WHAT IS CLAIMED IS:

1. A biosensor system for detecting G-protein activity, said biosensor system comprising the elements defined in (A) or (B):

(A)

(i) a first biosensor comprising:

a first component comprising a Gβγ interacting protein (βγIP) fused to (a) a resonance energy transfer (RET) donor; (b) a RET acceptor or (c) a first fragment of a reporter protein; and

a second component comprising a fused Gβ protein or a fused Gγ protein, wherein said Gβ protein or said Gγ protein is fused to (a) a RET donor; (b) a RET acceptor or (c) a second fragment of said reporter protein;

(ii) a second biosensor comprising:

the first and second components defined in (i); and

a third component comprising a recombinant Gα protein;

wherein (a) if said βγIP is fused to said RET donor, said Gβ or Gγ protein is fused to said RET acceptor; (b) if said βγIP is fused to said RET acceptor, said Gβ or Gγ protein is fused to said RET donor; and (c) if said βγIP is fused to said first fragment of said reporter protein, said Gβ or Gγ protein is fused to said second fragment of said reporter protein; or

(B)

(i) a biosensor comprising

a first component comprising a Gβγ interacting protein (βγIP) fused to (a) a RET donor; (b) a RET acceptor or (c) a first fragment of a reporter protein;

a second component comprising a fused G-protein coupled receptor (GPCR), wherein said GPCR is fused at its C-terminal to (a) a RET donor; (b) a RET acceptor or (c) a second fragment of said reporter protein;

a third component comprising a recombinant Gα protein;

wherein (a) if said βγIP is fused to said RET donor, said GPCR is fused to said RET acceptor; (b) if said βγIP is fused to said RET acceptor, said GPCR is fused to said RET donor; and (c) if said βγIP is fused to said first fragment of said reporter protein, said GPCR is fused to said second fragment of said reporter protein.

2. The biosensor system of claim 1, wherein said Gγ protein is fused to said RET donor, RET acceptor or second fragment.

3. The biosensor system of claim 1 or 2, wherein said RET donor, RET acceptor or second fragment is fused at the N-terminus of said Gβ or Gγ protein.

4. The biosensor system of any one of claims 1 to 3, wherein said RET donor, RET acceptor or first fragment is fused at the C-terminus of said βγIP.

5. The biosensor system of any one of claims 1 to 4, wherein said βγIP is fused to said RET acceptor and said Gβ protein, Gγ protein or GPCR is fused to said RET donor.

6. The biosensor system of any one of claims 1 to 5, wherein said RET donor is a bioluminescent protein.

7. The biosensor system of claim 6, wherein said bioluminescent protein is a luciferase.

8. The biosensor system of claim 7, wherein said luciferase is a *Renilla* luciferase.

9. The biosensor system of any one of claims 1 to 8, wherein said RET acceptor is a fluorescent protein.

10. The biosensor system of claim 9, wherein said fluorescent protein is a GFP.

11. The biosensor system of any one of claims 1 to 4, wherein said βγIP is fused to said first fragment, and said Gβ protein, Gγ protein or GPCR is fused to said second fragment.

12. The biosensor system of claim 11, wherein said reporter protein is a bioluminescent protein.

13. The biosensor system of claim 12, wherein said bioluminescent protein is a luciferase.

14. The biosensor system of claim 13, wherein said luciferase is a *Renilla* luciferase.

15. The biosensor system of claim 14, wherein said first fragment comprises about residues 1 to 110 of *Renilla* luciferase, and said second fragment comprises about residues 111 to 311 of *Renilla* luciferase.

16. The biosensor system of any one of claims 1 to 15, wherein the first component further comprises a plasma membrane (PM)-targeting moiety fused to said βγIP or said RET donor, RET acceptor or first fragment.

17. The biosensor system of claim 16, wherein said PM-targeting moiety is fused at the C-terminus of said RET donor, RET acceptor or first fragment.

18. The biosensor system of claim 16 or 17, wherein said PM-targeting moiety comprises a prenylation motif.

19. The biosensor system of claim 16, wherein said prenylation motif is the prenylation motif of human KRAS splice variant b.

20. The biosensor system of claim 19, wherein said PM-targeting moiety comprises the amino acid sequence KKKKKKSKTKCVIM (SEQ ID NO:37).

21. The biosensor system of any one of claims 16 to 20, further comprising a flexible linker between (i) said RET donor, RET acceptor or first fragment and (ii) said PM-targeting moiety.

22. The biosensor system of claim 21, wherein said flexible linker has a length corresponding to about 50 to about 500 amino acids.

