Interaction of Phosducin-like Protein with G Protein βγ Subunits*

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Phosducin-like protein (PhLP), a widely expressed ethanol-responsive gene (Miles, M. F., Barhite, S., Sanga, M., and Elliott, M. (1993) Proc. Natl. Acad. Sci. U. S. A. 90, 10831–10835), is a homologue of phosducin, a known major regulator of Gβγ signaling in retina and pineal gland. However, although phosducin has a well characterized role in retinal phototransduction, function of the PhLP remains unclear. In this study we examine the ability of PhLP to bind Gβγ dimer in vitro and in vivo. Using PhLP glutathione S-transferase fusion proteins, we show that PhLP directly binds Gβγ in vitro. Studies with a series of truncated PhLP fusion proteins indicate independent binding of Gβγ to both the amino- and C-terminal halves of PhLP. Protein-protein interactions between Gβγ and PhLP are inhibited by the α subunit of Ga and Gγ, suggesting that PhLP can bind only free Gβγ. Finally, we show that PhLP complexizes, at least partially, with Gβγ in vivo. Following overexpression of epitope-tagged PhLP together with Gβγ2a proteins in COS-7 cells, a PhLP-Gβγ complex is co-immunoprecipitated by monoclonal antibody directed against the epitope tag. Similarly, polyclonal anti-PhLP antibody co-precipitates endogenous PhLP and Gβγ proteins from NG108-15 cell lysates. These data are consistent with the hypothesis that PhLP is a widely expressed modulator of Gβγ function. Furthermore, because alternate forms of the PhLP transcript are expressed, there may be functional implications for the existence of two Gβγ-binding domains on PhLP.

Heterotrimeric guanine nucleotide-binding proteins (G proteins) play a major role in transmembrane signaling processes by transducing extracellular signals from the superfamily of heptahelical cell surface receptors to their appropriate intracellular effectors (1, 2). In its trimeric form, Gaγβδ is inactive, and the Ga subunit binds a molecule of GDP. Upon ligand binding, the receptor catalyzes the exchange of GDP for GTP on Ga that causes its activation and dissociation from the tightly bound Gβγ complex. Inactivation and reassociation of the heterotrimer is initiated by the hydrolysis of bound GTP into GDP by an intrinsic GT-Pase activity of the Ga subunit. It is now known that both the free GTP-bound Ga and the Gβγ dimer can bind and regulate downstream effectors including adenyl cyclases, phospholipases, and ion channels, and thereby modulate second messenger levels and ion flux (3).

The discovery of several specific Gβγ binding proteins has recently shed light on new roles for Gβγ in the propagation and termination of cellular signaling. The dimer has been shown to recruit β-adrenergic receptor kinase (βARK)2 to its membrane-associated receptor substrate and thus initiate receptor desensitization (4, 5). This process occurs via direct binding of Gβγ to the C terminus of a putative pleckstrin homology domain on βARK (6). Furthermore, the responsiveness of G protein-regulated signaling systems may be directly modulated through the interaction of Gβγ subunits with intracellular regulatory proteins. For instance, phosducin, a phosphoprotein mainly expressed in the retina and pineal gland, inhibits the phototransduction cascade by scavenging Gβγ subunits of the G protein transducin (GαT), thus preventing their reassociation with the GαT subunit (7, 8). Because phosducin has a higher affinity for Gβγ than does GαT, it has been suggested that the formation of the phosducin/Gβγ complex is a major factor regulating photoreceptor responsiveness (9). From in vitro binding and cotransfection assays, it was proposed that phosducin may also compete with other targets for Gβγ binding, such as βARK and phospholipase C type β2 (10, 11).

