G PROTEIN-COUPLED RECEPTORS (GPCRs) comprise one of the largest families of signaling molecules and play physiologically important roles in every cell type. Thousands of GPCRs are predicted to exist. Indeed, even in the tiny worm Caenorhabditis elegans, there are predicted to be ~650 different GPCRs (10). The GPCRs are activated by a diverse array of molecules, including hormones, neurotransmitters, paracrine factors, sensory molecules, ions, cyclic nucleotides, and proteases, among others. GPCRs signal in cells by activating heterotrimeric G proteins, which consist of $\alpha\beta\gamma$-subunits. Because there are multiple genes encoding $\alpha\beta\gamma$-subunits, a large number of different heterotrimers can theoretically assemble. Once activated, the $\alpha$- and $\beta\gamma$-subunits of G proteins dissociate, and each can modulate the activity of a growing list of effectors, including enzymes such as adenyl cyclases and phospholipases, as well as ion channels (14). In contrast to the large number of GPCRs, the number of identified effectors is considerably smaller. Because many cells have multiple GPCRs that signal through common effectors, it is not surprising that cross-regulation can occur in the signaling pathways from one GPCR to another. This “cross talk” is commonly seen as an inhibition (desensitization) induced by one GPCR onto another GPCR, although stimulation (sensitization) of signaling of one GPCR by another has also been frequently observed.

Desensitization of GPCRs, or an attenuation of signaling that occurs despite the continued presence of a ligand, is a physiologically important and complex process that participates in the turning off of the GPCRs (3, 6, 12, 17, 18). Traditionally, desensitization has been divided into two forms. Homologous desensitization is a process whereby only the activated GPCRs are “turned off” or desensitized, whereas heterologous desensitization refers to processes whereby the activation of one GPCR can result in the inhibition of another heterologous GPCR to signal. It is not clear why many GPCRs exhibit desensitization while others do not or why some GPCRs may exhibit either homologous or heterologous desensitization or both. For example, in the current article in focus, Willars et al. (Ref. 24, see page C859 in this issue) have observed heterologous desensitization of bradykinin B$_2$ receptors after activation of M$_3$ muscarinic receptors while the converse did not occur, i.e., the M$_3$ receptors were not desensitized by the B$_2$ receptors. Different cellular backgrounds can also influence the type and degree of desensitization that is observed, suggesting that multiple events are required for the different processes.

What are the molecular events underlying desensitization of GPCRs? Homologous desensitization has been extensively studied, and an important hallmark of this process is that it is exquisitely dependent on agonist occupancy of the GPCRs. Agonist-induced phosphorylation of GPCRs is an early event that is important in the process of homologous desensitization (3, 5, 17, 18). The processes underlying heterologous or cross-desensitization are less well understood, and it appears that there will be multiple mechanisms contributing to this type of inhibition. GPCRs that undergo heterologous desensitization do not need to be occupied by an agonist. Rather, the activation of one GPCR generates a signal that causes inhibition of signaling by a second, heterologous GPCR. This type of desensitization is due to cross talk in signaling pathways that involves modifications of the activities of GPCRs, G proteins, or effectors (8, 9, 12).

