37,43] However, the prediction of the functional outcomes (i.e., agonist/antagonist) remains woefully behind. Moreover, there are no current methods for designing biased ligands. This problem is exacerbated because many GPCRs couple with multiple transducers. In the case of the neurotensin receptor (NTSR1), it has been shown to promiscuously couple with every G-alpha subunit.[11,17] Furthermore, drug discovery campaigns often utilize the static structures deposited in the PDB, while GPCRs are dynamic, existing in an equilibrium of many conformationally relevant states.[37,57] Structural biologists and medicinal chemists regularly design compounds that interact with the orthosteric pocket and heedlessly try to target long-range network effects, pursuing fuzzy intracellular conformations. Additionally, the conformational transitions accompanying ligand binding/dissociation often lie outside of the time domain for molecular dynamics simulations to accurately sample in an unbiased fashion, leaving researchers needing a clear rationale on the structural basis of biased agonism.[58,59] Leaving this paradigm of designing molecules to target this "spooky action" at a

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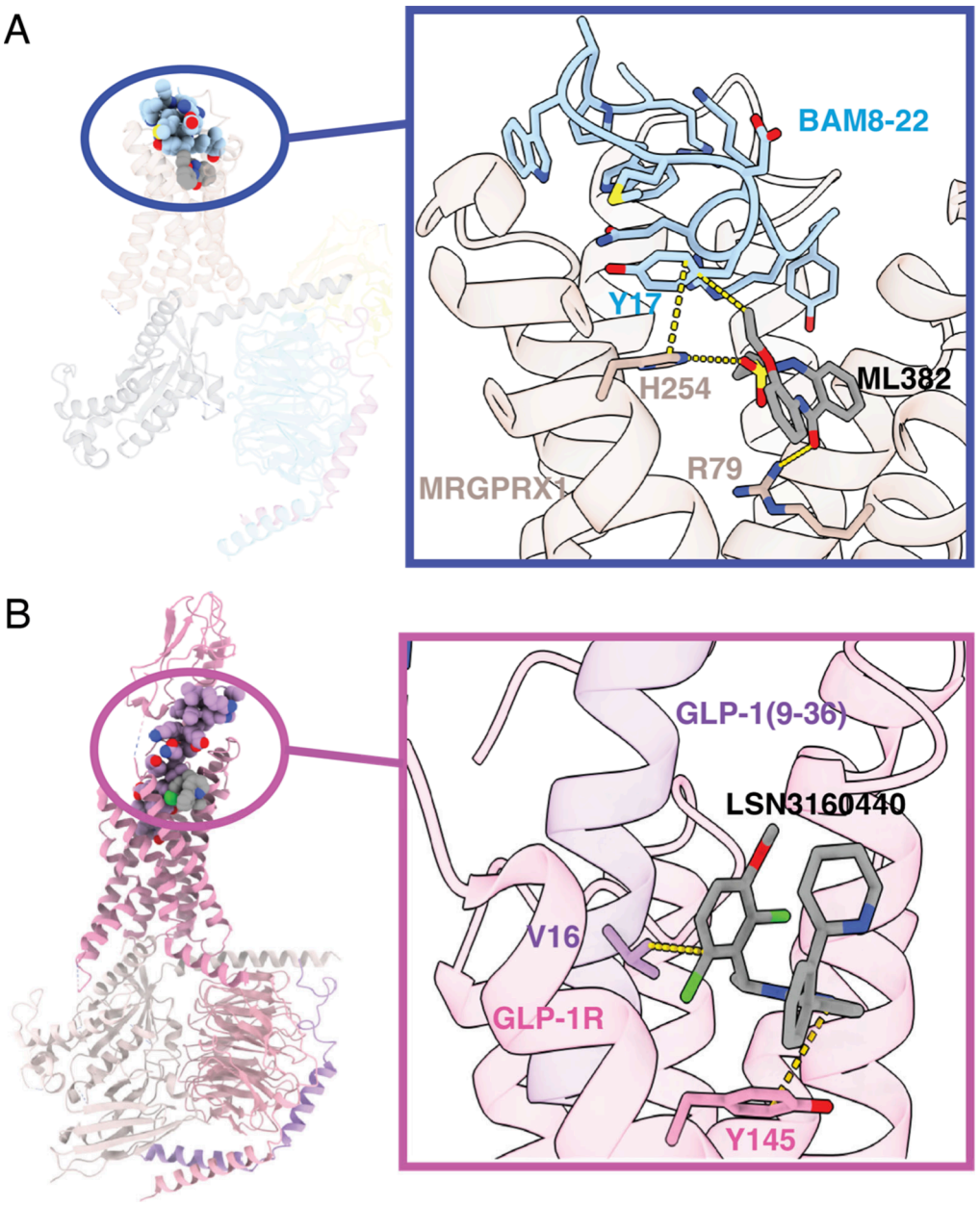