23. The biosensor system of claim 22, wherein said flexible linker has a length corresponding to about 200 amino acids.

24. The biosensor system of any one of claims 1 to 23, wherein said recombinant Gα protein is human $G\alpha_q$, $G\alpha_s$, $G\alpha_{i1}$, $G\alpha_{i2}$, $G\alpha_{i3}$, $G\alpha_{t\text{-}cone}$, $G\alpha_{t\text{-}rod}$, $G\alpha_{t\text{-}gust}$, $G\alpha_z$, $G\alpha_{oA}$, $G\alpha_{oB}$, $G\alpha_{olf}$, $G\alpha_{11}$, $G\alpha_{12}$, $G\alpha_{13}$, $G\alpha_{14}$, and $G\alpha_{15}/G\alpha_{16}$ protein, or promiscuous or non-selective Gα variant thereof.

25. The biosensor system of any one of claims 1 to 24, wherein said βγIP is GRK2 or GRK3.

26. The biosensor system of any one of claims 1 to 25, wherein (i) if said second component comprises a fused Gβ protein, said first and second biosensors further comprises a recombinant Gγ protein, or (ii) if said second component comprises a fused Gγ protein, said first and second biosensors further comprises a recombinant Gβ protein.

27. The biosensor system of any one of claims 1 to 25, wherein the biosensor system defined in (A) further comprises a G-protein-coupled receptor (GPCR).

28. The biosensor system of any one of claims 1 to 27, wherein the biosensor system defined in (B) further comprises a recombinant Gβ protein and/or a recombinant Gγ protein.

29. The biosensor system of any one of claims 1 to 28, wherein said first biosensor is present in a first cell and said second biosensor is present in a second cell.

30. The biosensor system of any one of claims 1 to 28, wherein in the biosensor system defined in (A), said first biosensor is present in a first membrane preparation and said second biosensor is present in a second membrane preparation.

31. The biosensor system of any one of claims 1 to 30, wherein the biosensor system defined in (A) comprises a plurality of second biosensors, wherein each of said second biosensors comprises a different recombinant Gα protein.

32. The biosensor system of claim 31, wherein said different recombinant Gα proteins are at least two of the following Gα proteins: $G\alpha_q$, $G\alpha_s$, $G\alpha_{i1}$, $G\alpha_{i2}$, $G\alpha_{i3}$, $G\alpha_{t\text{-}cone}$, $G\alpha_{t\text{-}rod}$, $G\alpha_{t\text{-}gust}$, $G\alpha_z$, $G\alpha_{oA}$, $G\alpha_{oB}$, $G\alpha_{olf}$, $G\alpha_{11}$, $G\alpha_{12}$, $G\alpha_{13}$, $G\alpha_{14}$, and $G\alpha_{15}/G\alpha_{16}$.

33. A biosensor for detecting G-protein activity comprising:

(i) a first component comprising a Gβγ interacting protein (βγIP) fused to (a) a RET donor; (b) a RET acceptor or (c) a first fragment of a reporter protein; and

(ii) a second component comprising a fused plasma membrane (PM)-targeting moiety, wherein said PM-targeting moiety is fused to (a) a RET donor; (b) a RET acceptor or (c) a second fragment of said reporter protein;

wherein (a) if said βγIP is fused to said RET donor, said PM-targeting moiety is fused to said RET acceptor; (b) if said βγIP is fused to said RET acceptor, said PM-targeting moiety is fused to said RET donor; and (c) if said βγIP is fused to said first fragment of said reporter protein, said PM-targeting moiety is fused to said second fragment of said reporter protein.

34. The biosensor of claim 33, wherein said PM targeting moiety is a PM protein or a fragment thereof that localizes to the PM.

35. The biosensor of claim 34, wherein said PM protein or fragment thereof comprises (a) a palmitoylation, myristoylation, and/or prenylation signal sequence and/or (b) a polybasic sequence.

36. The biosensor of claim 35, wherein said polybasic sequence and prenylation signal sequence are from human KRAS splice variant b.

37. The biosensor of claim 36, wherein said PM targeting moiety comprises the amino acid sequence KKKKKKSKTKCVIM (SEQ ID NO:37).

38. The biosensor of any one of claims 34 to 37, wherein said biosensor further comprises a third component that comprises a recombinant Gα protein.

