

Database tool

The systematic annotation of the three main GPCR families in Reactome

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Reactome is an open-source, freely available database of human biological pathways and processes. A major goal of our work is to provide an integrated view of cellular signalling processes that spans from ligand–receptor interactions to molecular readouts at the level of metabolic and transcriptional events. To this end, we have built the first catalogue of all human G protein-coupled receptors (GPCRs) known to bind endogenous or natural ligands. The UniProt database has records for 797 proteins classified as GPCRs and sorted into families A/1, B/2 and C/3 on the basis of amino acid sequence. To these records we have added details from the IUPHAR database and our own manual curation of relevant literature to create reactions in which 563 GPCRs bind ligands and also interact with specific G-proteins to initiate signalling cascades. We believe the remaining 234 GPCRs are true orphans. The Reactome GPCR pathway can be viewed as a detailed interactive diagram and can be exported in many forms. It provides a template for the orthology-based inference of GPCR reactions for diverse model organism species, and can be overlaid with protein–protein interaction and gene expression datasets to facilitate overrepresentation studies and other forms of pathway analysis.

Database URL: <http://www.reactome.org>

Introduction

G protein-coupled receptors (GPCRs), also known as 7-transmembrane (7TM) domain receptors, comprise the largest and most diverse gene super-family in humans—>1% of the total protein-coding human genome. Estimates of the exact number of GPCR genes vary but a recent phylogenetic analysis identified over 800 (1). Of these, 701 were classified within the rhodopsin family (type A) including 241 non-olfactory receptors. Many protein coding genes are alternatively spliced giving rise to isoforms so the true number of functionally unique receptors may be much higher than estimates based on gene numbers.

These GPCRs sense extracellular molecules and, through their interaction with G proteins, activate downstream signal transduction pathways. GPCRs respond to a huge range of stimuli, including light, odours, hormones, neurotransmitters and peptides (2). GPCRs represent around half

of cell surface drug targets (3) and are a very successful therapeutic target family for the pharmaceutical industry accounting for the majority of best-selling drugs, ~30% of all prescription pharmaceuticals on the market (4). The potential for further exploitation remains high, as only 10% of GPCRs are targeted by these marketed drugs (5).

Reactome is a free, open-source pathways database. Information in Reactome is captured by expert curators and peer-reviewed by experts in their fields of biology. The data is extensively cross-referenced to databases such as Ensembl [<http://www.ensembl.org/index.html> (6)], GO [<http://www.ebi.ac.uk/QuickGO/> (7)], PubMed (<http://www.ncbi.nlm.nih.gov/pubmed>), ChEBI [<http://www.ebi.ac.uk/chebi/index.jsp> (8)], UniProt [<http://www.uniprot.org/> (9)] and OMIM [<http://www.ncbi.nlm.nih.gov/omim> (10)]. Reactions for other species are inferred by orthology from curated human ones. Reactions can be viewed in the context of their pathways and interaction data can be overlaid

to further expand the data richness. Tools are available in Reactome to help users with analyses such as pathway over-representation (enrichment) and pathway differential expression, and data including tables of pairwise protein–protein interactions computed from manually curated reactions and complexes can be downloaded in a range of formats.

Several resources hold rich data for GPCRs. UniProt is a comprehensive protein knowledgebase of protein sequence and functional information. IUPHAR-db (International Union of basic and clinical PHARmacology, <http://www.iuphar-db.org/>) is a database of receptor nomenclature and drug classification. Its GPCR section is arranged according to the sequence homology and functional similarity of these receptors. It also contains orphan GPCR lists. These resources were used as a starting point to catalogue the GPCR project in Reactome.

Materials and methods

In UniProt, a query was constructed to search for all manually annotated and reviewed human GPCRs.

Information in Reactome is annotated by database curators. These in-house experts systematically reviewed the literature for the three GPCR families. GPCRs whose ligands were identified from published experimental data were captured via the Curator Tool, an interface which allows the curator to annotate and structure data in accord with Reactome's frame-based data model, and commit the results to a central repository (11). Data was organized into the three main GPCR families, *A/1*, *B/2* and *C/3*. Within each family, details were further structured based on the type of ligand. Attributes of a reaction captured by Reactome are:

- input molecule(s)
- output molecule(s)
- catalyst (where appropriate)
- compartment where reaction takes place
- literature reference
- species
- links to preceding reactions (e.g. the causal connection between ligand binding and G-protein interaction)
- membership in a pathway
- text summary of reaction
- curator
- expert reviewer

Input and output entities can be composed of proteins, simple chemicals or combinations of these entities (complexes).

Useful information captured from IUPHAR-db by the curation team included:

- the type of ligand the receptor bound, which helped in the organization of the project in Reactome.

Ligands were represented if they are generally accepted in the literature to be the endogenous or natural ones

- literature references associated with human receptors
- where appropriate, orthologous proteins in other species
- the primary transduction mechanism, i.e. the G protein(s) that coupled with the receptor: ligand complex
- receptor nomenclature

As of October 2009, there were 356 GPCRs captured by the IUPHAR database. The database also contains lists of orphans. Orphans are proteins classified as GPCR protein family members based on sequence similarity but whose endogenous ligands are unknown. These were investigated by Reactome curators to determine if recent advances assigned ligands to some of them.

Results

From UniProt, we retrieved records for the three main families of human GPCRs with the query:

family: 'G-protein coupled receptor' and organism:human and reviewed: yes

The query resulted in 836 protein matches. Of these, 797 proteins matched the three main families (*A/1*, *B/2* and *C/3*). We then queried IUPHAR-db and searched published literature to identify ligands for these proteins, with the results

Class *A/1*—726 UniProt records; ligands found for 519

Class *B/2*—49 UniProt records; ligands found for 29

Class *C/3*—22 UniProt records; ligands found for 15

Tables 1 (Class *A/1*) and 2 (classes *B/2* and *C/3*) show the GPCRs, their gene names and accession numbers, and the G protein with which each one interacts.

Of 797 GPCRs in families *A/1*–*C/3* screened from UniProt, we were able to catalogue 563 GPCRs that have ligands (70%), supported by information from IUPHAR and appropriate literature references.

We believe the remainder (234) to be true orphans i.e. no credible endogenous ligands have been determined for these receptors.

The project can be viewed here:

http://www.reactome.org/cgi-bin/eventbrowser?DB=gk_current&ID=372790

An on-line description of the features of these web pages and of additional pathway analysis tools that can be applied to the data is available here:

<http://www.reactome.org/userguide/Usersguide.html>

For each class, there are further subdivisions of the hierarchy, organized into ligand types that bind particular GPCRs.