Figure 3. Molecular glues acting on the orthosteric pocket. A. MRGPRX1 (tan) in complex with the ligand BAM8−22 (blue) and the molecular glue ML382 (PDB: 8DWG). **B.** GLP-1R (pink) bound to the inactive ligand GLP-1(9−36) (purple) and the molecular glue LSN3160440 (PDB: 6VCB). LSN3160440 changes GLP-1(9−36) from a weak partial agonist to a full agonist.

distance, molecular glues that can directly target the PPIs between the receptor and transducer are highly valuable (Figure 2D).

MOLECULAR GLUES STABILIZING ORTHOSTERIC LIGANDS

There are several examples of molecular glues mediating the interaction between a protein/peptide-based ligand and the orthosteric pocket of a GPCR[60,61] (Figure 2D). Below, we will outline two examples of molecular glues mediating ligand/receptor interactions (Figure 3). However, we encourage the reader to examine the known structures in the PDB from this perspective. In both examples, the molecular glue technically acts as an allosteric modulator of an orthosteric ligand but also mediates further contact between the ligand and the receptor.

The first example is the human MAS-related G protein-coupled receptor X1 (MRGPRX1). MRGPRX1 has emerged as a novel target for treating pruritis and chronic pain.[60,62−64] It is mainly expressed in sensory neurons of the dorsal root ganglion, making it an excellent candidate for novel analgesics as activation of the receptor would avoid the potential addiction and respiratory depression observed with the current opioid treatment modalities.[63] Additionally, peripheral receptor activation may play a role in the sensation of itch. MRGPRX1 is activated by several endogenous enkephalin fragments including the bovine adrenal medulla peptide 8−22 (BAM8−22).[60] Several active state cryoEM structures were recently solved for MRGPRX1 in complex with BAM8−22 (alone) and in concert with the PAM ML382[60] (Figure 3A). These structures revealed that ML382 acts as a molecular glue between BAM8−22 and the receptor (Figure 3A). Acting as a