39. The biosensor of claim 38, wherein said recombinant Gα protein is of the Gq family.

40. The biosensor of claim 39, wherein said recombinant Gα protein is $G\alpha_q$ or $G\alpha_{11}$.

41. A method for determining whether a test agent modulates the activity of a GPCR, said method comprising:

(1) providing a biosensor comprising the elements defined in (A), (B) or (C):

(A)

(i) a first component comprising a Gβγ interacting protein (βγIP) fused to (a) a RET donor; (b) a RET acceptor or (c) a first fragment of a reporter protein;

(ii) a second component comprising a fused Gβ protein or a fused Gγ protein, wherein said Gβ protein or said Gγ protein is fused to (a) a RET donor; (b) a RET acceptor or (c) a second fragment of said reporter protein,

wherein (a) if said βγIP is fused to said RET donor, said Gβ or Gγ protein is fused to said RET acceptor; (b) if said βγIP is fused to said RET acceptor, said Gβ or Gγ protein is fused to said RET donor; and (c) if said βγIP is fused to said first fragment of said reporter protein, said Gβ or Gγ protein is fused to said second fragment of said reporter protein;

(iii) a third component comprising a recombinant Gα protein; and

(iv) a fourth component comprising said GPCR;

(B)

(i) a first component comprising a Gβγ interacting protein (βγIP) fused to (a) a RET donor; (b) a RET acceptor or (c) a first fragment of a reporter protein;

(ii) a second component comprising said GPCR fused at its C-terminal to (a) a RET donor; (b) a RET acceptor or (c) a second fragment of said reporter protein;

(iii) a third component comprising a recombinant Gα protein;

wherein (a) if said βγIP is fused to said RET donor, said GPCR is fused to said RET acceptor; (b) if said βγIP is fused to said RET acceptor, said GPCR is fused to said RET donor; and (c) if said βγIP is fused to said first fragment of said reporter protein, said GPCR is fused to said second fragment of said reporter protein; or

(C)

(i) a first component comprising a Gβγ interacting protein (βγIP) fused to (a) a RET donor; (b) a RET acceptor or (c) a first fragment of a reporter protein;

(ii) a second component comprising a fused plasma membrane (PM)-targeting moiety, wherein said PM-targeting moiety is fused to (a) a RET donor; (b) a RET acceptor or (c) a second fragment of said reporter protein;

wherein (a) if said βγIP is fused to said RET donor, said PM-targeting moiety is fused to said RET acceptor; (b) if said βγIP is fused to said RET acceptor, said PM-targeting moiety is fused to said RET donor; and (c) if said βγIP is fused to said first fragment of said reporter protein, said PM-targeting moiety is fused to said second fragment of said reporter protein;

(iii) a third component comprising a recombinant Gα protein; and

(iv) a fourth component comprising said GPCR; and

(2) measuring the signal emitted by said RET acceptor or reporter protein in the presence and absence of said test agent;

wherein a higher signal measured in the presence of the agent is indicative that said test agent increases the activity of said GPCR, and a lower signal measured in the presence of the agent is indicative that said agent inhibits the activity of said GPCR.

42. The method of claim 41, wherein said biosensor is the biosensors defined in any one of claims 2 to 32 and 34 to 40.

43. A method for determining whether a Gα protein is activated by a GPCR agonist, said method comprising:

(a) measuring the signal emitted by said RET acceptor or reporter protein in the presence and absence of said GPCR agonist in the first and second biosensors of the biosensor system of any one of claims 1 to 32, and

(b) identifying whether the Gα protein is activated by said GPCR agonist based on the signal emitted by said RET acceptor or reporter protein;

wherein a higher increase of the signal measured in the presence of the GPCR agonist in said second biosensor relative to said first biosensor is indicative that the Gα protein is activated by said GPCR agonist, and wherein a similar or lower increase, or a decrease, of the signal measured in the presence of the GPCR agonist in said second biosensor relative to said first biosensor is indicative that said the Gα protein is not activated by said GPCR agonist.

44. A method for determining whether a Gα protein is activated by a GPCR agonist, said method comprising:

(a) measuring the signal emitted by a RET acceptor or reporter protein in the presence and absence of said GPCR agonist in a first biosensor comprising:

(i) a first component comprising a Gβγ interacting protein (βγIP) fused to (a) a RET donor; (b) a RET acceptor or (c) a first fragment of a reporter protein; and

(ii) a second component comprising a fused G-protein coupled receptor (GPCR), wherein said GPCR is fused at its C-terminal to (a) a RET donor; (b) a RET acceptor or (c) a second fragment of said reporter protein;

(b) measuring the signal emitted by a RET acceptor or reporter protein in the presence and absence of said GPCR agonist in a second biosensor comprising:

(i) the first and second components defined in (a); and

(ii) a third component comprising a recombinant form of said Gα protein;

wherein (a) if said βγIP is fused to said RET donor, said GPCR is fused to said RET acceptor; (b) if said βγIP is fused to said RET acceptor, said GPCR is fused to said RET donor; and (c) if said βγIP is fused to said first fragment of said reporter protein, said GPCR is fused to said second fragment of said reporter protein;

wherein a higher increase of the signal measured in the presence of the GPCR agonist in said second biosensor relative to said first biosensor is indicative that the Gα protein is activated by said GPCR agonist, and wherein a similar or lower increase, or a decrease, of the signal measured in the presence of the GPCR agonist in said second biosensor relative to said first biosensor is indicative that said the Gα protein is not activated by said GPCR agonist.