Table 1. Family A GPCR names, accession numbers and G protein coupled (Receptor names link out to the corresponding reaction in Reactome)

GPCR family A Receptors	Gene Name	Acc. No.	G α subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	G α subunit(s) coupled
5-hydroxytryptamine receptor 1A	HTR1A	P08908	i	Type-2 angiotensin II receptor (AT2)	AGTR2	P50052	i, q/11
5-hydroxytryptamine receptor 1B	HTR1B	P28222	i	Apelin receptor	APLNR	P35414	i
5-hydroxytryptamine receptor 1D	HTR1D	P28221	i	B1 bradykinin receptor	BDKRB1	P46663	i, q/11
5-hydroxytryptamine receptor 1E	HTR1E	P28566	i	B2 bradykinin receptor	BDKRB2	P30411	i, q/11
5-hydroxytryptamine receptor 1F	HTR1F	P30939	i	C5a anaphylatoxin chemotactic receptor	C5AR1	P21730	i
5-hydroxytryptamine receptor 2A	HTR2A	P28223	q/11	Cholecystokinin receptor type A	CCKAR	P32238	q/11
5-hydroxytryptamine receptor 2B	HTR2B	P41595	q/11	C-C chemokine receptor type 10	CCR10	P46092	i
5-hydroxytryptamine receptor 2C	HTR2C	P28335	q/11	C-C chemokine receptor type 1	CCR1	P32246	i
5-hydroxytryptamine receptor 4	HTR4	Q13639	s	C-C chemokine receptor type 2	CCR2	P41597	i
5-hydroxytryptamine receptor 5A	HTR5A	P47898	i	C-C chemokine receptor type 3	CCR3	P51677	i
5-hydroxytryptamine receptor 6	HTR6	P50406	s	C-C chemokine receptor type 4	CCR4	P51679	i
5-hydroxytryptamine receptor	HTR7	P34969	s	C-C chemokine receptor type 5	CCR5	P51681	i
Adenosine receptor A1	ADORA1	P30542	i	C-C chemokine receptor type 6	CCR6	P51684	i
Adenosine receptor A2a	ADORA2A	P29274	s	C-C chemokine receptor type 7	CCR7	P32248	i
Adenosine receptor A2b	ADORA2B	P29275	s	C-C chemokine receptor type 8	CCR8	P51685	i
Adenosine A3 receptor	ADORA3	P33765	i	C-C chemokine receptor type 9	CCR9	P51686	i
Muscarinic acetylcholine receptor M1	CHRM1	P11229	q/11	Cysteinyl leukotriene receptor 1	CYSLTR1	Q9Y271	q/11
Muscarinic acetylcholine receptor M2	CHRM2	P08172	i	Cysteinyl leukotriene receptor 2	CYSLTR2	Q9N575	q/11
Muscarinic acetylcholine receptor M3	CHRM3	P20309	q/11	Cannabinoid receptor 1	CNR1	P21554	i
Muscarinic acetylcholine receptor M4	CHRM4	P08173	i	Cannabinoid receptor 2	CNR2	P34972	i
Muscarinic acetylcholine receptor M5	CHRM5	P08912	q/11	CX3C chemokine receptor 1	CX3CR1	P49238	unknown
Adrenocorticotrophic hormone receptor	MC2R	Q01718	s	High affinity interleukin-8 receptor A	IL8RA	P25024	i
α -1A adrenergic receptor	ADRA1A	P35348	q/11	High affinity interleukin-8 receptor B	IL8RB	P25025	i
α -1B adrenergic receptor	ADRA1B	P35368	q/11	C-X-C chemokine receptor type 3	CXCR3	P49682	i
α -1D adrenergic receptor	ADRA1D	P25100	q/11	C-X-C chemokine receptor type 4	CXCR4	P61073	i
α -2A adrenergic receptor	ADRA2A	P08913	i	C-X-C chemokine receptor type 6	CXCR6	O00574	i
α -2B adrenergic receptor	ADRA2B	P18089	i	C-X-C chemokine receptor type 7	CXCR7	P25106	i
α -2C adrenergic receptor	ADRA2C	P18825	i	D(1A) dopamine receptor	DRD1	P21728	s
β -1 adrenergic receptor	ADRB1	P08588	s	D(2) dopamine receptor	DRD2	P14416	i
β -2 adrenergic receptor	ADRB2	P07550	s	D(3) dopamine receptor	DRD3	P35462	i
β -3 adrenergic receptor	ADRB3	P13945	s	D(4) dopamine receptor	DRD4	P21917	i
Type-1 angiotensin II receptor	AGTR1	P30556	q/11	D(1B) dopamine receptor	DRD5	P21918	s

(Continued)

