

An Update On Signaling In GO

GO Consortium Meeting

May 2011

Los Angeles

Rebecca Foulger and the Signaling WG

GO Signaling Workshop

- ❖ *Where:* EBI
- ❖ *When:* February 16th and 17th 2011

- ❖ *Who:*

Andrea Townsend-Nicholson

Andrew Chatry-aryamontri

~~Andrew Winter~~

Rebecca Foulger

Midori Harris

Paola Roncaglia

Chris Mungall (17th)

Sandra Orchard (Intact)

Ruth Lovering (BHF-UCL)

Varsha Khodiyar (BHF-UCL)

Peter D'Eustachio (Reactome)

Val Wood (PomBase)

Rachael Huntley (GOA)

Susan Tweedie (FlyBase)

Alex Diehl (remotely)

http://gocwiki.geneontology.org/index.php/Signaling_Workshop_February_2011

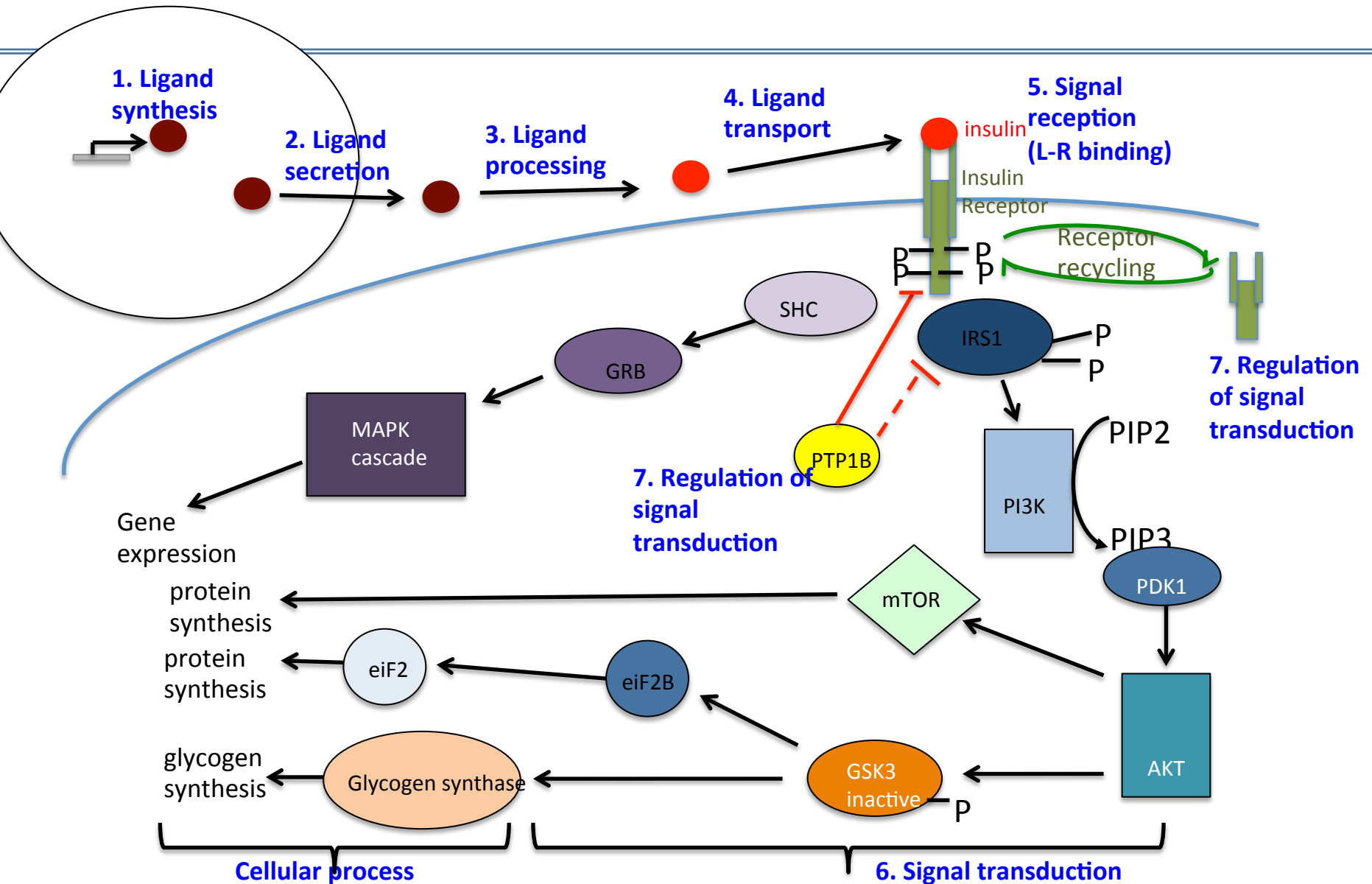
Topics Discussed At the Workshop

- ❖ What should and shouldn't be covered by signaling?
- ❖ Defining the start and end points of all signaling processes
 - signaling
 - generation of a signal
 - signal transduction
 - initiation of signal transduction
 - termination of signal transduction
 - signaling pathway
 - signaling cascade
 - paracrine/autocrine/endocrine signaling
- ❖ Defining a receptor
- ❖ Connecting cell-surface receptors with the intracellular signaling cassettes
- ❖ Dealing with non 1:1 ligand:receptor combinations
- ❖ How does receptor-mediated endocytosis fit with signaling?
- ❖ Is it important HOW a signal reaches the receiving cell?

KEY DECISIONS & POINTS

from the workshop

signaling ; GO:0023052



SIGNALING: what does it include?

- ❖ Grouping term for:
 - **TYPES** of signaling (e.g. epithelial-mesenchymal cell signaling)
 - **PROCESSES** in signaling (E.g. signal transduction)
- ❖ **DOES INCLUDE**
 - Everything from ligand-receptor down (signal transduction)
 - The signal travelling from the producing cell to the receiving cell
- ❖ **DOES NOT INCLUDE:**
 - Synthesis of the signal (E.g. Transcription of the insulin gene)
 - Synthesis of the receptor (E.g. Transcription of the insulin receptor gene).
 - Processing of the receptor prior to ligand binding (eg glycosylation of the insulin receptor)

(NB: These steps would **REGULATE** signaling).
- ❖ **COMPLICATED BECAUSE:**
 - *In some cases, the stimulus is the signal (eg nutrient signaling pathway).*
 - *In some cases, the signal is synthesised in response to the stimulus.*
 - *In some cases, the signal is released in response to the stimulus (eg insulin)*
- ❖ **OUTSTANDING QUESTION:**
 - *Is secretion the first step in signaling?*

signal and signaling

Signaling GO:0023052

Old Def: The entirety of a process whereby information is transmitted. This process **begins with the initiation of the signal** and ends when a response has been triggered.