Because phosphorylation of the GPCRs plays a central role in homologous desensitization, many have asked whether receptor phosphorylation might play a role in heterologous desensitization. Activation of second messenger-dependent protein kinases, such as protein kinase A and protein kinase C (PKC), by one GPCR can result in the phosphorylation of other GPCRs in the same cell and result in heterologous desensitization (9, 12). Indeed, many GPCRs are phosphorylated by PKC, and an illustration of the role of PKC in cross-desensitization is provided from studies of chemoattractant receptors (1). Notably, PKC-mediated phosphorylation of platelet-activating factor (PAF) receptors is induced by activation of other chemoattractant receptors and inhibits both coupling of the PAF receptors to G proteins and mobilization of intracellular calcium stores (20). Interestingly, the cross-regulation is unidirectional, because activation of wild-type PAF receptors does not cross-regulate the chemoattractant receptors that induced the cross-regulation of the PAF receptors (20).
A unidirectional regulation of GPCRs was also found in the study by Willars et al. (24); however, the consequence of phosphorylation by PKC was markedly different. Willars et al. demonstrate that activation of bradykinin B$_2$ receptors induced a PKC-dependent phosphorylation of the M$_3$ muscarinic receptors. However, this phosphorylation did not appear to alter the ability of the M$_3$ receptors to increase phosphoinositide or calcium signaling. On the other hand, activation of the M$_3$ receptors did not induce phosphorylation of the bradykinin B$_2$ receptors but did induce cross-inhibition of phosphoinositide and calcium signaling. Thus these data provide an example where the cross-regulation of receptor signaling did not appear to be linked to receptor cross-phosphorylation, whereas, in other cases, such as that cited above for the PAF receptors (1, 20), receptor phosphorylation plays an obligatory role in cross-desensitization. It is likely that multiple events participate in heterologous regulation of GPCRs. It is becoming increasingly clear that receptor phosphorylation is not the exclusive mediator of heterologous desensitization (1, 8) and that events downstream are clearly involved. Although the exact nature of these downstream events is not known in detail, many have suggested the involvement of, and/or modification of, G proteins, downstream effectors, and/or stores of small signaling molecules (1, 8, 9, 12).

What else might be contributing to cross-desensitization of GPCRs? In recent years, the identification and characterization of the growing family of regulators of G protein signaling (RGS) proteins has added another player to desensitization pathways (2, 11). Because RGS proteins have been demonstrated to be GTPase-activating proteins (GAPs) that accelerate the turnover of activated G proteins, it is likely that RGS proteins contribute to desensitization. The role of RGS proteins in the regulation of GPCRs is under intensive study, and we are certain to learn more about how they participate in desensitization. Of interest, there are indications that the inhibition of activated G$_{\alpha}$-subunits by RGS proteins might allow for enhanced expression of G$_{\beta\gamma}$-dependent pathways (4, 8, 16). Interestingly, certain effectors such as phospholipase C-$\beta$1 possess GAP activity, and this can be blocked by G$_{\beta\gamma}$-subunits (7). It remains to be determined how GAPs contribute to cross-regulation.

In addition, previously unappreciated inhibitory signaling mechanisms are becoming more obvious contributors to heterologous desensitization. For example, it is possible that cross-desensitization might result when two GPCRs regulate a common effector by different signaling pathways. Recent studies of the regulation of voltage-dependent calcium channels by GPCRs have suggested this possibility (19). Potentially, the ability of one GPCR to regulate one effector may be more dominant and preclude the ability of another GPCR to exert its effect. Another area of developing interest is the role of lipids or lipid-derived signaling molecules. Arachidonic acid production has been linked to the ability of endothelin A receptors to heterologously inhibit the ability of $\mu$-opiate receptors to stimulate G protein-activated inwardly rectifying potassium channels (GIRKs) (21). This is interesting because the endothelin receptors by themselves do not appear to exert effects on the GIRK channels (21).

Other mechanisms of GPCR regulation also may involve lipid-dependent regulation. The findings that arrestin-dependent internalization of GPCRs may possess a requirement for phosphoinositides (13), and that agonist-dependent internalization of GPCRs may be modulated by depletion of phosphoinositides (22), both suggest that the status of lipid stores may influence the desensitization of GPCRs. In addition, there appears to be a requirement for phosphoinositides in the GPCR-dependent regulation of GIRK channels (15, 23). Consequently, depletion of phosphatidylinositol 4,5-bisphosphate stores by GPCR-mediated activation of phospholipases could contribute to heterologous desensitization of GIRKs. Future studies should reveal other previously unrecognized signaling events that have roles in the desensitization of responses elicited by GPCRs. From what we know so far, it is likely that the molecular events underlying cross-regulation of GPCRs will be complex and that a universal mechanism will not be found responsible.

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