We recently isolated a rat brain cDNA encoding a phosducin-like protein (PhLP), which has 65% amino acid homology to phosducin (12). We also described several 5’-end splice variants that generate two predicted isoforms of the protein: PhLP long (PhLP) of 301 amino acids containing the entire coding sequence and PhLP short (PhLPS) of 218 amino acids missing the first 83 N-terminal residues of PhLP (12, 13). Based on sequence and PhLP short (PhLPS) of 218 amino acids missing the first 83 N-terminal residues of PhLP (12, 13). Based on sequence, a two-domain Gβγ-binding domain. To more directly characterize the interaction of PhLP with Gβγ, we studied PhLP binding to Gβγ both in vivo and in vitro. Our results here, using in vitro binding studies with a series of truncated PhLP glutathione S-transferase (GST) fusion proteins, show that PhLP binds Gβγ through a bipartite binding domain. The Gβγ-PhLP interaction was confirmed by co-immunoprecipitation of the complex from cell lysates. Our findings support the hypothesis that PhLP can modulate Gβγ function in signaling in nonretinal tissues. In favor of this hypothesis, a recent report showed that recombinant PhLPγ inhibits several Gβγ functions in vitro (14). Interestingly, these authors suggested that unlike phosducin (11, 15), the N terminus of PhLP was unlikely to contain a Gβγ-binding domain.

1 Gβγ are thought to exist as an obligate complex and are thus referred to here as Gβγ even in instances where direct interaction may only involve Gβ.

2 The abbreviations used are: βARK, β-adrenergic receptor kinase; DMEM, Dulbecco’s modified Eagle’s medium; GST, glutathione S-transferase; HA, hemagglutinin; PhLP, phosducin-like protein; PhLPγ, phosducin-like protein, short form (PhLP12253–301); PBS, phosphate-buffered saline; PAGE, polyacrylamide gel electrophoresis.

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(14) in many tissues through direct protein-protein interactions. Regulation of PhLP/Gβγ interactions could be an important factor in controlling G protein signaling.

**EXPERIMENTAL PROCEDURES**

**Materials**—PhLP and PhLPγ cDNAs were cloned in our laboratory (12). The GST-phosducin construct was obtained from Dr. Cheryl Burg (University of Southern California). Gβγ and Gα proteins purified from bovine brain were kindly provided by Dr. Eva Neer (Brighman and Woman’s Hospital, Boston, MA). Gαβγ and Gγ2 expression vectors were kindly gifts from Dr. H. Bourne (University of California at San Francisco). Purified recombinant Gαβγ2 was generously provided by Dr. Rene Or Lustre in the Bourne laboratory. Recombinant Gα protein was from Calbiochem.

**DNA Constructs**—The full-length PhLP (amino acids 1–310) as well as different regions of the protein corresponding to amino acid residues 84–301 (referred to as PhLPγα), 1–167, 1–115, 1–70, 1–301, 161–301, and 200–301 were expressed as GST fusion proteins. DNA fragments encoding PhLP and its derivatives were amplified by polymerase chain reaction using rat PhLP cDNA as template and 5’- and 3’-primers containing BamHI and EcoRI sites, respectively. The amplified fragments were ligated in frame with the 5’ end of the coding region of GST into BamHI and EcoRI sites of the pHexas His-2 vector (Pharmacia Biotech Inc.). The resultant constructs were verified by DNA sequencing using the chain termination method (Sequenase version 2.0, U.S. Biochemical Corp.) and used to transform Escherichia coli strain BL21.

An epitope-tagged PhLP expression vector was generated by fusing an 8 amino acid peptide from the hemagglutinin (HA) of influenza virus to the C terminus of PhLP. Sense and antisense oligonucleotides corresponding to the HA epitope (VDVPDYAS) were synthesized on a 5’ EcoRI site and a 3’ NotI site, were synthesized, annealed, and inserted into pCDNA3 (Invitrogen) by the Sarkosyl method (16). In a typical binding assay, following immobilization on glutathione-Sepharose beads, fusion proteins at a final concentration of 0.5–1.0 μM were incubated with 50–100 nM Gβγ purified from bovine brain in 50 μl of phosphate-buffered saline (PBS) containing 0.01% Lubrol for 2 h at 4 °C. Following six washes in 200 μl of PBS containing 0.1% Lubrol, the beads were resuspended in SDS sample buffer and boiled for 10 min. The eluted proteins were separated with 10% SDS–polyacrylamide gel electrophoresis (PAGE) and electrotransferred onto nitrocellulose membranes using standard methods. The blots were probed with a polyclonal anti-α antisera (1:1000; DuPont NEN) and processed using the enhanced chemiluminescent immunonosense detection system (Amersham Corp.). Occasionally, blots were stripped and reprobed with a polyclonal anti-GST antisera (1:4000; Santa Cruz).