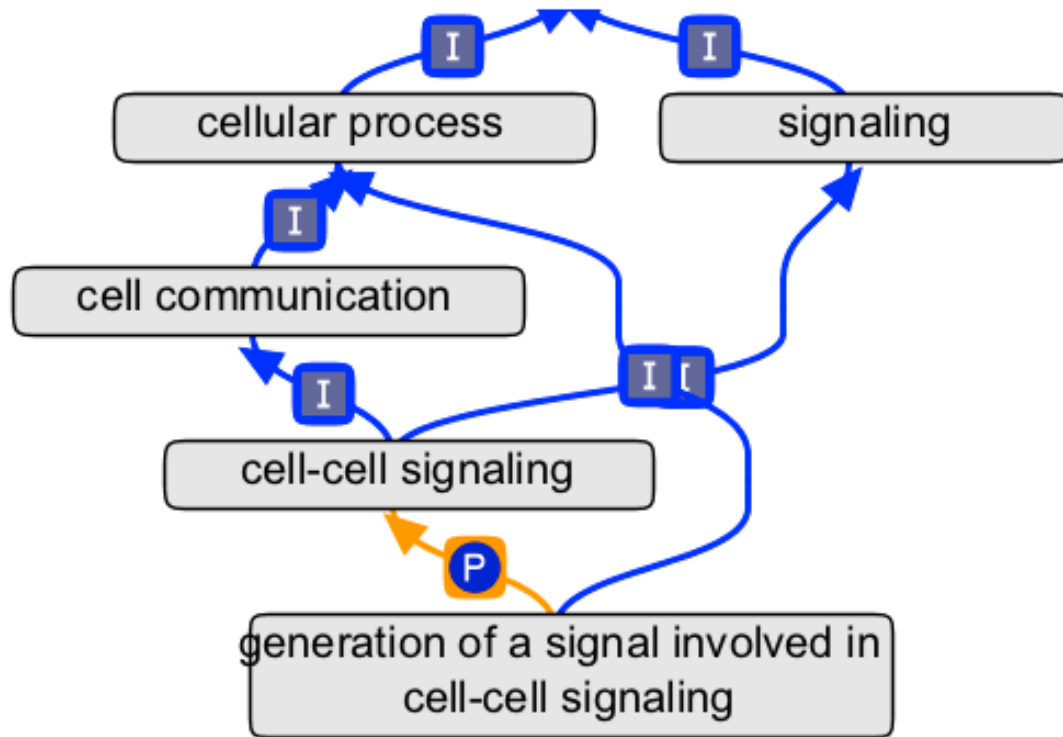
New Def: The entirety of a process in which information is transmitted. This process **begins with an active signal** and ends when a response has been triggered.

Signal (Glossary):

Any variable property or parameter that serves to convey information, and may be a physical entity such as a gene product or small molecule, a photon, or a change in state such as movement or voltage change.

TO DO...

- ❖ re-look at placement of:
 - ‘generation of a signal involved in cell-cell signaling ; GO:0003001’



Definition:

The cellular process that creates a physical entity or change in state, i.e. a signal, that originates in one cell and is used to transfer information to another cell. This process begins with the initial formation of the signal and ends with the mature form and placement of the signal.

Endocrine & paracrine signaling

❖ ENDOCRINE AND PARACRINE TERMS ADDED TO GO:

[endocrine signaling](#) ; GO:0038002

Def: The transfer of information from one cell to another, where an endocrine hormone is transported from the signal-producing cell to the receiving cell via the circulatory system (via blood, lymph or cerebrospinal fluid). The signaling cell and the receiving cell are often distant to each other.

is_a: cell-cell signaling ; GO:0007267

part_of: endocrine process ; GO:0050886

part_of: multicellular organismal signaling ; GO:0035637

ISBN:3527303782, ISBN:0199264678

[paracrine signaling](#) ; GO:0038001

Def: The transfer of information from one cell to another, where the signal travels from the signal-producing cell to the receiving cell by passive diffusion or bulk flow in intercellular fluid. The signaling cell and the receiving cell are usually in the vicinity of each other.

ISBN:3527303782

is_a: cell-cell signaling ; GO:0007267

(‘paracrine signaling’ is currently a narrow synonym of cell-cell signaling ; GO:0007267)

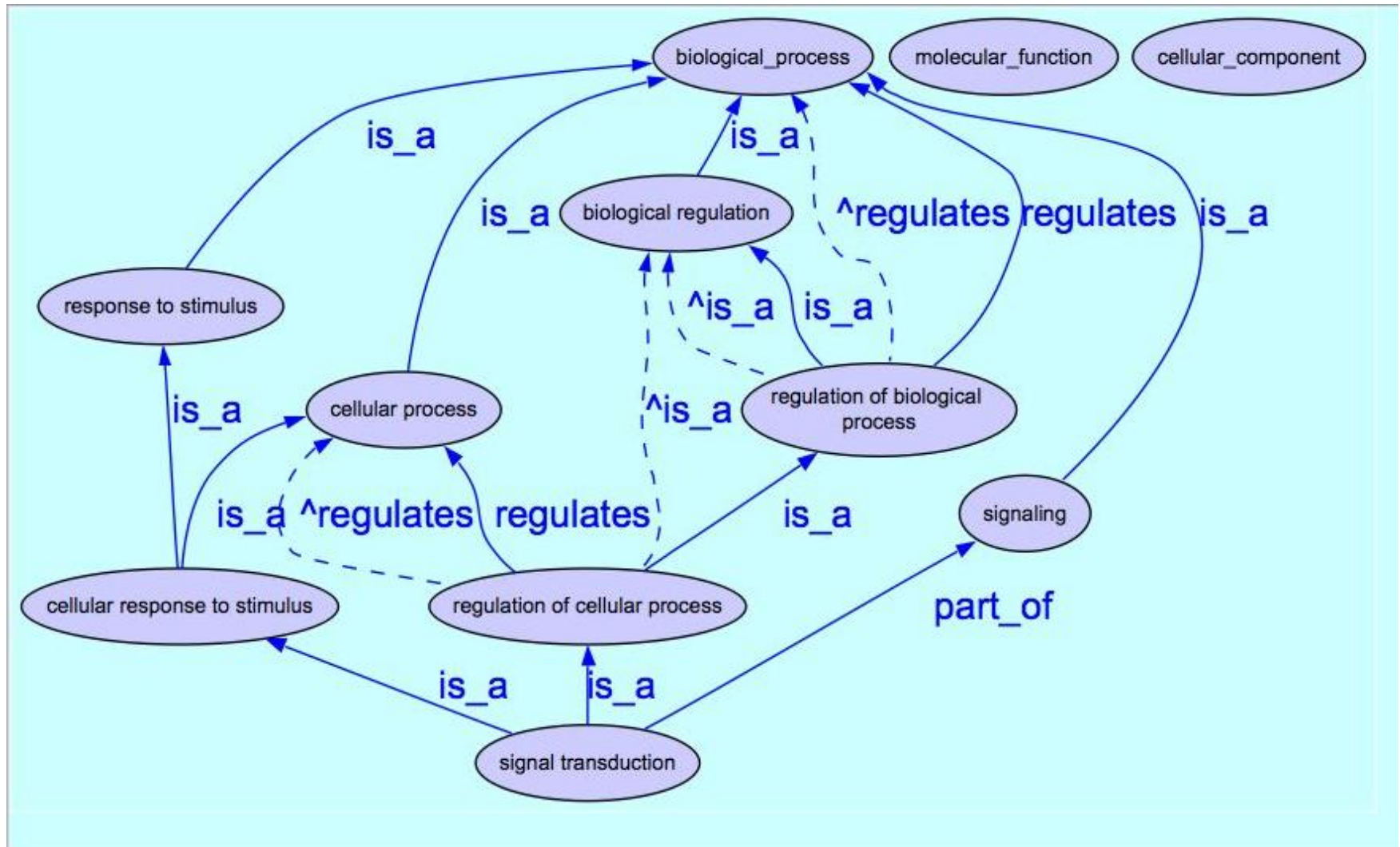
Thank you to Stan for contacting endocrinology experts too!