Table 1. Continued

GPCR family A Receptors	Gene Name	Acc. No.	Gα subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	Gα subunit(s) coupled
Duffy antigen/chemokine receptor	DARC	Q16570	unknown	Melanocyte-stimulating hormone receptor	MCTR	Q01726	s
Endothelin-1 receptor	EDNRA	P25101	q/11	Melatonin receptor type 1A	MTNR1A	P48039	i
Endothelin B receptor	EDNRB	P24530	q/11	Melatonin receptor type 1B	MTNR1B	P49286	i
N-formyl peptide receptor 2	FPR2	P25090	q/11	Substance-P receptor	TACR1	P25103	q/11
Follicle-stimulating hormone receptor	FSHR	P23945	s	Substance-K receptor	TACR2	P21452	q/11
Galanin receptor type 1	GALR1	P47211	i	Neuromedin-K receptor	TACR3	P29371	q/11
Galanin receptor type 2	GALR2	O43603	i	Neuromedin-B receptor	NMBR	P28336	q/11
Galanin receptor type 3	GALR3	O60755	i	Neuropeptides BAW receptor type 1	NPBWR1	P48145	i
Gastrin/cholecystokinin type B receptor	CCKBR	P32239	q/11	Neuropeptides BAW receptor type 2	NPBWR2	P48146	i
Gonadotropin-releasing hormone receptor	GNRHR	P30968	q/11	Neuropeptide FF receptor 1	NPFRR1	Q9GZQ6	q/11
Putative gonadotropin-releasing hormone II receptor	GNRHR2	Q96P88	q/11	Neuropeptide FF receptor 2	NPFRR2	Q9Y5X5	q/11
G-protein coupled oestrogen receptor 1	GPOR	Q99527	i	Neuropeptide Y receptor type 1	NPY1R	P25929	i
Uracil nucleotide/cysteinyl leukotriene receptor	GPR17	Q13304	i, q/11	Neuropeptide Y receptor type 2	NPY2R	P49146	i
Putative G-protein coupled receptor 44	GPR44	Q9Y5Y4	i	Neuropeptide Y receptor type 4	PPYR1	P50391	i
G-protein coupled receptor 55	GPR55	Q9Y2T6	i	Neuropeptide Y receptor type 5	NPY5R	Q15761	i
Gastrin-releasing peptide receptor	GRPR	P30550	q/11	Neurotensin receptor type 1	NTSR1	P30989	q/11
Histamine H1 receptor	HRH1	P35367	q/11	Neurotensin receptor type 2	NTSR2	O95665	q/11
Histamine H2 receptor	HRH2	P25021	s	Olfactory receptor 10A2	OR10A2	Q9H208	olf
Histamine H3 receptor	HRH3	Q9Y5N1	i	Olfactory receptor 10A3	OR10A3	P58181	olf
Histamine H4 receptor	HRH4	Q9H3N8	i	Olfactory receptor 10A4	OR10A4	Q9H209	olf
Kiss-1 receptor	KISS1R	Q969F8	q/11	Olfactory receptor 10A5	OR10A5	Q9H207	olf
Lysophosphatidic acid receptor 1	LPAR1	Q92633	i, q/11	Olfactory receptor 10A6	OR10A6	Q8NH74	olf
Lysophosphatidic acid receptor 2	LPAR2	Q9HBW0	i, q/11	Olfactory receptor 10A7	OR10A7	Q8NGE5	olf
Lysophosphatidic acid receptor 3	LPAR3	Q9UBY5	i, q/11	Olfactory receptor 10AD1	OR10AD1	Q8NGE0	olf
Lysophosphatidic acid receptor 4	LPAR4	Q99677	q/11	Olfactory receptor 10AG1	OR10AG1	Q8NH19	olf
Lysophosphatidic acid receptor 6	LPAR6	P43657	q/11	Olfactory receptor 10C1	OR10C1	Q96KK4	olf
Lutropin-choriogonadotropic hormone receptor	LHCGR	P22888	s	Olfactory receptor 10G2	OR10G2	Q8NGC3	olf
Leukotriene B4 receptor 1	LTB4R	Q15722	q/11	Olfactory receptor 10G3	OR10G3	Q8NGC4	olf
Leukotriene B4 receptor 2	LTB4R2	Q9NPC1	q/11	Olfactory receptor 10G4	OR10G4	Q8NGN3	olf
Melanocortin receptor 3	MC3R	P41968	s	Olfactory receptor 10G6	OR10G6	Q8NH81	olf
Melanocortin receptor 4	MC4R	P32245	s	Olfactory receptor 10G7	OR10G7	Q8NGN6	olf
Melanocortin receptor 5	MC5R	P33032	s	Olfactory receptor 10G8	OR10G8	Q8NGN5	olf

(Continued)

Table 1. Continued

GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled
Olfactory receptor 10G9	OR10G9	Q8NGN4	olf	Olfactory receptor 2T10	OR2T10	Q8NGZ9	olf
Olfactory receptor 10H1	OR10H1	Q9Y4A9	olf	Olfactory receptor 2T11	OR2T11	Q8NH01	olf
Olfactory receptor 10H2	OR10H2	Q60403	olf	Olfactory receptor 2T12	OR2T12	Q8NG77	olf
Olfactory receptor 10H3	OR10H3	Q60404	olf	Olfactory receptor 2T27	OR2T27	Q8NH04	olf
Olfactory receptor 10H4	OR10H4	Q8NGA5	olf	Olfactory receptor 2T29	OR2T29	Q8NH02	olf
Olfactory receptor 10H5	OR10H5	Q8NGA6	olf	Olfactory receptor 2T33	OR2T33	Q8NG76	olf
Olfactory receptor 10J1	OR10J1	P30954	olf	Olfactory receptor 2T34	OR2T34	Q8NGX1	olf
Olfactory receptor 10J3	OR10J3	Q5JRS4	olf	Olfactory receptor 2T35	OR2T35	Q8NGX2	olf
Olfactory receptor 10J5	OR10J5	Q8NHC4	olf	Olfactory receptor 4A15	OR4A15	Q8NGL6	olf
Olfactory receptor 10K1	OR10K1	Q8NGX5	olf	Olfactory receptor 4A16	OR4A16	Q8NH70	olf
Olfactory receptor 10K2	OR10K2	Q6IF99	olf	Olfactory receptor 4A47	OR4A47	Q6IF82	olf
Olfactory receptor 10P1	OR10P1	Q8NGE3	olf	Olfactory receptor 4C45	OR4C45	A6NMZ5	olf
Olfactory receptor 10Q1	OR10Q1	Q8NGQ4	olf	Olfactory receptor 4C46	OR4C46	A6NHA9	olf
Olfactory receptor 10R2	OR10R2	Q8NGX6	olf	Olfactory receptor 4F15	OR4F15	Q8NGB8	olf
Olfactory receptor 10S1	OR10S1	Q8NGN2	olf	Olfactory receptor 4F17	OR4F17	Q8NGA8	olf
Olfactory receptor 10T2	OR10T2	Q8NGX3	olf	Olfactory receptor 4F21	OR4F21	O95013	olf
Olfactory receptor 10V1	OR10V1	Q8NGI7	olf	Olfactory receptor 5A2	OR5A2	Q8NGJ7	olf
Olfactory receptor 10W1	OR10W1	Q8NGF6	olf	Olfactory receptor 5A4	OR5A4	Q8NGJ6	olf
Olfactory receptor 14A2	OR14A2	Q96R54	olf	Olfactory receptor 5A7	OR5A7	Q8NH64	olf
Olfactory receptor 14C36	OR14C36	Q8NHC7	olf	Olfactory receptor 5B2	OR5B2	Q9Y5P1	olf
Olfactory receptor 14I1	OR14I1	A6ND48	olf	Olfactory receptor 5B4	OR5B4	Q9Y5P0	olf
Olfactory receptor 14J1	OR14J1	Q9UGF5	olf	Olfactory receptor 5B5	OR5B5	Q9H339	olf
Olfactory receptor 14K1	OR14K1	Q8NGZ2	olf	Olfactory receptor 5B5	OR5B6	Q9H340	olf
Olfactory receptor 2A12	OR2A12	Q8NGT7	olf	Olfactory receptor 5D1	OR5D1	Q8NGF3	olf
Olfactory receptor 2A14	OR2A14	Q96R47	olf	Olfactory receptor 5E1	OR5E1	Q8TCB6	olf
Olfactory receptor 2A25	OR2A25	A4D2G3	olf	Olfactory receptor 5E2	OR5E2	Q9H255	olf
Olfactory receptor 2AG1	OR2AG1	Q9H205	olf	Olfactory receptor 5F1	OR5F1	A6NGY5	olf
Olfactory receptor 2AG2	OR2AG2	A6NM03	olf	Olfactory receptor 5F2	OR5F2	Q8NH61	olf
Olfactory receptor 2AJ1	OR2AJ1	Q8NGZ0	olf	Olfactory receptor 5G1	OR5G1	Q8NGK1	olf
Olfactory receptor 2AK2	OR2AK2	Q8NG84	olf	Olfactory receptor 5G2	OR5G2	Q8NGK0	olf
Olfactory receptor 2AP1	OR2AP1	Q8NGE2	olf	Putative olfactory receptor 51H1	OR51H1P	Q8NH63	olf
Olfactory receptor 2AT4	OR2AT4	A6NND4	olf	Olfactory receptor 51I1	OR51I1	Q9H343	olf