**Anti-PhLP Antiserum**—The entire coding region of PhLPγ cDNA was amplified by polymerase chain reaction and fused in frame with the maltose binding protein coding region in the vector pMal-c2 (New England Biolabs). The maltose binding protein-PhLPγ fusion protein migrated at approximately 72 kDa on SDS-polyacrylamide gels as expected. The fusion protein was purified to apparent homogeneity by amylose resin chromatography exactly as described by the manufacturer (New England Biolabs) and was injected into rabbits by a commercial source (CalTag) for generation of a polyclonal antiserum. This antiserum was affinity-purified over a column of GST-PhLPγ coupled to CaBr2-activated Sepharose 4B (Pharmacia).

**Cell Culture and Transient DNA Transfection**—NG108-15 cells were grown as described (12) in Dulbecco’s modified Eagle’s medium (DMEM) containing 10% fetal bovine serum (JRH Biosciences). COS-7 cells (3 × 10^6 cells/well) were seeded 48 h before transfection in 6-well plates in DMEM supplemented with 10% fetal bovine serum. Cells were incubated for 5 h in serum-free DMEM with DNA plasmids premixed with lipofectAmine (Life Technologies, Inc.) and then infected over night at 37 °C in DMEM containing 10% fetal bovine serum. The total amount of DNA in all transfections was 2 μg/well. When required, the empty pCDNA3 vector was used to maintain a constant amount of DNA.

**Immunoprecipitation**—Two × 10^6 transfected COS-7 cells or 4 × 10^6 NG108-15 cells were washed twice with ice-cold PBS and lysed in Nonidet P-40 lysis buffer (50 mM Tris-HCl, pH 8.0, 150 mM NaCl, 1%}

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**FIG. 1.** Binding of bovine brain Gβγ by GST-phosducin and GST-PhLP. A, Western blot (upper panel) analysis of Gβγ binding to GST fusion proteins. GST fusion proteins (500 nM) or GST (1.5 μM) were incubated with 70 nM of purified bovine brain Gβγ. Bound Gβγ was fractionated by SDS-PAGE and visualized by Western blot using a polyclonal anti-β antisera as described under “Experimental Procedures.” Lane 1, GST; lane 2, GST-PhLPγ; lane 3, GST-PhLP; lane 4, GST-phosducin. Also shown is 500 ng of purified Gγ alone. The lower panel shows Coomassie Blue staining of expressed GST fusion proteins following purification on glutathione-Sepharose resin. Approximate molecular masses (in kDa) are indicated at the right. B, titration of Gβγ binding to GST-PhLP (left) and GST-phosducin (right). GST fusion proteins were present at 1 μM in all reactions, and increasing concentrations of Gβγ were added as indicated. The amounts of GST fusion proteins and bound Gβγ were monitored by Western blot using anti-GST (lower panels) and anti-β (upper panels) antisera, respectively. The results are representative of experiments repeated at least three times.

Nonidet P-40, 2 μg/ml aprotinin and leupeptin, 20 μg/ml soybean trypsin inhibitor). After 15 min of incubation on ice, insoluble material was removed by centrifugation at 10,000 × g for 4°C for 10 min, and the lysate was precleared in the presence of protein A-agarose (Santa Cruz) for 30 min. COS-7 cell lysate was then incubated with 5 μg of monoclonal antibody 12CA5 (Boehringer Mannheim) to the HA tag, whereas NG108-15 cell lysate was incubated with 5 μg of affinity-purified polyclonal anti-PhLP or an equivalent amount of preimmune serum. The incubations were conducted overnight at 4°C before precipitation in the presence of protein A-agarose. The immunoprecipitates were washed four times with PBS containing 0.1% Lubrol, and protein complexes were eluted in SDS sample buffer and analyzed by Western blot using the 12CA5 antibody and/or a monoclonal antibody to the Gβ subunit (Transduction Laboratories).