Signal Transduction

Signal Transduction

- ❖ **Revised Def:** The cellular process in which a signal is conveyed to trigger a change in the activity or state of a cell. Signal transduction begins with reception of a signal, e.g. a ligand binding to a receptor or receptor activation by a stimulus such as light, and ends with regulation of a downstream cellular process, e.g. regulation of transcription or regulation of a metabolic process. Signal transduction covers signaling from receptors located on the surface of the cell, and signaling via molecules located within the cell. For signaling between cells, signal transduction is restricted to events at and within the receiving cell.

Signal transduction regulates cellular processes



Initiation and Termination of Signal Transduction

Initiation/termination of signal transduction

- ❖ initiation of signal transduction ; GO:0023036
 - Definition: The process in which a signal causes activation of a receptor
 - Comment: Note that this term is intended for the annotation of both ligands and receptors.
- ❖ Definitions don't match the comments.
- ❖ The end of 'initiation of signal transduction' was unclear.
- ❖ For many of the children the ligand was not a GP (signal initiation by light).
- ❖ => **WAS OBSOLETE**
- ❖ termination of signal transduction ; GO:0023021
 - Definition: The signaling process in which signal transduction is brought to an end rather than being reversibly modulated.

Signaling Pathway

After removal of the process/pathway split, all pathway terms were put back under 'signal transduction', so there is no generic signaling pathway term....

How it used to look
(up to January 2011)

- ⊖ ← **signaling**
 - ⊕ ← **behavioral signaling**
 - ⊕ ← **cell-cell signaling**
 - ⊕ ← **extracellular matrix-cell signaling**
 - ⊕ ← **motogenic signaling initiating cell movement in cerebral cortex**
 - ← **motogenic signaling involved in interneuron migration from the subpallium to the cortex**
 - ← **motogenic signaling involved in postnatal olfactory bulb interneuron migration**

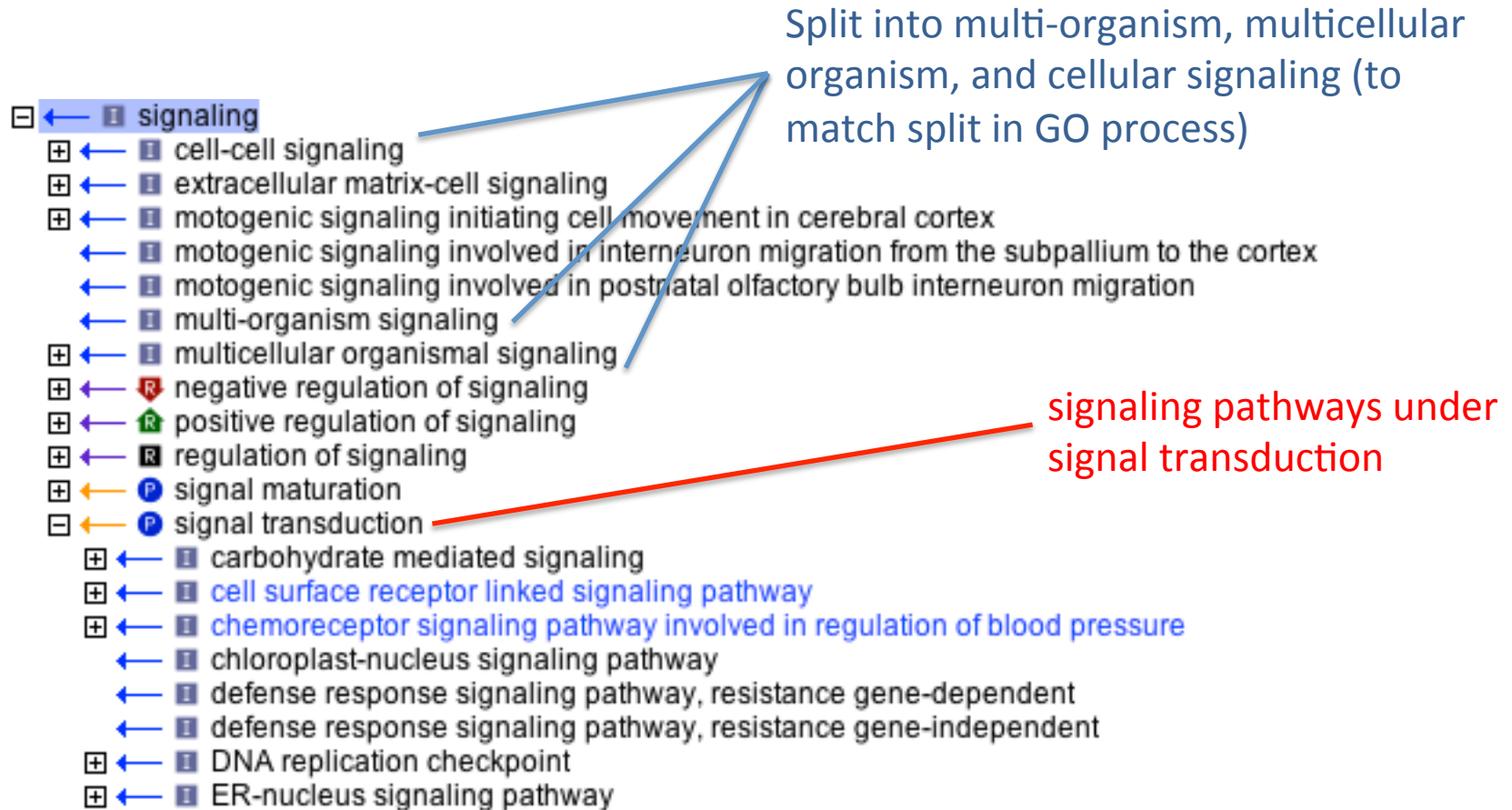
- ⊖ ← **signaling pathway**
 - ⊕ ← **carbohydrate mediated signaling**
 - ⊕ ← **cell surface receptor linked signaling pathway**
 - ⊕ ← **chemoreceptor signaling pathway involved in regulation of blood pressure**
 - ← **defense response signaling pathway, resistance gene-dependent**
 - ← **defense response signaling pathway, resistance gene-independent**
 - ⊕ ← **hormone-mediated signaling pathway**
 - ← **hydrogen peroxide mediated signaling pathway**
 - ⊕ ← **immune response-regulating signaling pathway**
 - ⊕ ← **intracellular signaling pathway**
 - ⊕ ← **jasmonic acid mediated signaling pathway**
 - ⊕ ← **lipoprotein mediated signaling**
 - ⊕ ← **negative regulation of signaling pathway**
 - ⊕ ← **nitric oxide mediated signal transduction**
 - ⊕ ← **osmosensory signaling pathway**
 - ⊕ ← **positive regulation of signaling pathway**
 - ⊕ ← **red or far-red light signaling pathway**
 - ⊕ ← **regulation of signaling pathway**
 - ⊕ ← **salicylic acid mediated signaling pathway**
 - ⊕ ← **sphingolipid signaling pathway**