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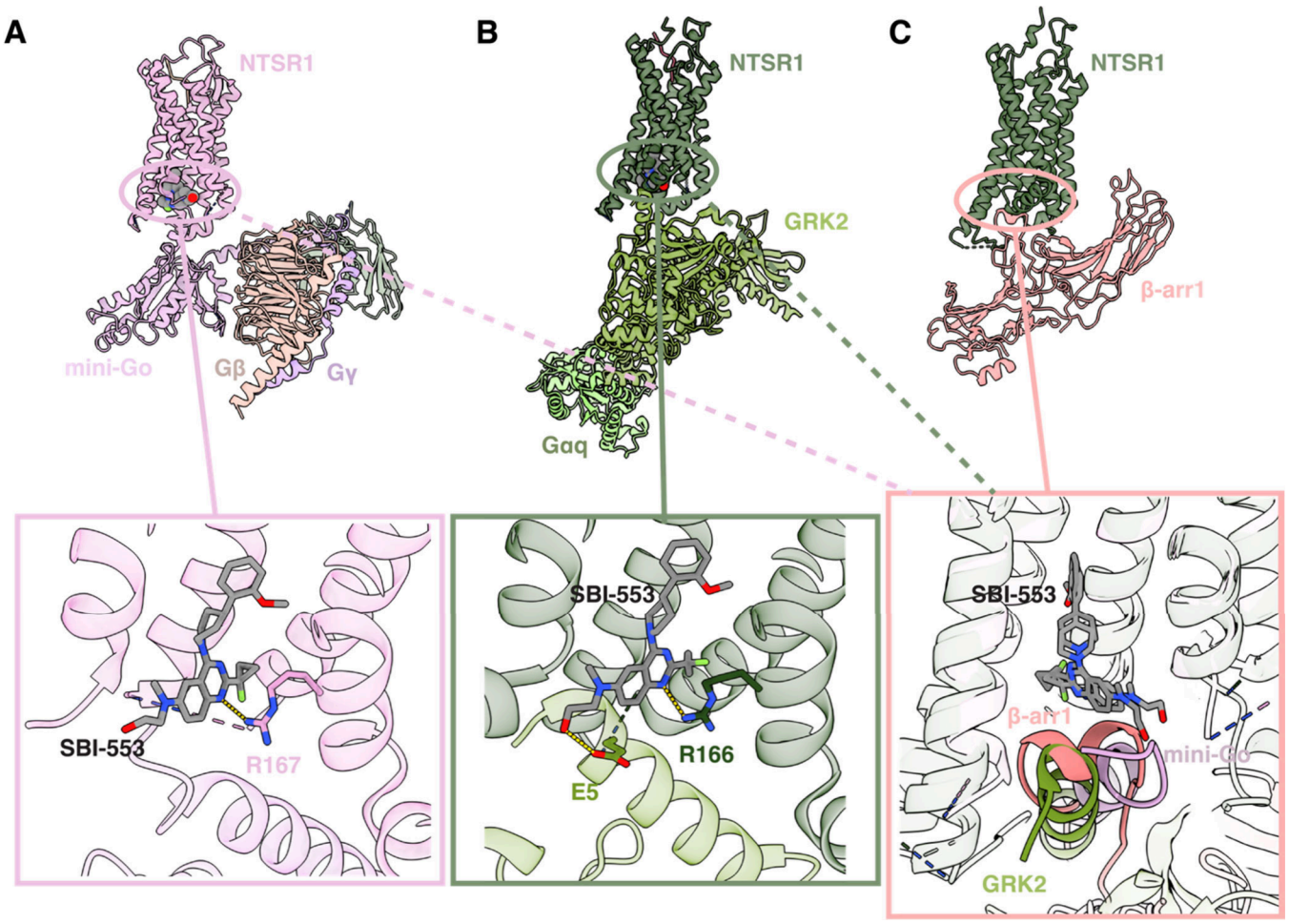

Figure 4. Structures of NTSR1 bound to SBI-553. A. The structure of NTSR1 and mini-G_o bound to SBI-553 a transducer specific molecular glue (PDB: 8FN0). **B.** The structure of NTSR1 bound to GRK/$G_{\alpha q}$ and SBI-553 (PDB: 8JPC). **C.** Top panel shows the structure of NTSR1/β-arr 1 complex (PDB: 6UP7). The bottom panel shows the overlay of all the structures of NTSR1 highlighted in this figure.

PAM, ML382 dramatically enhances the potency of BAM8−22 (~300 fold) for MRGPRX1 and exhibits strong probe dependence (it acts as a weak PAM for another agonist, compound 16). Structurally, there is no difference in the binding mode of BAM8−22 compared to the ML382/BAM8−22 complexes, but the molecular glue structurally stabilized the peptide, allowing the side chains to be better resolved. ML382 stabilizes BAM8−22 through hydrophobic and $\pi-\pi$ interactions (Figure 3A) and hydrogen bonds through its sulfonamide group and H254 while R79 interacts with the benzamide carbonyl. Also, ML382 is stabilized through $\pi-\pi$ interactions with Y82. ML382 binding is also responsible for the significant rearrangement of the ligand binding pocket, indicating the potential plasticity of MRGPRX1 and further ligand development of this molecular glue. While it is unclear if ML382 affects the signaling profile of BAM8−22 for other transducers, it is a perfect example of how molecular glues can act as an allosteric modulator mediating interactions between the orthosteric ligand and the receptor.

Another example of a molecular glue stabilizing an orthosteric ligand is the glucagon-like peptide 1 receptor (GLP-1R) PAM LSN3160440.[61] GLP-1R is the target of several FDA-approved peptides for treating type-2 diabetes and weight loss.[65−68] LSN3160440 was found through screening a diverse 220,000 compound library and is a molecular glue for the otherwise inactivated endogenous peptide GLP-1(9−36)[61] (Figure 3B). It changes GLP-1(9−36) from a weak partial agonist with an efficacy of around 3% to a full agonist and shifts its potency ~1500 fold.[61] It targets the ternary complex of GLP-1R/GLP-1(9−36) and mediates interactions between the receptor and peptide (Figure 3B). Interestingly, LSN3160440 wedges itself at the interface of TM1, TM2, and GLP-1(9−36). This ligand is prominently stabilized through hydrophobic and $\pi-\pi$ interactions (Figure 3B) but does not make any direct electrostatic/h-bonding interactions. The authors hypothesized that there are additional water-mediated interactions between the receptor and LSN3160440, which was confirmed via molecular dynamics simulations. Again, little insight is given into the modulation of various downstream transducers and their signaling profiles. However, this is a powerful example of how a molecular glue can act as an activating agent to an otherwise inactive molecule.