**RESULTS AND DISCUSSION**

To determine whether PhLP directly interacts with Gβγ, we generated PhLP and PhLPγ GST fusion proteins and examined their ability to bind Gβγ purified from bovine brain. Following immobilization of the fusion proteins on glutathione-agarose beads, Gβγ binding was detected by Western blot analysis using an anti-β antisera. As controls, we tested Gβγ binding by the GST protein itself and GST fused to phosducin (GST-phosducin), a known Gβγ-binding protein. The fusion proteins migrate at their expected molecular weight as visualized by Coomassie Blue-staining (Fig. 1A, lower panel) and can be detected with an anti-GST antibody on Western blot analysis (Fig. 1B, lower panel). In addition, GST-PhLPγ proteins are recognized by an affinity-purified polyclonal antisera directed against PhLPγ (data not shown).

As previously reported by other investigators (11), GST-
phosphducin bound detectable amounts of Gβγ at a molar ratio of βγ:GST-phosphducin of approximately 1:7 (Fig. 1A, upper panel). Under similar conditions, GST-PhLP and GST-PhLP also retained Gβγ (Fig. 1A, upper panel). By contrast, GST protein did not bind Gβγ subunits even when present at a 3-fold higher concentration than the GST-PhLP proteins (Fig. 1A, upper panel), indicating that Gβγ binding by the fusion proteins is specified by the PhLP protein sequence. The affinity of phosphducin for Gβγ was previously reported to be in the nanomolar range (11, 14). Under our experimental conditions, GST-PhLP appeared to have a slightly lower affinity for Gβγ than GST-phosphducin but of the same order of magnitude because titration with varying amounts of Gβγ protein showed that GST-PhLP retained only slightly lower amounts of Gβγ than did GST-phosphducin (Fig. 1B, upper panel).

To map the Gβγ binding domain of PhLP, we examined recombinant Gγ1γ2 interaction in vitro with GST fusion proteins containing various regions of PhLP. Fig. 2 shows an alignment of PhLP deletion constructs with the full-length PhLP. Each construct produced a protein that migrated at the expected molecular weight on SDS-PAGE (data not shown). Surprisingly, we found Gβγ binding activity of PhLP at two areas in the N- and C-terminal regions. In the N-terminal half of PhLP (PhLP1–167), amino acids 50–115 appeared sufficient for Gβγ binding, because PhLP50–115 and PhLP50–167 retained Gβγ, whereas PhLP1–50 and PhLP168–301 did not (Fig. 2). The 50–150 region contains an 11-amino acid stretch (57–67: TGPKGVINDWR) that is perfectly conserved between PhLP and phosphducin (11). Furthermore, the crystal structure of the Gγ1γ2-phosphducin complex, reported while this manuscript was in preparation, showed that this highly conserved sequence has extensive and tight interactions with the center of the Gγ propeller (17). This conserved sequence region of PhLP (amino acids 57–67) may be important for binding of Gγ1γ2 because PhLP84–301 totally lacked βγ binding, whereas PhLP50–167 had essentially full binding activity. However, although the 57–67 region may be important for Gβγ binding by PhLP, additional elements seem required because PhLP1–70 did not bind Gβγ.

Additional deletions revealed that the C-terminal half of PhLP (PhLP161–301) also binds Gβγ. This binding activity was further localized to the C-terminal 101 residues of PhLP (PhLP200–301) (Fig. 2). This C-terminal binding domain may explain why PhLP9 (PhLP84–301), which does not contain the 57–67 conserved sequence, still retained significant Gβγ binding activity.

The crystal structure of Gγ1γ2-phosphducin also showed two spatially and possibly functionally distinct domains in phosphducin (17). These two domains roughly correspond to the N-terminal and C-terminal halves of the molecule and do not interact with each other but both contact Gγ1γ2. The N-terminal domain may compete with Gγ3, whereas the C-terminal, thioredoxin-like domain was suggested to be responsible for Gγ1γ2 translocation away from the membrane (17). Based on sequence homology, Gaudet et al. proposed a similar structure for PhLP and predicted the Gγ1γ2 interacting residues in this protein (17). The regions occupied by these amino acids, depicted in Fig. 2, correspond to residues 54–69 and 114–152 in the N-terminal domain and to residues 240–247 and 270–277 in the C-terminal domain. Our results are in perfect agreement with these predictions and also suggest that the N-terminal and C-terminal domains can interact with Gγ1γ2 independently.