'signaling pathway' &
'signaling process' are
siblings

- ⊖ ← **signaling process**
 - ⊕ ← **consequence of signal transmission**
 - ⊖ ← **generation of a signal involved in cell-cell signaling**
 - ⊕ ← **creation of an inductive signal**
 - ⊕ ← **signal release**
 - ⊕ ← **initiation of signal transduction**
 - ⊕ ← **receptor recycling**
 - ⊕ ← **regulation of signaling process**
 - ⊕ ← **signal release**
- ⊖ ← **signal transmission**
 - ⊕ ← **signal release**
 - ⊕ ← **signal transduction**
 - ← **signal transmission via air**
 - ← **signal transmission via conformational transition**
 - ⊕ ← **signal transmission via diffusible molecule**
 - ⊕ ← **signal transmission via phosphorylation event**
 - ← **signal transmission via transcytosis**
 - ⊕ ← **signal transmission via vascular system**
 - ⊕ ← **transmission of nerve impulse**
 - ⊕ ← **two-component signal transduction system (phosphorelay)**
 - ⊕ ← **termination of signal transduction**

'signaling pathway' and
'signal transduction' aren't
connected

Once the split was removed.....



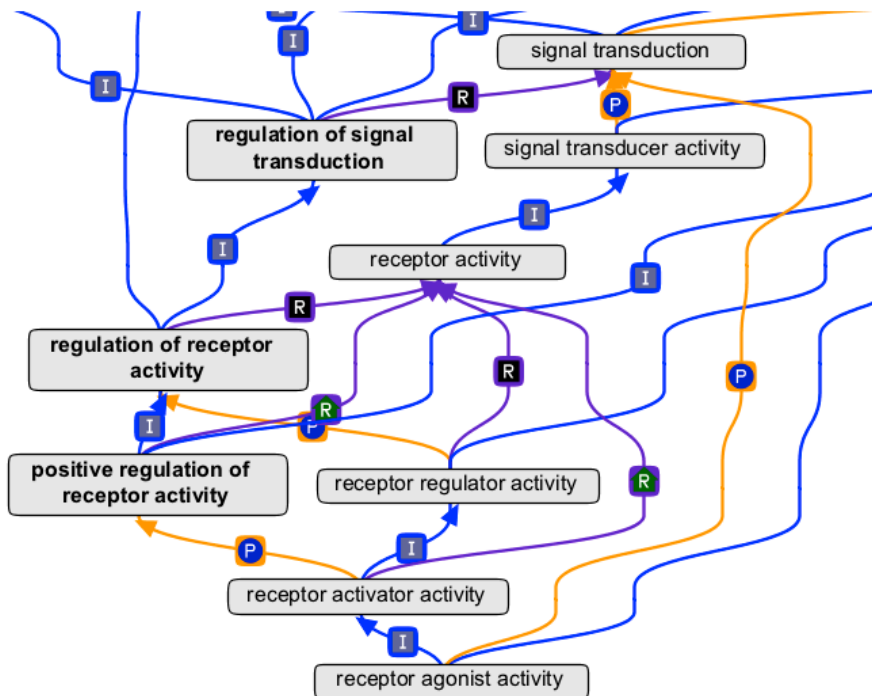
...simplified to aid more consistent annotation

SIGNALING PATHWAYS

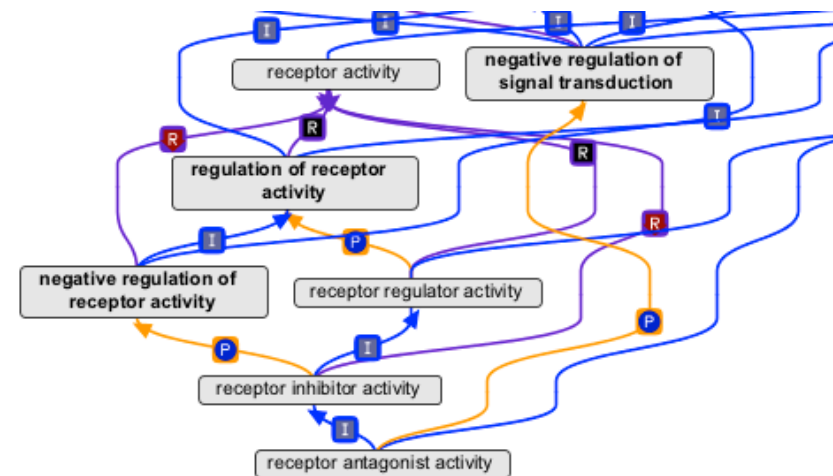
- ❖ Limited to events **WITHIN THE RECEIVING CELL**.
- ❖ Begin with ligand binding to a receptor (or for non-protein ligands [e.g. light], a receptor being activated).
- ❖ End with **REGULATION** of a downstream **CELLULAR process**.
- ❖ A ligand is part of the pathway (so can be annotated to 'signal transduction').
- ❖ **Specific transcription factors ARE** part of a pathway because they pass the signal on to RNA polymerase. They are the last GP in the pathway (eg FOXO).
- ❖ **Glycogen synthase** is **NOT** part of the pathway because it is not passing the signal on... it is part of the downstream cellular process of glycogen synthesis.