MOLECULAR GLUES TARGETING THE RECEPTOR/TRANSDUCER INTERFACE

The most promising use of molecular glues is in targeting/modulating the PPIs between the receptor and a specific transducer. Not until recently have several examples appeared in the literature to potently modulate these interactions and directly modify signal transduction (Figure 2D). Below we will

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Biochemistry 2025, 64, 749−759

examine the structural details of NTSR1 and SBI-553,[11,12] parathyroid hormone 1 receptor (PTHR1) and PC0371,[13] type 2 taste receptor 14 (TAS2R14) and compound28.1 (cmpd28.1),[14] and the serotonin-1A receptor (5-HT_{1A}R) and phosphatidylinositol 4-phosphate (PtdIns4P)[69] (Figures 4-6).

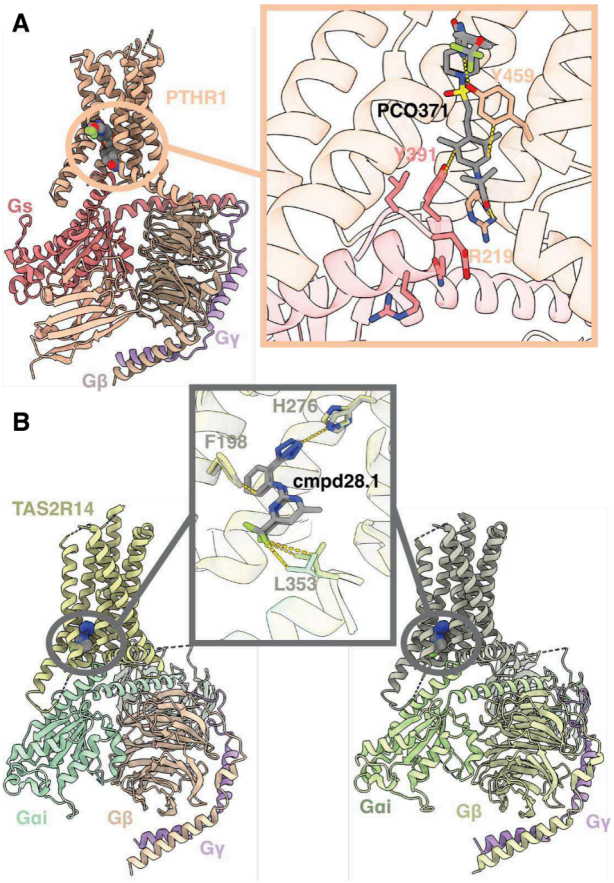

Figure 5. Structures of intracellular molecular glues. A. Structure of PTHR1 bound to the intracellular molecular glue PCO371 (PDB: 8JR9). **B.** TAS2R14 in complex with cmpd28.1 and GαI and Ggust (PDBs: 8VY7 and 8VY9).

These cases could serve as a paradigm shift and open a novel way of targeting GPCR driven signaling pathways. These examples are not meant to be an exhaustive search of the current literature but are intended to serve as an objective in which to view new and existing structural information.