(Continued)

Table 1. Continued

GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled
Olfactory receptor 51I2	OR51I2	Q9H344	olf	Olfactory receptor 56A1	OR56A1	Q8NGH5	olf
Olfactory receptor 51J1	OR51J1	Q9H342	olf	Olfactory receptor 56A3	OR56A3	Q8NH54	olf
Olfactory receptor 51L1	OR51L1	Q8NGJ5	olf	Olfactory receptor 56A4	OR56A4	Q8NGH8	olf
Olfactory receptor 51M1	OR51M1	Q9H341	olf	Olfactory receptor 56A5	OR56A5	P0C7T3	olf
Olfactory receptor 51Q1	OR51Q1	Q8NH59	olf	Olfactory receptor 56B1	OR56B1	Q8NGI3	olf
Olfactory receptor 51S1	OR51S1	Q8NGJ8	olf	Olfactory receptor 56B4	OR56B4	Q8NH76	olf
Olfactory receptor 51T1	OR51T1	Q8NGJ9	olf	Olfactory receptor 5AC2	OR5AC2	Q9NZP5	olf
Olfactory receptor 51V1	OR51V1	Q9H2C8	olf	Olfactory receptor 5AK2	OR5AK2	Q8NH90	olf
Olfactory receptor 52A1	OR52A1	Q9UKL2	olf	Olfactory receptor 5AN1	OR5AN1	Q8NGI8	olf
Olfactory receptor 52A5	OR52A5	Q9H2C5	olf	Olfactory receptor 5AP2	OR5AP2	Q8NGF4	olf
Olfactory receptor 52B2	OR52B2	Q96RD2	olf	Olfactory receptor 5AR1	OR5AR1	Q8NGP9	olf
Olfactory receptor 52B4	OR52B4	Q8NGK2	olf	Olfactory receptor 5AS1	OR5AS1	Q8N127	olf
Olfactory receptor 52B6	OR52B6	Q8NGF0	olf	Olfactory receptor 5AU1	OR5AU1	Q8NGC0	olf
Olfactory receptor 52D1	OR52D1	Q9H346	olf	Olfactory receptor 5H14	OR5H14	A6NHG9	olf
Olfactory receptor 52E2	OR52E2	Q8NGJ4	olf	Olfactory receptor 5H15	OR5H15	A6NDH6	olf
Olfactory receptor 52E4	OR52E4	Q8NGH9	olf	Olfactory receptor 6C65	OR6C65	A6NIZ3	olf
Olfactory receptor 52E5	OR52E5	Q8NH55	olf	Olfactory receptor 6C68	OR6C68	A6NDL8	olf
Olfactory receptor 52E6	OR52E6	Q96RD3	olf	Olfactory receptor 6C70	OR6C70	A6NIJ9	olf
Olfactory receptor 52E8	OR52E8	Q6IFG1	olf	Olfactory receptor 6C74	OR6C74	A6NCV1	olf
Olfactory receptor 52H1	OR52H1	Q8NGJ2	olf	Olfactory receptor 6C75	OR6C75	A6NL08	olf
Olfactory receptor 52I1	OR52I1	Q8NGK6	olf	Olfactory receptor 6C76	OR6C76	A6NM76	olf
Olfactory receptor 52I2	OR52I2	Q8NH67	olf	Olfactory receptor 7E24	OR7E24	Q6IFN5	olf
Olfactory receptor 52K1	OR52K1	Q8NGK4	olf	Opsin-3	OPN3	Q9H1Y3	t
Olfactory receptor 52K2	OR52K2	Q8NGK3	olf	Melanopsin	OPN4	Q9UHM6	q/11
Olfactory receptor 52L1	OR52L1	Q8NGH7	olf	Opsin-5	OPN5	Q6U736	t
Olfactory receptor 52M1	OR52M1	Q8NGK5	olf	δ-type opioid receptor	OPRD1	P41143	i
Olfactory receptor 52N1	OR52N1	Q8NH53	olf	κ-type opioid receptor	OPRK1	P41145	i
Olfactory receptor 52N2	OR52N2	Q8NGI0	olf	μ-type opioid receptor	OPRM1	P35372	i
Olfactory receptor 52N4	OR52N4	Q8NGI2	olf	Nociceptin receptor	OPRL1	P41146	i
Olfactory receptor 52N5	OR52N5	Q8NH56	olf	Blue-sensitive opsin	OPN1SW	P03999	t
Olfactory receptor 52R1	OR52R1	Q8NGF1	olf	Rhodopsin	RHO	P08100	t
Olfactory receptor 52W1	OR52W1	Q6IF63	olf	Green-sensitive opsin	OPN1MW	P04001	t

(Continued)