Process-Function Relationships

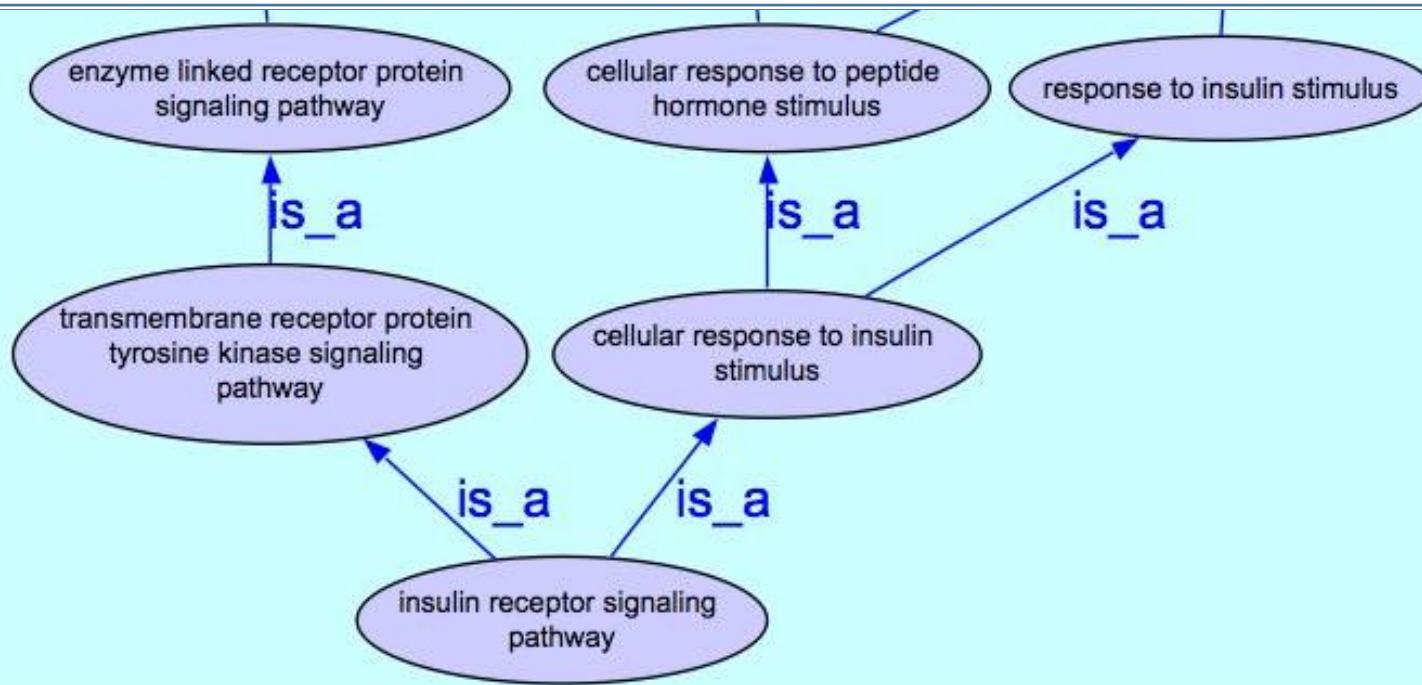
- Receptor agonist



- Receptor antagonist



A note about ‘response to’...



- The ligand (insulin) is part of the pathway. Therefore, if a user queries for ‘cellular response to insulin stimulus’ they will get back hits to insulin.
- We decided we could live with this (as it was felt that the ligand SHOULD be part of the pathway, and we didn’t want to split up the pathways into everything from receptor-downwards).

Pathways and downstream processes

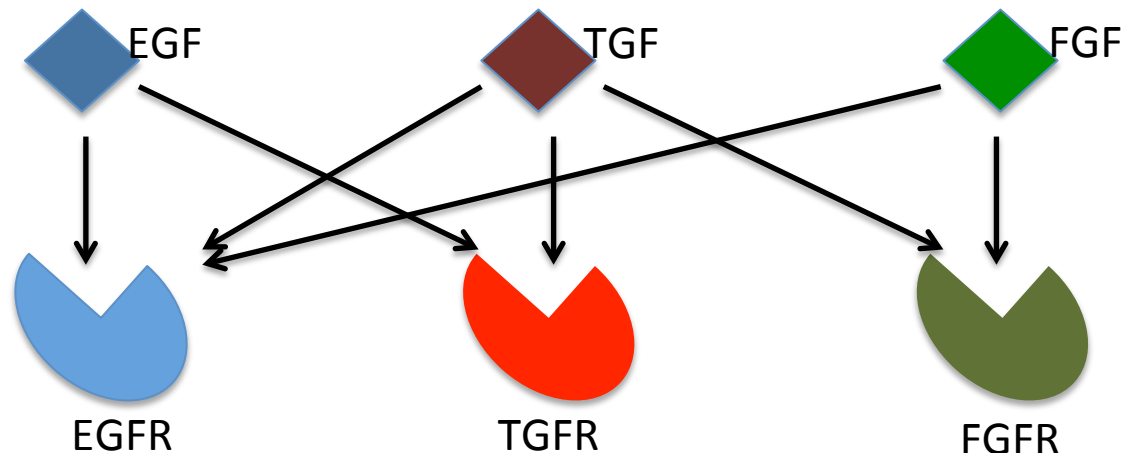
- ❖ One thing we needed to do was to connect up the pathways to their downstream processes. There are 2 'stages' of downstream processes:
 - **CELLULAR** (E.g. transcription, protein synthesis, *apoptosis*)
 - **MULTI-CELLULAR** (E.g. heart development, wound healing, pattern formation)
- ❖ Each pathway **REGULATES** a **CELLULAR PROCESS**.
- ❖ And these **cellular processes** are involved in **multi-cellular processes** (E.g. heart development, pattern formation, lung induction).
- ❖ So the GO term:
 - **Wnt receptor signaling pathway involved in kidney development ; GO:0061289**
means Wnt receptor signaling pathway involved in **regulating x cellular process** (probably transcription) **that contributes to kidney development**.