As previously mentioned, NTSR1 is a Class A GPCR that has been found to promiscuously couple to every G-alpha subunit. It plays an essential role in the modulation of dopaminergic neuronal activity, regulation of food intake, and has been implicated as a target in opioid-independent analgesia.[11,12] Recently, several groups have reported the structures of the PAM agonist SBI-553[11,12] (Figure 4). SBI-553 was identified through medicinal chemistry efforts to optimize the pharmacokinetic and solubility properties of an initial parent compound, which was determined via a high-content screen.[11] In the presence of the endogenous ligand neurotensin (NTS), SBI-553 acts as a PAM-agonist for β-arrestin, a NAM for G_q and G_{15}, and is neutral for G_i and G_{12} signaling.[11] This signaling bias is a direct effect of the ligand acting as a molecular glue between the receptor and the transducer. Currently, two structures are available with SBI-553: one in complex with mini-G_o[11] (Figure 4A) and the other in complex with GRK2 and $G_{\alpha q}$[12] (Figure 4B). While the structure of NTSR1 coupled to β-arrestin 1 has been solved, this was not in the presence of SBI-553[70] (Figure 4C). Figure 4A/B shows the interactions with SBI-553 with mini-G_o and GRK2, respectively. Interestingly, comparing the mini-G_o structure to the GRK2 structure, no electrostatic interactions are found between SBI-553 and the transducer, but in the GRK2 structure, there is a clear H-bond with E5 and the hydroxyl tail of SBI-553. Aligning the two structures one notices a remarkable superposition between the ligands, while the inserting helix of each transducer is shifted slightly (Figure 4C). Overlaying the arrestin structure shows that the finger-loop of arrestin seems to span the entire region between the helices of mini-G_o and GRK2, giving an overall larger interaction area than the other transducers (Figure 4C). Additionally, some residues that may be important for interactions between the arrestin finger-loop and SBI-553 are unresolved in the current structure. Finally, R166 yields an important interaction between SBI-553 and the receptor (Figure 4A/B), which would also be recapitulated in the arrestin structure. However, it is essential to note that Krumm et al. state that the mini-G_o pocket was rearranged upon SBI-553 binding, so it would only serve to imagine that the receptor:arrestin interface would also be structurally altered for optimal binding of SBI-553.[11]

The PTHR1 receptor is a class B receptor and regulates calcium homeostasis and skeletal development via its endogenous agonists parathyroid hormone (PTH) and PTH-related peptide (PTHRP).[71] PTH or PTHRP analogs are used in the clinic to treat hypothyroidism and osteoporosis.[71] PCO371 was found in a cell-based functional screen to be a biased PAM-agonist for Gs activation while showing no recruitment of either β-arrestin 1 or β-arrestin 2.[13] Interestingly, much like SBI-553 and NTSR1, the cryoEM structure of PTHR1 bound to PCO371 showed it acting as a molecular glue mediating the contact between the receptor and G_s[13] (Figure 5A). PCO371 mediates these receptor/transducer contacts through extensive H-bonding and hydrophobic interactions with the receptor and edge-to-face $\pi-\pi$ interactions with the $G_{\alpha s}$. Without a PTHR1:arrestin structure, the reasons for signaling bias lie with the preferred interactions observed between the receptor and Gα_s.

An example from Class T GPCRs is the TAS2R14 receptor. The TAS2R family has been implicated in the recognition and perception of bitterness.[14] TAS2R14 has been associated with various extraoral physiological functions including stimulation and relaxation of airway smooth muscle.[14,72] Recently, the cryoEM structure of TAS2R14 was solved bound to both G_i and G_{gust} in the presence of a bitter tastant cmpd28.1[14] (Figure 5B). Cmpd28.1 was initially derived from modifying a known TAS2R14 agonist, flufenamic acid, a nonsteroidal anti-inflammatory drug.[14,73] Remarkably, the canonical orthosteric pocket was found to be occupied by cholesterol, while the tastant sat in an allosteric pocket between the receptor and G-protein. Interestingly, when the structures are overlaid with the different G-proteins, they are nearly identical (Figure 5B). Additionally, cmpd28.1 makes minimal hydrophobic interactions with the transducer through L353 and the trifluoro moiety, makes a singular H-bond to H276 of the receptor, and makes potential π-interactions with F198 (Figure 5B). These interactions would presumably be recapitulated with the other known agonist, flufenamic acid. Interestingly, it does not seem