Table 1. Continued

GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled
Red-sensitive opsin	OPN1LW	P04000	t	Olfactory receptor 2B11	OR2B11	Q5JQ55	olf
Visual pigment-like receptor peropsin	RRH	O14718	t	Olfactory receptor 2C1	OR2C1	Q95371	olf
Olfactory receptor 1A1	OR1A1	Q9P1Q5	olf	Olfactory receptor 2C3	OR2C3	Q8N628	olf
Olfactory receptor 1A2	OR1A2	Q9Y585	olf	Olfactory receptor 2D2	OR2D2	Q9H210	olf
Olfactory receptor 1B1	OR1B1	Q8NGR6	olf	Olfactory receptor 2D3	OR2D3	Q8NGH3	olf
Olfactory receptor 1C1	OR1C1	Q15619	olf	Olfactory receptor 2F1	OR2F1	Q13607	olf
Olfactory receptor 1D2	OR1D2	P34982	olf	Olfactory receptor 2F2	OR2F2	Q95006	olf
Olfactory receptor 1F1	OR1F1	Q43749	olf	Olfactory receptor 2G2	OR2G2	Q8NGZ5	olf
Olfactory receptor 1F12	OR1F12	Q8NHA8	olf	Olfactory receptor 2G3	OR2G3	Q8NGZ4	olf
Olfactory receptor 1G1	OR1G1	P47890	olf	Olfactory receptor 2G6	OR2G6	Q5TZ20	olf
Olfactory receptor 1I1	OR1I1	O60431	olf	Olfactory receptor 2H1	OR2H1	Q9GZK4	olf
Olfactory receptor 1J1	OR1J1	Q8NGS3	olf	Olfactory receptor 2H2	OR2H2	Q95918	olf
Olfactory receptor 1J2	OR1J2	Q8NGS2	olf	Putative olfactory receptor 2I1	OR2I1P	Q8NGU4	olf
Olfactory receptor 1J4	OR1J4	Q8NGS1	olf	Olfactory receptor 2I1	OR2J1	Q9GZK6	olf
Olfactory receptor 1K1	OR1K1	Q8NGR3	olf	Olfactory receptor 2J2	OR2J2	O76002	olf
Olfactory receptor 1L1	OR1L1	Q8NH94	olf	Olfactory receptor 2J3	OR2J3	O76001	olf
Olfactory receptor 1L3	OR1L3	Q8NH93	olf	Olfactory receptor 2K2	OR2K2	Q8NGT1	olf
Olfactory receptor 1L4	OR1L4	Q8NGR5	olf	Olfactory receptor 2L2	OR2L2	Q8NH16	olf
Olfactory receptor 1L6	OR1L6	Q8NGR2	olf	Olfactory receptor 2L3	OR2L3	Q8NG85	olf
Olfactory receptor 1L8	OR1L8	Q8NGR8	olf	Olfactory receptor 2L5	OR2L5	Q8NG80	olf
Olfactory receptor 1M1	OR1M1	Q8NGA1	olf	Olfactory receptor 2L8	OR2L8	Q8NGY9	olf
Olfactory receptor 1N1	OR1N1	Q8NGS0	olf	Olfactory receptor 2L13	OR2L13	Q8N349	olf
Olfactory receptor 1N2	OR1N2	Q8NGR9	olf	Olfactory receptor 2M2	OR2M2	Q96R28	olf
Olfactory receptor 1Q1	OR1Q1	Q15612	olf	Olfactory receptor 2M3	OR2M3	Q8NG83	olf
Olfactory receptor 1S1	OR1S1	Q8NH92	olf	Olfactory receptor 2M4	OR2M4	Q96R27	olf
Olfactory receptor 1S2	OR1S2	Q8NGQ3	olf	Olfactory receptor 2M5	OR2M5	A3KFT3	olf
Olfactory receptor 2A2	OR2A2	Q6IF42	olf	Olfactory receptor 2M7	OR2M7	Q8NG81	olf
Olfactory receptor 2A4	OR2A4	Q95047	olf	Olfactory receptor 2S2	OR2S2	Q9NQN1	olf
Olfactory receptor 2B2	OR2B2	Q9GZK3	olf	Olfactory receptor 2T1	OR2T1	O43869	olf
Putative olfactory receptor 2B3	OR2B3	O76000	olf	Olfactory receptor 2T2	OR2T2	Q6IF00	olf
Olfactory receptor 2B6	OR2B6	P58173	olf	Olfactory receptor 2T3	OR2T3	Q8NH03	olf
Putative olfactory receptor 2B8	OR2B8P	P59922	olf	Olfactory receptor 2T4	OR2T4	Q8NH00	olf

(Continued)

Table 1. Continued

GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled
Olfactory receptor 2T5	OR2T5	Q6IEZ7	olf	Olfactory receptor 4E1	OR4E1	P0C645	olf
Olfactory receptor 2T6	OR2T6	Q8NHC8	olf	Olfactory receptor 4E2	OR4E2	Q8NGC2	olf
Olfactory receptor 2T8	OR2T8	A6NH00	olf	Olfactory receptor 4F3/4F16/4F29	OR4F3	Q6IEY1	olf
Olfactory receptor 2V1	OR2V1	Q8NHB1	olf	Olfactory receptor 4F4	OR4F4	Q96R69	olf
Olfactory receptor 2V2	OR2V2	Q96R30	olf	Olfactory receptor 4F5	OR4F5	Q8NH21	olf
Olfactory receptor 2W1	OR2W1	Q9Y3N9	olf	Olfactory receptor 4F6	OR4F6	Q8NGB9	olf
Olfactory receptor 2W3	OR2W3	Q7Z3T1	olf	Olfactory receptor 4K1	OR4K1	Q8NGD4	olf
Olfactory receptor 2Y1	OR2Y1	Q8NGV0	olf	Olfactory receptor 4K2	OR4K2	Q8NGD2	olf
Olfactory receptor 2Z1	OR2Z1	Q8NG97	olf	Olfactory receptor 4K3	OR4K3	Q96R72	olf
Olfactory receptor 3A1	OR3A1	P47881	olf	Olfactory receptor 4K5	OR4K5	Q8NGD3	olf
Olfactory receptor 3A2	OR3A2	P47893	olf	Olfactory receptor 4K13	OR4K13	Q8NH42	olf
Olfactory receptor 3A3	OR3A3	P47888	olf	Olfactory receptor 4K14	OR4K14	Q8NGD5	olf
Olfactory receptor 3A4	OR3A4	P47883	olf	Olfactory receptor 4K15	OR4K15	Q8NH41	olf
Putative olfactory receptor 4A4	OR4A4P	Q8NGN8	olf	Olfactory receptor 4K17	OR4K17	Q8NGC6	olf
Olfactory receptor 4A5	OR4A5	Q8NH83	olf	Olfactory receptor 4L1	OR4L1	Q8NH43	olf
Olfactory receptor 4A8	OR4A8P	P0C604	olf	Olfactory receptor 4M1	OR4M1	Q8NGD0	olf
Olfactory receptor 4B1	OR4B1	Q8NGF8	olf	Olfactory receptor 4M2	OR4M2	Q8NGB6	olf
Olfactory receptor 4C3	OR4C3	Q8NH37	olf	Olfactory receptor 4N2	OR4N2	Q8NGD1	olf
Olfactory receptor 4C5	OR4C5	Q8NGB2	olf	Olfactory receptor 4N4	OR4N4	Q8N0Y3	olf
Olfactory receptor 4C6	OR4C6	Q8NH72	olf	Olfactory receptor 4N5	OR4N5	Q8IXE1	olf
Olfactory receptor 4C11	OR4C11	Q6IEV9	olf	Olfactory receptor 4P4	OR4P4	Q8NGL7	olf
Olfactory receptor 4C12	OR4C12	Q96R67	olf	Olfactory receptor 4Q2	OR4Q2	P0C623	olf
Olfactory receptor 4C13	OR4C13	Q8NGP0	olf	Olfactory receptor 4Q3	OR4Q3	Q8NH05	olf
Olfactory receptor 4C15	OR4C15	Q8NGM1	olf	Olfactory receptor 4S1	OR4S1	Q8NGB4	olf
Olfactory receptor 4C16	OR4C16	Q8NGL9	olf	Olfactory receptor 4S2	OR4S2	Q8NH73	olf
Olfactory receptor 4D1	OR4D1	Q15615	olf	Olfactory receptor 4X1	OR4X1	Q8NH49	olf
Olfactory receptor 4D2	OR4D2	P58180	olf	Olfactory receptor 4X2	OR4X2	Q8NGF9	olf
Olfactory receptor 4D5	OR4D5	Q8NGN0	olf	Olfactory receptor 5A1	OR5A1	Q8NGJ0	olf
Olfactory receptor 4D6	OR4D6	Q8NGJ1	olf	Olfactory receptor 5A2	OR5A2	Q8NGI9	olf
Olfactory receptor 4D9	OR4D9	Q8NGE8	olf	Olfactory receptor 5B2	OR5B2	Q96R09	olf
Olfactory receptor 4D10	OR4D10	Q8NGI6	olf	Olfactory receptor 5B3	OR5B3	Q8NH48	olf
Olfactory receptor 4D11	OR4D11	Q8NGI4	olf	Olfactory receptor 5B12	OR5B12	Q96R08	olf