Non 1:1 receptor:ligand binding

Non 1:1 receptor:ligand binding

❖ PROBLEMS

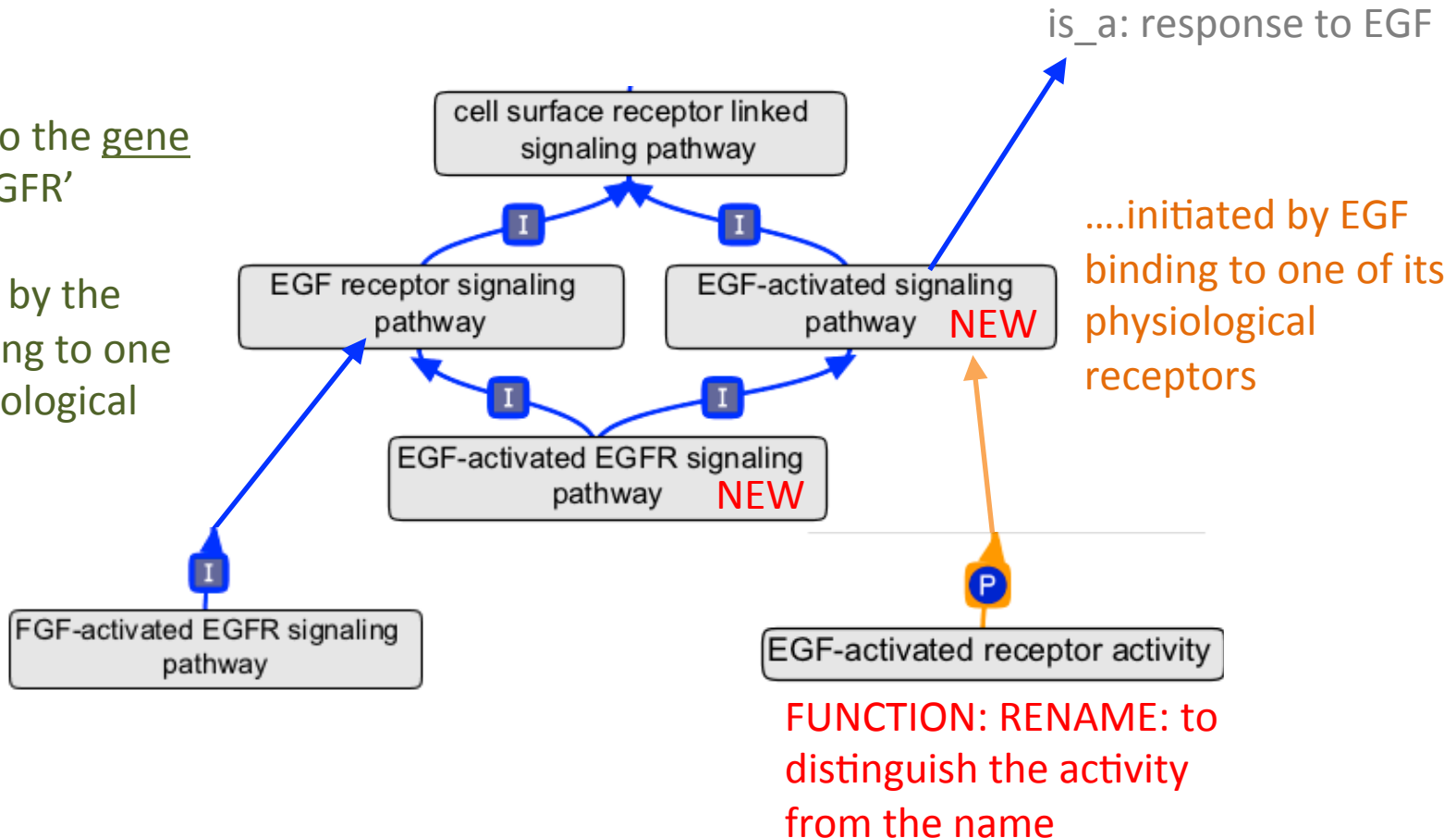
- Many receptors bind >1 ligand. Many ligands bind >1 receptor.
- Many receptors are named after the first ligand they bind to (called the EGFR even though it binds EGF, TGF etc)...
- Mixture of definitions at the moment:
 - ...epidermal growth factor receptor binding to one of its physiological ligands (*epidermal growth factor receptor signaling pathway*)
 - ...any member of the activin family binding to a cell surface receptor (*activin receptor signaling pathway*)



PROPOSAL...

Referring to the gene product 'EGFR'

....initiated by the EGFR binding to one of its physiological ligands



Still to work out...

- ❖ Where do the receptor agonists and antagonists fit in?
- ❖ E.g. EGFR agonist activity ; GO:NEW

Is this:

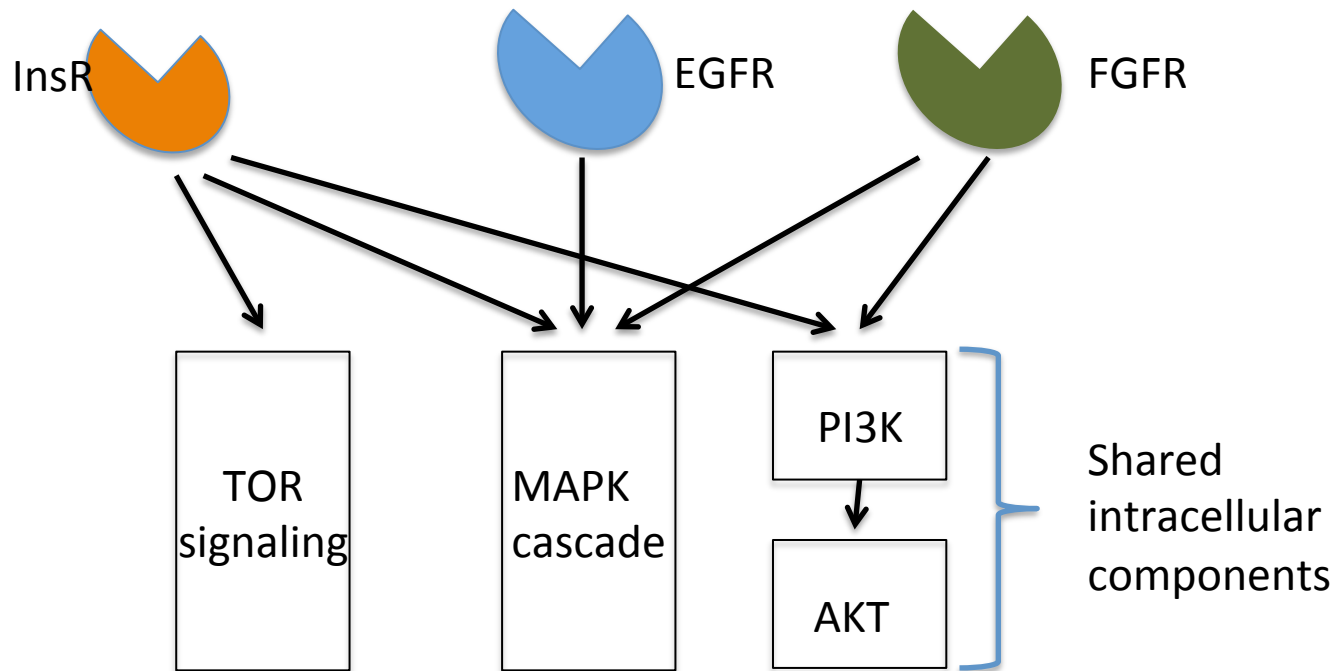
- i) an agonist to the gene product called 'EGFR'?
- ii) an agonist to an EGF-activated receptor (eg EGF)?
- iii) anything that starts the EGFR signaling pathway?