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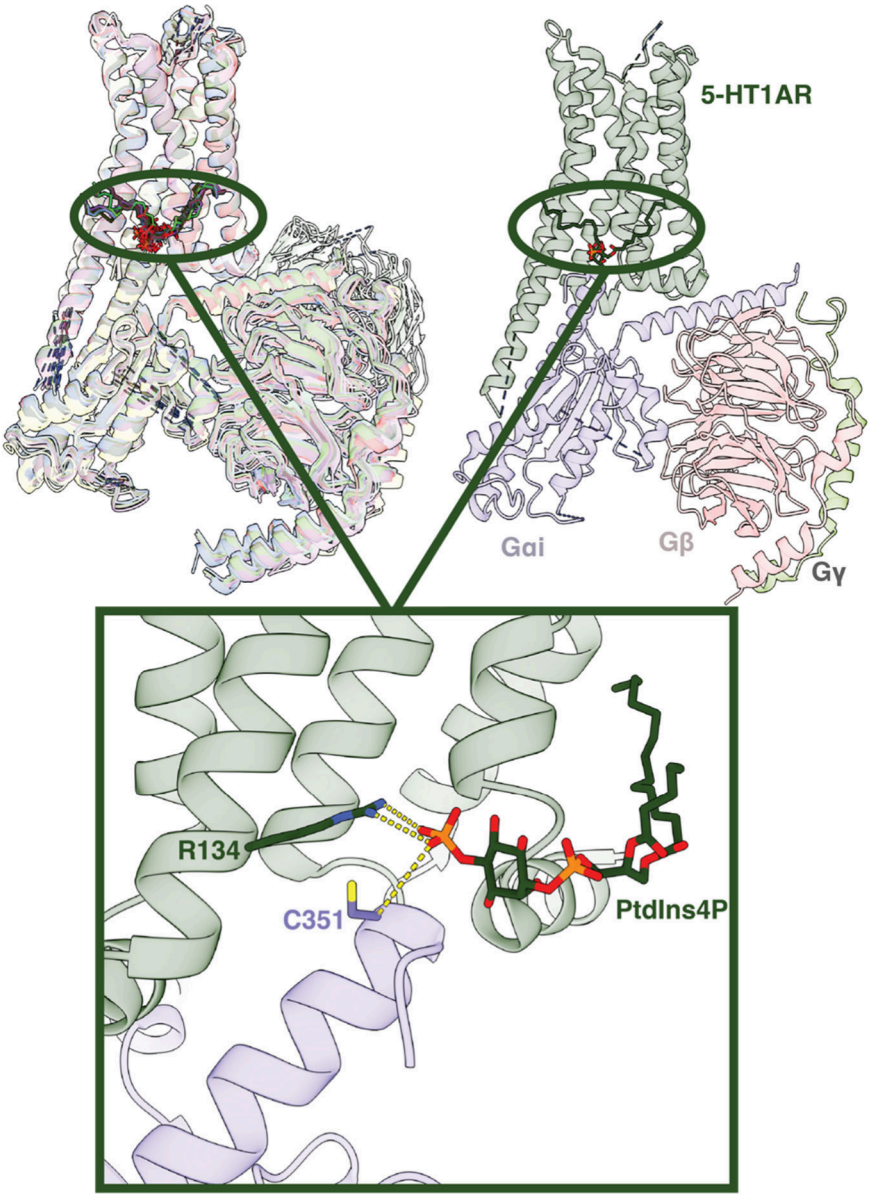

Figure 6. Structures of 5-HT1AR highlighting the intracellular molecular glue PtdIns4P. Top left panel shows the overlay of all 13 structures available of 5-HT_{1A}R in the PDB across a variety of agonists. 11 out of the 13 structures contain the PtdIns4P, while the Coulombic density in the other two is indicative of occupancy of the phospholipid at this interface. Top right panel show the 5-HT_{1A}R structure bound to serotonin and highlights PtdIns4P (PDB: 7E2Y). The bottom panel is a zoom in of the interactions mediated between the transducer and the receptor by PtdIns4P.

that cmpd28.1 shows any transducer preference for Gi or Ggust, and β-arrestin recruitment has yet to be explored. This pathway invariance could be due to the promiscuous nature of the number of bitter compounds TAS2R14 needs to recognize.