Because these domains may affect different functions of Gγ1γ2, they might be useful tools to study different aspects of Gβγ regulation as suggested by Gaudet et al. (17).

Previous studies have demonstrated that the binding of Gβγ to Go subunit inhibits its interaction with phosphducin or βARK (15, 18). In contrast, the N-terminal domain of the G protein-gated K⁺ channel as well as the small GTPase, ADP-ribosylation factor were shown to interact with either Gβγ alone or trimeric Goβγδ (19, 20). We found that recombinant Gγ3 inhibited Gβγ binding to GST-PhLP (Fig. 3). Similarly, Goα,GTPγS but not Goα,GTPδS partially abolished the interaction of Gβγ with GST-PhLP (data not shown). Together, these results suggest that only free Gβγ can interact with PhLP. Western blot analysis with a common anti-α antisemur (DuPont NEN) indicated that neither Gγ1γ2 or Gγ3 was retained on GST-PhLP along with the Gγ1γ2 dimer (Fig. 3, and data not shown). In addition, Gγ1γ2 by itself did not bind to GST-PhLP (Fig. 3). It should be noted that a relatively high concentration of recombinant Gγ1γ2 was required to totally eliminate Gβγ binding to GST-PhLP (Fig. 3). This may reflect the fact that Gγ1γ2 interacts more tightly with PhLP than with Go subunit, as was previously found for phosphducin and Gγ3 interaction with Gβγ (17).

To demonstrate Gγ1γ2-PhLP interaction in situ, the complex was immunoprecipitated following overexpression of the proteins in COS-7 cells. For these experiments, the C terminus of PhLP was tagged with a HA epitope. COS-7 cells were transiently transfected with plasmids encoding PhLP-HA, Gβγ1γ2, and Gγ2 subunits, or a combination of these proteins. Expression of the proteins in COS-7 cells was monitored by Western blot
FIG. 4. Co-immunoprecipitation of Gβγ with PhLP. A, COS-7 cells were transiently transfected either alone or in combination with PhLP-HA expression vector, Gβ1 and Gγ2 expression vectors, or with empty vector as indicated. Anti-HA immunoprecipitates (upper panel) from COS-7 cells transfected as shown were analyzed for the presence of Gβ1 and PhLP-HA proteins by immunoblotting with anti-Gβ1 and anti-HA antibodies. Aliquots of the precleared whole cell lysates (lower panels) were also monitored for expression levels of PhLP-HA and Gβ1. 1/30 volume of each lysate was loaded on the gel. B, endogenous PhLP protein was immunoprecipitated from NG108-15 cell lysates using pre-immune serum (PI) or a polyclonal anti-PhLP antibody. The precipitated proteins were then analyzed by Western blot for the presence of Gβ subunits using a monoclonal anti-Gβ1 antibody. The position of immunoglobulin heavy and light chains is indicated (*). Experiments were repeated at least twice with similar results.

analysis using monoclonal anti-HA and anti-β1 antibodies (Fig. 4A, lower panels). PhLP-HA was specifically precipitated by anti-HA antibody (Fig. 4A, upper panel). In addition, this antibody co-precipitated overexpressed Gβ1,γ2 subunits, only in cells co-expressing PhLP-HA (Fig. 4A, upper panel). We also detected Gβ in immunoprecipitates from cells transfected only with PhLP-HA plasmid (Fig. 4A, upper panel), suggesting that PhLP-HA interacts with both overexpressed and endogenous Gβγ subunits.

Interaction of endogenous PhLP and Gβγ was examined in NG108-15 neuroblasto
toma × glioma cells because this cell line expresses high basal levels of PhLP.3 Endogenous PhLP protein from NG108-15 cell lysates was immunoprecipitated by an affinity-purified polyclonal antiserum directed against PhLP. On Western blot analysis of NG108-15 cell lysates, this antiserum recognized a single band migrating at 46 kDa, the expected molecular mass for full-length PhLP protein (data not shown) (12). The antiserum also immunoprecipitated a 46-kDa protein from [35S]methionine-labeled NG108-15 cells (data not shown). Anti-PhLP immunoprecipitates contained Gβ as detected by Western blot analysis (Fig. 4B). Preimmune serum did not precipitate Gβ (Fig. 4B). These results confirm the overexpression studies (Fig. 4A) and suggest that PhLP might exist, at least partially, as a complex with Gβγ subunits in vivo.