Intracellular signaling units

Types of intracellular signaling

- ❖ Groups of proteins (cascades)
 - MAPK cascade
 - JAK-STAT cascade
 - Hippo cascade
- ❖ One protein/mediator
 - Activation/inhibition of adenylate cyclase (GPCR signaling)
 - *PI3K*
 - *PKB (aka AKT)*
 - *TOR*
 - Ca^{++} mediated

Connecting receptors and intracellular signaling components

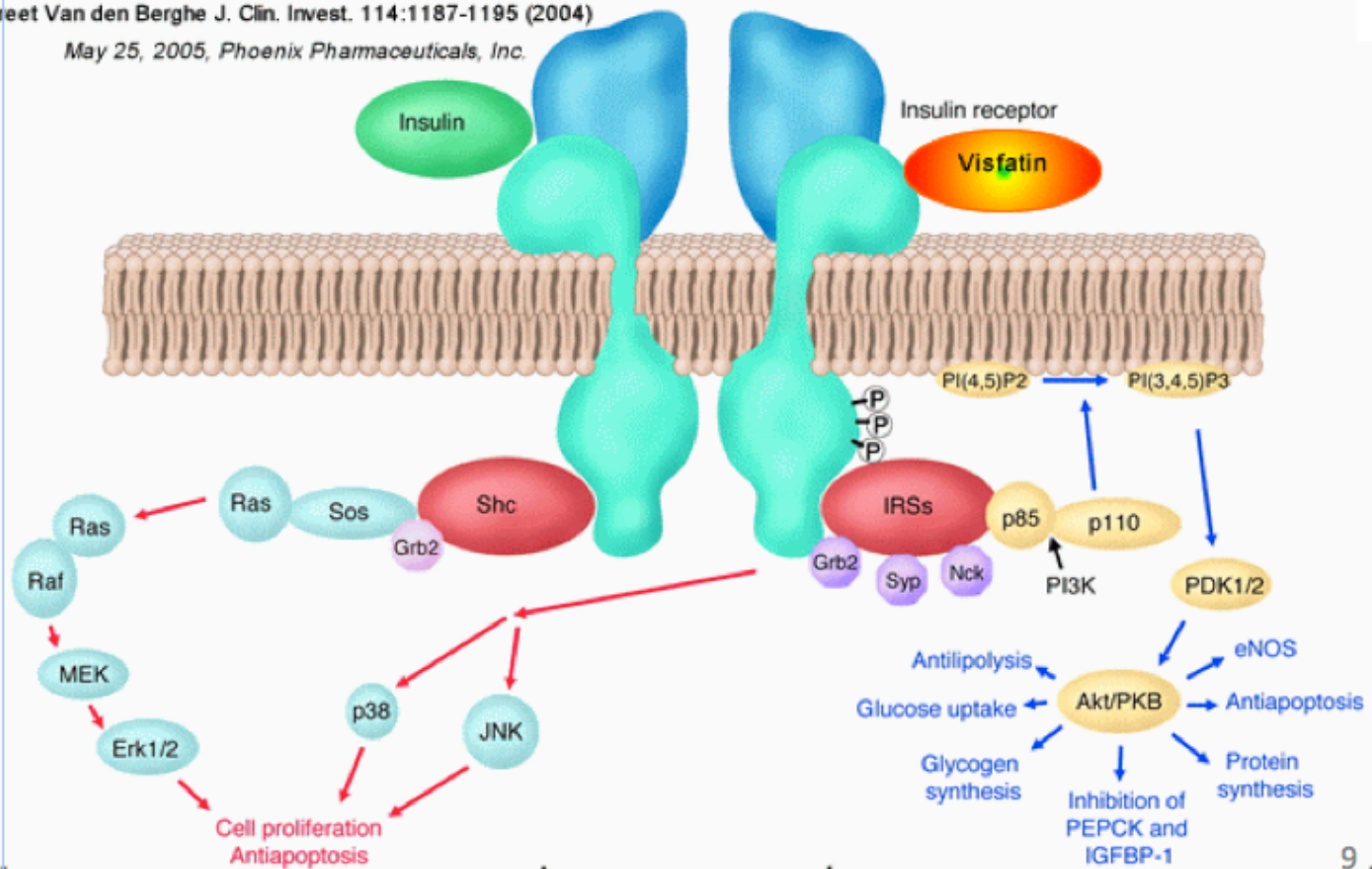


Can look at from perspective of:

- ❖ receptor signaling via multiple routes.

OR

- ❖ AKT being a hub to multiple upstream receptors



IRS1 activates PI3K (signals via PI3K) downstream of the insulin receptor

4 options discussed

- ❖ 1/ insulin receptor signaling pathway VIA TOR signaling cascade
- ❖ 2/ TOR signaling cascade INVOLVED IN insulin receptor signaling pathway
- ❖ 3/ REGULATION OF TOR signaling cascade by insulin receptor signaling pathway
- ❖ 4/ ~~Don't connect them~~

Each option would involve annotating a different subset of the pathway.

The best of the bunch ?

- ❖ 1/ insulin receptor signaling pathway VIA TOR signaling cascade
- ❖ 2/ ~~TOR signaling cascade~~ INVOLVED IN ~~insulin receptor signaling pathway~~
- ❖ 3/ REGULATION ~~OF TOR signaling cascade by insulin receptor signaling pathway~~
- ❖ 4/ ~~Don't connect them~~

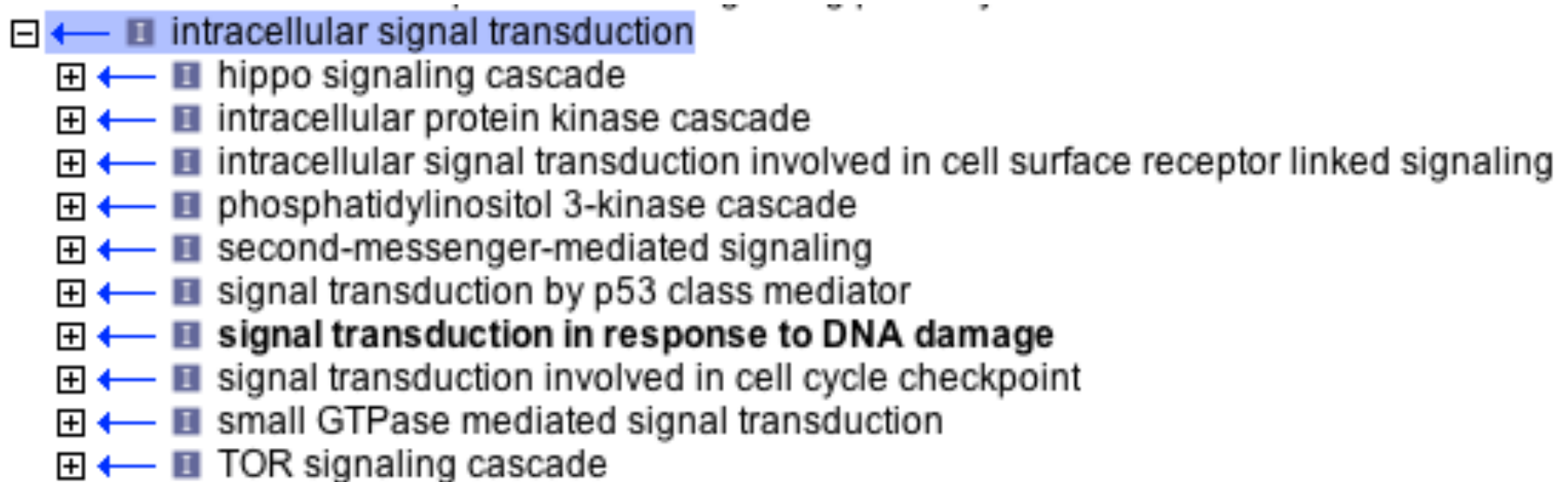
insulin receptor signaling pathway VIA TOR signaling cascade