(Continued)

Table 1. Continued

GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	Gz. subunit(s) coupled
Olfactory receptor 5B17	OR5B17	Q8NGF7	olf	Olfactory receptor 6A2	OR6A2	O95222	olf
Olfactory receptor 5B21	OR5B21	A6NL26	olf	Olfactory receptor 6B1	OR6B1	O95007	olf
Olfactory receptor 5C1	OR5C1	Q8NGR4	olf	Olfactory receptor 6B2	OR6B2	Q6IFH4	olf
Olfactory receptor 5D13	OR5D13	Q8NGL4	olf	Olfactory receptor 6B3	OR6B3	Q8NGW1	olf
Olfactory receptor 5D14	OR5D14	Q8NGL3	olf	Olfactory receptor 6C1	OR6C1	Q96RD1	olf
Olfactory receptor 5D16	OR5D16	Q8NGK9	olf	Olfactory receptor 6C2	OR6C2	Q9NZP2	olf
Olfactory receptor 5D18	OR5D18	Q8NGL1	olf	Olfactory receptor 6C3	OR6C3	Q9NZP0	olf
Olfactory receptor 5F1	OR5F1	O95221	olf	Olfactory receptor 6C4	OR6C4	Q8NGE1	olf
Olfactory receptor 5H1	OR5H1	A6NKK0	olf	Olfactory receptor 6C6	OR6C6	A6NF89	olf
Olfactory receptor 5H2	OR5H2	Q8NGV7	olf	Olfactory receptor 6F1	OR6F1	Q8NGZ6	olf
Olfactory receptor 5H6	OR5H6	Q8NGV6	olf	Olfactory receptor 6J1	OR6J1	Q8NGC5	olf
Olfactory receptor 5I1	OR5I1	Q13606	olf	Olfactory receptor 6K2	OR6K2	Q8NGY2	olf
Olfactory receptor 5J2	OR5J2	Q8NH18	olf	Olfactory receptor 6K3	OR6K3	Q8NGY3	olf
Olfactory receptor 5K1	OR5K1	Q8NHB7	olf	Olfactory receptor 6K6	OR6K6	Q8NGW6	olf
Olfactory receptor 5K2	OR5K2	Q8NHB8	olf	Olfactory receptor 6M1	OR6M1	Q8NGM8	olf
Olfactory receptor 5K3	OR5K3	A6NET4	olf	Olfactory receptor 6N1	OR6N1	Q8NGY5	olf
Olfactory receptor 5K4	OR5K4	A6NMS3	olf	Olfactory receptor 6N2	OR6N2	Q8NGY6	olf
Olfactory receptor 5L1	OR5L1	Q8NGL2	olf	Olfactory receptor 6P1	OR6P1	Q8NGX9	olf
Olfactory receptor 5L2	OR5L2	Q8NGL0	olf	Olfactory receptor 6Q1	OR6Q1	Q8NGQ2	olf
Olfactory receptor 5M1	OR5M1	Q8NGP8	olf	Olfactory receptor 6S1	OR6S1	Q8NH40	olf
Olfactory receptor 5M3	OR5M3	Q8NGP4	olf	Olfactory receptor 6T1	OR6T1	Q8NGN1	olf
Olfactory receptor 5M8	OR5M8	Q8NGP6	olf	Olfactory receptor 6V1	OR6V1	Q8N148	olf
Olfactory receptor 5M9	OR5M9	Q8NGP3	olf	Olfactory receptor 6X1	OR6X1	Q8NH79	olf
Olfactory receptor 5M10	OR5M10	Q6IEU7	olf	Olfactory receptor 6Y1	OR6Y1	Q8NGX8	olf
Olfactory receptor 5M11	OR5M11	Q96RB7	olf	Olfactory receptor 7A5	OR7A5	Q15622	olf
Olfactory receptor 5P2	OR5P2	Q8WZ92	olf	Olfactory receptor 7A10	OR7A10	O76100	olf
Olfactory receptor 5P3	OR5P3	Q8WZ94	olf	Olfactory receptor 7A17	OR7A17	O14581	olf
Olfactory receptor 5R1	OR5R1	Q8NH85	olf	Olfactory receptor 7C1	OR7C1	O76099	olf
Olfactory receptor 5T1	OR5T1	Q8NG75	olf	Olfactory receptor 7C2	OR7C2	O60412	olf
Olfactory receptor 5T2	OR5T2	Q8NGG2	olf	Olfactory receptor 7D4	OR7D4	Q8NG98	olf
Olfactory receptor 5T3	OR5T3	Q8NGG3	olf	Olfactory receptor 7G1	OR7G1	Q8NGA0	olf
Olfactory receptor 5W2	OR5W2	Q8NH69	olf	Olfactory receptor 7G2	OR7G2	Q8NG99	olf