45. The method of claim 43, wherein said biosensors comprise one or more of the features defined in claims 34 to 40.

46. The method of claim 41, further comprising

(3) measuring the signal emitted by said RET acceptor or reporter protein in

(a) the second biosensor(s) defined in element (A) of any one of claims 1 to 31,

(b) the biosensor defined in element (B) of any one of claims 1 to 31, or

(c) the biosensor of any one of claims 38 to 40,

in the presence and absence of a test agent and in the presence of a GPCR agonist, wherein said recombinant Gα protein is coupled to said GPCR; and

(4) determining whether said test agent is an inhibitor of said Gα protein;

wherein a lower signal measured in the presence of the test agent is indicative that said test agent is an inhibitor of said Gα protein, and a similar or higher signal measured in the presence of the test agent is indicative that said test agent is not an inhibitor of said Gα protein.

47. A method for determining whether a test agent is an inhibitor of a Gα protein of interest, said method comprising:

(1) contacting

(a) the second biosensor(s) defined in element (A) of any one of claims 1 to 32,

(b) the biosensor defined in element (B) of any one of claims 1 to 32, or

(c) the biosensor of any one of claims 38 to 40;

with a GPCR agonist, wherein said recombinant Gα protein corresponds to said Gα protein of interest;

(2) measuring the signal emitted by said RET acceptor or reporter protein in the presence and absence of said test agent; and

(c) determining whether said test agent is an inhibitor of said Gα protein,

wherein a lower signal measured in the presence of the test agent is indicative that said test agent is an inhibitor of said Gα protein of interest, and a similar or higher signal measured in the

presence of the test agent is indicative that said test agent is not an inhibitor of said Gα protein of interest.

48. A method for determining whether a test agent is an activator of a Gα protein of interest, said method comprising:

(1) contacting

(a) the second biosensor(s) defined in element (A) of any one of claims 1 to 32,

(b) the biosensor defined in element (B) of any one of claims 1 to 32, or

(c) the biosensor of any one of claims 38 to 40;

with a GPCR antagonist, wherein said recombinant Gα protein corresponds to said Gα protein of interest;

(2) measuring the signal emitted by said RET acceptor or reporter protein in the presence and absence of said test agent; and

(3) determining whether said test agent is an activator of said Gα protein,

wherein a higher signal measured in the presence of the test agent is indicative that said test agent is an activator of said Gα protein of interest, and a similar or lower signal measured in the presence of the test agent is indicative that said test agent is not an activator of said Gα protein of interest.

49. The method of any one of claims 41 to 48, wherein said RET donor is a bioluminescent protein, and wherein said method further comprises contacting the biosensor with a substrate for said donor bioluminescent protein.

50. The method of claim 49, wherein said substrate is a luciferin.

51. The method of claim 50, wherein said luciferin is a coelenterazine.

52. The method of claim 51, wherein said coelenterazine is Coelenterazine 400A.

53. The method of any one of claims 41 to 52, wherein the biosensor comprises a RET donor and a RET acceptor, and wherein said method further comprises: (i) measuring signal emitted by said RET donor, and (ii) determining the ratio [RET acceptor signal / RET donor signal].

54. A mutated Gα polypeptide comprising a mutation at a position corresponding to residue 67 and/or residue 75 of human $G\alpha_q$ protein, wherein residue 67 of human $G\alpha_q$ protein corresponds to the tyrosine residue at position 28 of SEQ ID NO:1, and residue 75 of human $G\alpha_q$ protein corresponds to the phenylalanine residue at position 36 of SEQ ID NO:1.

55. The mutated Gα polypeptide of claim 54, wherein said mutation is a substitution.

56. The mutated Gα polypeptide of claim 54 or 55, wherein said mutation is at a position corresponding to residue 67 of human $G\alpha_q$ protein.

57. The mutated Gα polypeptide of claim 56, wherein said mutation is a substitution for a non-aromatic residue.

58. The mutated Gα polypeptide of claim 57, wherein non-aromatic residue is cysteine.

59. The mutated Gα polypeptide of claim 54 or 55, wherein said mutation is at a position corresponding to residue 75 of human $G\alpha_q$ protein.

60. The mutated Gα polypeptide of claim 59, wherein said mutation is a substitution for a non-aromatic residue.

61. The mutated Gα polypeptide of claim 60, wherein said non-aromatic residue is glycine.

62. A nucleic acid comprising a sequence encoding the mutated Gα polypeptide of any one of claims 54 to 61.

63. A plasmid or vector comprising the nucleic acid of claim 62.

64. A cell comprising the nucleic acid of claim 62 or the plasmid of claim 63.