In conclusion, these studies have documented the direct interaction of PhLP with Gβγ through both in vivo and in vitro analyses. Our deletion analysis, together with the recent crystal structure of the phodocin-Gbγ complex, suggests that two potentially independent domains on PhLP interact with Gβγ. This complements prior studies on PhLP that suggested that regions beyond the N terminus were involved in inhibition of Gβγ function in vitro (14). Because the two domains of PhLP are predicted to interact with functionally different regions of Gb (17) and we have previously shown the existence of multiple forms of the PhLP transcript, it is tempting to speculate that alternate forms of PhLP might produce distinct changes in Gβγ signaling. For example, PhLPs, which predominate the C-terminal Gβγ-binding domain and thus might produce different kinetics or extent of changes in Gβγ function than the full-length PhLP protein. It remains to be determined which of the diverse Gβγ cellular effects are functionally modified by PhLP-Gbγ interactions.

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REFERENCES
1. Gilman, A. G. (1987) Annu. Rev. Biochem. 56, 615–649
2. Simon, M. I., Strathmann, M. P., and Gautam, N. (1991) Science 252, 802–808
3. Neer, E. J. (1995) Cell 80, 249–257
4. Haga, K., and Haga, T. (1992) J. Biol. Chem. 267, 5222–5227
5. Pitcher, J. A., Inglese, J., Higgins, J. B., Arriza, J. L., Casey, P. J., Kim, C., Benovic, J. L., Kwarar, M. M., Caron, M. G., and Lefkowitz, R. J. (1992) Science 257, 1264–1267
6. Pitcher, J. A., Touhara, K., Payne, E. S., and Lefkowitz, R. J. (1995) J. Biol. Chem. 270, 11707–11710
7. Lee, B. H., Lieberman, B. S., and Lolley, R. N. (1987) Biochemistry 26, 3983–3990
8. Lee, R. H., Ting, T. D., Lieberman, B. S., Tobias, D. B., Lolley, R. N., and Ho, Y.-K. (1992) J. Biol. Chem. 267, 25104–25112
9. Yoshida, T., Willardson, B. M., Wilkins, J. F., Jensen, G. J., Thornton, B. D., and Bitesensky, M. M. (1994) J. Biol. Chem. 269, 24056–24057
10. Hekman, M., Bauer, P. H., Söllemann, P., and Lohse, M. J. (1994) FEBS Lett. 343, 120–124
11. Xu, J., Wu, D., Slepak, V. Z., and Simon, M. I. (1995) Proc. Natl. Acad. Sci. U. S. A. 92, 2086–2090
12. Miles, M. F., Barhite, S., Sanga, M., and Elliott, M. (1993) Proc. Natl. Acad. Sci. U. S. A. 90, 10831–10835
13. Wang, J.-F., Barhite, S., Thibault, C., and Miles, M. F. (1995) Alcohol. Clin. Exp. Res. 19, 34 (abstr.)
14. Schroeder, S., and Lohse, M. J. (1996) Proc. Natl. Acad. Sci. U. S. A. 93, 2100–2104
15. Hawes, B. E., Touhara, K., Kurose, H., Lefkowitz, R. J., and Inglese, J. (1994) J. Biol. Chem. 269, 29825–29830
16. Frangioni, J. V., and Neel, B. G. (1993) Ann. Biochem. 210, 179–187
17. Gaudet, R., Bohn, A., and Sigler, P. B. (1996) Cell 87, 577–588
18. Touhara, K., Inglese, J., Pitcher, J. A., Shaw, G., and Lefkowitz, R. J. (1994) J. Biol. Chem. 269, 10217–10220
19. Huang, C.-L., Slesinger, P. A., Casey, P. J., Jan, Y. N., and Jan, L. Y. (1995) Neuron 15, 1133–1143
20. Colombo, M. I., Inglese, J., D’Souza-Sheehy, C., Beron, W., and Stahl, P. D. (1995) J. Biol. Chem. 270, 24564–24571

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