(Continued)

Table 1. Continued

GPCR family A Receptors	Gene Name	Acc. No.	Gα subunit(s) coupled	GPCR family A Receptors	Gene Name	Acc. No.	Gα subunit(s) coupled
Olfactory receptor 7G3	OR7G3	Q8NG95	olf	Prostaglandin F2-α receptor	PTGFR	P43088	q/11
Olfactory receptor 8A1	OR8A1	Q8NGG7	olf	Prostaglandin receptor	PTGIR	P43119	s
Olfactory receptor 8B3	OR8B3	Q8NGG8	olf	Prolactin-releasing peptide receptor	PRLHR	P49683	unknown
Olfactory receptor 8B4	OR8B4	Q96RC9	olf	Platelet-activating factor receptor	PTAFR	P25105	q/11
Olfactory receptor 8B8	OR8B8	Q15620	olf	Pyroglutamylated RFamide peptide receptor	QRFRP	Q96P65	q/11
Olfactory receptor 8B12	OR8B12	Q8NGG6	olf	RPE-retinal G protein-coupled receptor	RGR	P47804	t
Olfactory receptor 8D1	OR8D1	Q8WZ84	olf	Sphingosine 1-phosphate receptor 1	S1PR1	P21453	i
Olfactory receptor 8D2	OR8D2	Q9GZM6	olf	Sphingosine 1-phosphate receptor 2	S1PR2	Q95136	i, q/11
Olfactory receptor 8D4	OR8D4	Q8NGM9	olf	Sphingosine 1-phosphate receptor 3	S1PR3	Q99500	i, q/11, 12/13
Orexin receptor type 1	HCRTR1	O43613	q/11	Sphingosine 1-phosphate receptor 4	S1PR4	O95977	i
Orexin receptor type 2	HCRTR2	O43614	q/11	Sphingosine 1-phosphate receptor 5	S1PR5	Q9H228	i
Oxoecosanoid receptor 1	OXER1	Q8TDS5	i	Somatostatin receptor type 1	SSTR1	P30872	i
Oxytocin receptor	OXTR	P30559	q/11	Somatostatin receptor type 2	SSTR2	P30874	i
P2Y purinoceptor 1	P2RY1	P47900	q/11	Somatostatin receptor type 3	SSTR3	P32745	i
P2Y purinoceptor 2	P2RY2	P41231	q/11	Somatostatin receptor type 4	SSTR4	P31391	i
P2Y purinoceptor 4	P2RY4	P51582	i	Somatostatin receptor type 5	SSTR5	P35346	i
P2Y purinoceptor 6	P2RY6	Q15077	q/11	Thromboxane A2 receptor	TBXA2R	P21731	q/11, 13
P2Y purinoceptor 8	P2RY8	Q86VZ1	unknown	Trace amine-associated receptor 1	TAAAR1	Q96RJ0	s
Putative P2Y purinoceptor 10	P2RY10	O00398	q/11	Trace amine-associated receptor 2	TAAAR2	Q9P1P5	s
P2Y purinoceptor 11	P2RY11	Q96G91	s, q/11	Putative trace amine-associated receptor 3	TAAAR3	Q9P1P4	s
P2Y purinoceptor 12	P2RY12	Q9H244	i	Trace amine-associated receptor 5	TAAAR5	O14804	s
P2Y purinoceptor 13	P2RY13	Q9BPV8	i	Trace amine-associated receptor 6	TAAAR6	Q96R18	s
P2Y purinoceptor 14	P2RY14	Q15391	i	Trace amine-associated receptor 8	TAAAR8	Q969N4	s
Proteinase-activated receptor 1	F2R	P25116	q/11	Trace amine-associated receptor 9	TAAAR9	Q96R19	s
Proteinase-activated receptor 2	F2RL1	P55085	q/11	Thyrotropin receptor	TSHR	P16473	s
Proteinase-activated receptor 3	F2RL2	O00254	q/11	Vasopressin V1a receptor	AVPR1A	P37288	q/11
Proteinase-activated receptor 4	F2RL3	Q96R10	q/11	Vasopressin V1b receptor	AVPR1B	P47901	q/11
Prostaglandin D2 receptor	PTGDR	Q13258	s	Vasopressin V2 receptor	AVPR2	P30518	s
Prostaglandin E2 receptor EP1 subtype	PTGER1	P34995	q/11	Chemokine XC receptor 1	XCR1	P46094	q/11
Prostaglandin E2 receptor EP2 subtype	PTGER2	P43116	s				
Prostaglandin E2 receptor EP3 subtype	PTGER3	P43115	i				
Prostaglandin E2 receptor EP4 subtype	PTGER4	P35408	s				

Table 2. Family B and C GPCR names, accession numbers and G protein coupled (GPCR names link out to the corresponding Reactome reaction)