Finally, the last example returns to a Class A GPCR, the 5-HT_{1A} receptor. 5-HT_{1A} receptor agonists are widely known for their anxiolytic and antidepressant effects.[74,69] For example, buspirone is a current FDA-approved compound and is a partial agonist for the 5-HT_{1A} receptor.[75] Additionally, both partial agonists and antagonists have been shown to enhance the therapeutic effects of antidepressants in the clinic.[76] Furthermore, 5-HT_{1A} has also recently been implicated in the potential therapeutic effects of some psychedelic compounds, namely 5-methoxy-dimethyltryptamine (5-MeO−DMT), the active ingredient in the poison of the Colorado river toad (*Incilius alvarius*).[75] Several papers have been published over the past 3 years where the active state structure of the 5-HT_{1A} receptor coupled to G_i has been solved across a wide range of agonists[69,75,77,78] (Figure 6). Surprisingly, in the first structures solved, Xu et al. noted the presence of the phospholipid PtdIns4P mediating interactions between the receptor and the Gα-subunit[69] (Figure 6). Currently, 13 structures of 5-HT_{1A} are available in the PDB, with 11 containing PtdIns4P mediating the interactions between the receptor and G_i. However, both structures that do not contain PtdIns4p in their models show evidence of occupancy based on the Coulombic density in their deposited maps. All structures would indicate that the phosphate of PtdIns4p forms a salt-bridge between the conserved R134 on the receptor and makes a hydrogen bond through the backbone of C351 on Gα_i, while many other interactions are occurring between the myoinositol and the receptor (Figure 6). Additionally, Xu et al. found that the

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presence of PtdIns4p increased the rate of GTP-hydrolysis by 2.4 fold and increased the basal activity of the receptor.[69] Since this phospholipid is ubiquitously found across all the 5-HT_{1A} structures, this indicates that it plays a significant role in receptor-mediated G_i signaling, but its effects on other transducers have yet to be examined. This phospholipid acts as a molecular glue to enhance PPI interactions and could be an optimal site for small molecule design.

CONCLUDING REMARKS

Given the exciting structural revolution in GPCR chemical biology, novel methods of targeting individual signal transduction pathways are needed. While searching for allosteric modulators for GPCRs is undoubtedly not a new concept, viewing these small molecules as molecular glues yields a distinct philosophical shift in the field. Many of the biased ligands currently rely on complicated and long-range network effects stemming from the orthosteric pocket, which modulate intracellular conformations of the receptor and then lead to differences in coupling to various transducers. Given the current structural evidence across many different classes of GPCRs, much of it outlined in this Perspective, this allosteric pocket seems ubiquitous for all GPCRs. Moreover, using a molecular glue to directly modulate the signaling bias of an endogenous ligand (or even in a multidrug approach) by directly influencing PPI in this pocket is a profound paradigm shift in GPCR drug/molecular probe discovery.

Also, molecular glues can be utilized as allosteric modulators to influence PPI between peptide/protein-based orthosteric ligands and the receptor. In the case of LSN3160440, it can impart activity to an endogenous ligand (GLP-1(9−36)) which is known to be inactive.[61] Furthermore, many GPCRs are known to form homo/heterodimers, which are important to their downstream signaling events.[79] Utilizing the structural information available, one could hypothesize that molecular glues could precisely enhance these phenomena. Some evidence of this can already be found for the family C GPCR, $GABA_B$, where the PAM acts as a molecular glue to stabilize an active state TM6-TM6 dimer interface,[80−82] but it is unclear whether these ligands have a direct effect on signaling bias. However, recent evidence shows that apparent signaling bias can be achieved by targeting the dimer interface of the metabotropic glutamate receptor 3 (mGluR3), another family C GPCR.[83] GPCRs are also known to interact with many different scaffolding partners and effectors.[18] One example for which some structural information is available is receptor activity-modifying proteins (RAMPs).[84−87] A molecular glue targeting these types of interactions would yield a potentially powerful probe for understanding the basic structural biology and signaling of GPCRs but could also be an additional drug target.

Since there are now several examples in the literature of molecular glues modulating PPI to affect the signaling bias of GPCRs, *in silico* methods can now be used to screen for small molecules that target this site. Also, with DNA encoded libraries (DEL) being successfully used for GPCRs to discover allosteric modulators,[88] one could imagine a well-designed experiment that utilizes this technology to screen for novel molecular glues. However, one potential downside in targeting this intracellular site will be finding compounds that can cross the membrane, which is not the case for typical orthosteric ligands.

Overall, this shift in perspective yields a novel method for developing GPCR probes and drug discovery. Even a couple of years ago, the power to directly modulate GPCR signaling activity through modulating PPI between the receptor and transducer was not considered. With a surplus of structural information available, more examples of molecular glues and GPCRs will surely surface, adding another tool to the researcher's tool chest.

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Notes

The authors declare the following competing financial interest(s): RHG is an active consultant of 2A Biosciences.

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Biochemistry 2025, 64, 749−759