GPCR family 2/B Receptors	Gene Name	Acc. No.	G α subunit(s) coupled	GPCR family 3/C Receptors	Gene Name	Acc. No.	G α subunit(s) coupled
Brain-specific angiogenesis inhibitor 1	BAI1	O14514	unknown	Extracellular calcium-sensing receptor	CASR	P41180	i, q/11
Brain-specific angiogenesis inhibitor 2	BAI2	O60241	unknown	GABA type B receptor 1 subunit 1	GABBR1	Q9UB55	i
Brain-specific angiogenesis inhibitor 3	BAI3	O60242	unknown	GABA type B receptor subunit 2	GABBR2	O75899	i
Calcitonin receptor	CALCR	P30988	s	GPCR family C group 6 member A	GPRC6A	Q5T6X5	q/11
Calcitonin gene-related peptide type 1 receptor	CALCRL	Q16602	s	Metabotropic glutamate receptor 1	GRM1	Q13255	q/11
Corticotropin-releasing factor receptor 1	CRHR1	P34998	s	Metabotropic glutamate receptor 2	GRM2	Q14416	i
Corticotropin-releasing factor receptor 2	CRHR2	Q13324	s	Metabotropic glutamate receptor 3	GRM3	Q14832	i
Growth hormone-releasing hormone receptor	GHRHR	Q02643	s	Metabotropic glutamate receptor 4	GRM4	Q14833	i
Gastric inhibitory polypeptide receptor	GIPR	P48546	s	Metabotropic glutamate receptor 5	GRM5	P41594	q/11
Glucagon-like peptide 1 receptor	GLP1R	P43220	s	Metabotropic glutamate receptor 6	GRM6	O15303	i
Glucagon-like peptide 2 receptor	GLP2R	O95838	s	Metabotropic glutamate receptor 7	GRM7	Q14831	i
Glucagon receptor	GCCR	P47871	q/11, s	Metabotropic glutamate receptor 8	GRM8	O00222	i
Pituitary adenylate cyclase-activating polypeptide type 1 receptor	ADCYAP1R1	P41586	s	Taste receptor 1 member 1	TAS1R1	Q7RTX1	unknown
Parathyroid hormone receptor 1	PTH1R	Q03431	s	Taste receptor 1 member 2	TAS1R2	Q8TE23	unknown
Parathyroid hormone 2 receptor	PTH2R	P49190	s	Taste receptor 1 member 3	TAS1R3	Q7RTX0	unknown
Secretin receptor	SCTR	P47872	s				
Vasoactive intestinal polypeptide receptor 1	VIPR1	P32241	s				
Vasoactive intestinal polypeptide receptor 2	VIPR2	P41587	s				
Frizzled-10	FZD10	Q9ULW2	i				
Frizzled-1	FZD1	Q9UP38	i				
Frizzled-2	FZD2	Q14332	i				
Frizzled-3	FZD3	Q9NPG1	i				
Frizzled-4	FZD4	Q9ULV1	i				
Frizzled-5	FZD5	Q13467	i				
Frizzled-6	FZD6	O60353	i				
Frizzled-7	FZD7	O75084	i				
Frizzled-8	FZD8	Q9H461	i				
Frizzled-9	FZD9 (FZD3)	O00144	i				
Smoothed homologue	SMO (SMOH)	Q99835	i				

Discussion

First and foremost, this addition to Reactome provides a computationally-accessible resource for information about ligand-binding GPCRs. The three main families in human are annotated, together with downstream signalling events mediated by coupling to the appropriate G protein. Each receptor protein record has multiple link-outs to key databases related to sequence, genetic disorders, ontology and literature, further enriching the information a user can view. These annotations of GPCRs by protein family complement the extensive annotation by ligand specificity previously compiled by Alexander and colleagues (12).

A total of 563 ligand-binding GPCRs were identified and included in Reactome; an additional 234 with no identifiable ligand were not. Notably, we included a set of GPCRs thought to function as olfactory receptors. In many cases, these GPCRs have been identified and classified based on their interaction when expressed in a model cultured cell with members of a small set of standard test odorant molecules. These studies are generally accepted as establishing the olfactory receptor function of these GPCRs, albeit without identifying the odorant molecule(s) with which they interact under physiological conditions (13).

Though the absence of any identified ligand presents problems for the pharmaceutical industry and for researchers wishing to study a receptor using a tool agonist, orphan receptors can be of interest when linked to a particular subcellular location and/or physiological process. For instance, the predominantly dorsal root ganglion expressed MRGX family of receptors have been extensively studied because of their narrow and therapeutically interesting expression profile (14). The pathway contexts provided by Reactome annotation provide an additional functional grouping that may be useful in generating testable hypotheses about roles of orphan GPCRs.

Orphan GPCRs have been the subject of intensive research including ligand screening by pharmaceutical companies for many years (15,16), so why do so many GPCRs have no identified ligand? There are several reasons why the endogenous ligands are still undetermined for some orphan GPCRs:

- They may be accessory proteins that enable another GPCR to function. The classic example of this is the GABAB receptor, which functions as an obligate dimer (17). Dimerization may be the minimum functional state of GPCRs (18) and heterodimers may be common. Some orphan receptors may only be functional when they form a heterodimer. This adds a level of complexity that has not been systematically addressed by ligand screening because of the difficulties inherent in simultaneously expressing two receptors.
- They themselves may need an as yet unknown accessory protein to respond to their ligand. The prime example of this is the amylin (AMY) receptor which is a dimer composed of a calcitonin receptor and one of three RAMPs to create AMY1-3 receptors (19,20). Other examples of accessory proteins that modify receptor surface expression include the REEP and RTPs (21).
- They may not signal via G proteins i.e. they may have some other downstream effector. There are numerous examples of GPCR signalling that is independent of G proteins (22); a receptor that only signals via these mechanisms is unlikely to be identified by a ligand screening exercise using any of the established technologies.
- They may require the presence of an accessory protein for successful signalling (23) that is not present in the assay systems routinely used for ligand identification.
- Ligand screening platforms used to date are unable to detect G12/13 coupled receptors, indeed G12/13 signalling is rarely measured directly, but inferred from remote downstream events (24). The G12/13 signalling route has only recently been studied in any detail (25) so perhaps a portion of orphans fall into this category. This point could be extended to suggest that none of the ligand screening technologies are optimal for every receptor subtype. It could be that some ligand-receptor interactions have been missed because the signal was not detectable using the technology available. Newly emerging technologies such as Surface Plasmon Resonance may help to address this (26).

Overlaying protein-protein interaction data e.g. from IntAct on the curated Reactome GPCR dataset may provide a powerful approach for identifying candidate heterodimer partners and their potential functions and thus provides a novel tool for the study of orphan receptors (27). Overlaying protein-small molecule data from resources such as PubChem, ChEMBL or proprietary sources may enable identification of cofactors or modulators and could identify novel lead compounds.

Reactome contains several tools for the analysis of large-scale data sets that the user can submit to the resource. Results of such analyses are exportable in many formats from PNG images to systems biology data standards such as BioPAX and SBML. Some key features of the data in Reactome are:

- data in Reactome is free and open-source
- data can be analysed by the 'Skypainter' tool in Reactome to perform pathway enrichment analysis or expression analysis
- Reactome contains orthologue predictions to many model organisms, providing a template for analysing structure-function conservation in GPCR gene families across species

- bulk querying of Reactome data can be easily performed using the 'BioMart' tool.

Features accessed from the pathway diagrams page (Entity Level Views or ELVs)

- Protein: Protein interaction overlays could potentially provide researchers with a starting point for novel drug targets
- Protein: drug interaction overlays could help to distinguish potentially useful drugs that act on a narrow range of similar processes from ones likely to have adverse effects because they interact with many proteins or with proteins involved in a diverse range of processes.

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