[is_a] insulin receptor signaling pathway

[HAS_PART] TOR signaling pathway

Nomenclature

- ❖ Discussed options of ‘cascade, cassette, module’.
- ❖ Concern that ‘cascade’ implied amplification of a signal.
- ❖ Decided to stick with ‘**cascade**’ for intracellular ‘modules’ in GO.



Receptors

RECEPTORS

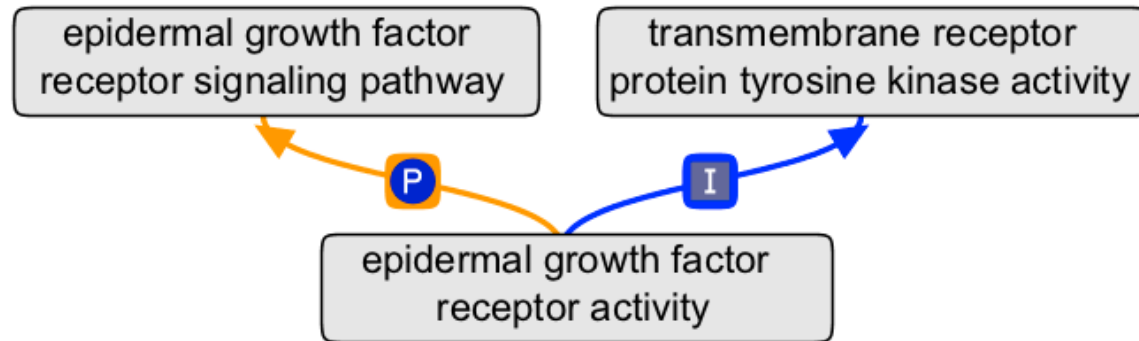
- ❖ Q: *Should receptors be split into those that signal and those that transport?*
- ❖ Q: *Are ion-gated ligand channels (eg P2X receptor) receptors?*
- ❖ Q: *How does receptor-mediated endocytosis fit with signaling?*

- ❖ AI: Rename ‘receptor activity ; GO:0004872’ to ‘signaling receptor activity’
- ❖ AI: Receptor activity should stay in function.
- ❖ AI: Rename ‘insulin receptor activity’ to ‘insulin-activated receptor activity’.
- ❖ AI: Add comment: “This does not refer to receptors such as transporters, “mop-up” receptors, decoy receptors, adhesion receptors, importin receptor, nutrient receptors”, to make clear that there are other types of receptors.

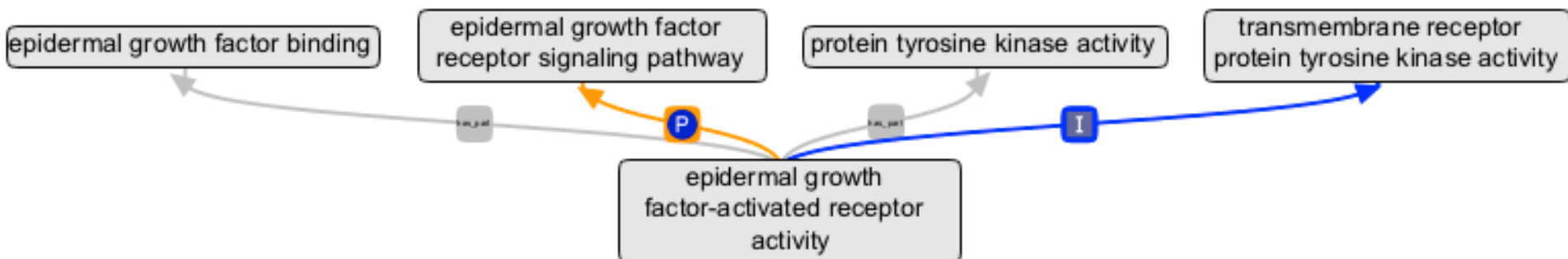
TO DO: Need to work out how signaling and RME fit together. Is there a signaling step in RME? Are receptors involved in RME, signaling receptors? Are they passing the signal on or just internalizing it?

Receptors and HAS_PART

current:



proposed:



Curation Manual:

*Ontology development together with
annotation*

Signaling Curation Manual

LIGAND AND RECEPTOR					
COMPONENT	ROLE	UNIPROT AC (human unless stated otherwise)	PMID (R = review)	SUGGESTED GO TERMS FOR ANNOTATION	NOTES
insulin	ligand	P01308	PMID:11238471 (R)		
insulin receptor	receptor	P06213	PMID:2986534 (R)		

Something to consider....

Mammals have an insulin receptor and an insulin-like growth factor receptor to bring about changes in metabolism and growth, respectively. *Drosophila* have one receptor for both 'jobs', which binds to insulin-like peptides (DILPs) rather than insulin. Nomenclature-wise, it is termed the *Drosophila* insulin receptor. Does it have 'insulin receptor activity' and is it involved in the 'insulin receptor signalling pathway'? We should bear in mind that GO is required for some curation conventions.

DOWNSTREAM SIGNALING MOLECULES (SIGNALING VIA IRS-1)

COMPONENT	ROLE	UNIPROT AC (human unless stated otherwise)	PMID (R = review)	SUGGESTED GO TERMS FOR ANNOTATION	NOTES
IRS1	Phosphorylated by active insulin receptor	P35568	PMID:9609109 (R)		
p85	regulatory subunit of PI3kinase	P27986 (A) O00459 (B)	PMID:9677303 (R) PMID:8909804 (R) PMID:10209156 (R)		
p110	catalytic subunit of PI3kinase	P42336	PMID:9677303 (R) PMID:8909804 (R) PMID:10209156 (R)		
PDK1	Activates Akt/PKB	O15530	PMID:9094314 PMID:9247112 (R)		
PDK2	Activates Akt/PKB	Q6A1A2	PMID:16014356 (R)		
Akt/PKB	Ser/Thr kinase with many downstream targets.	P31749	PMID:20517722 (R)		

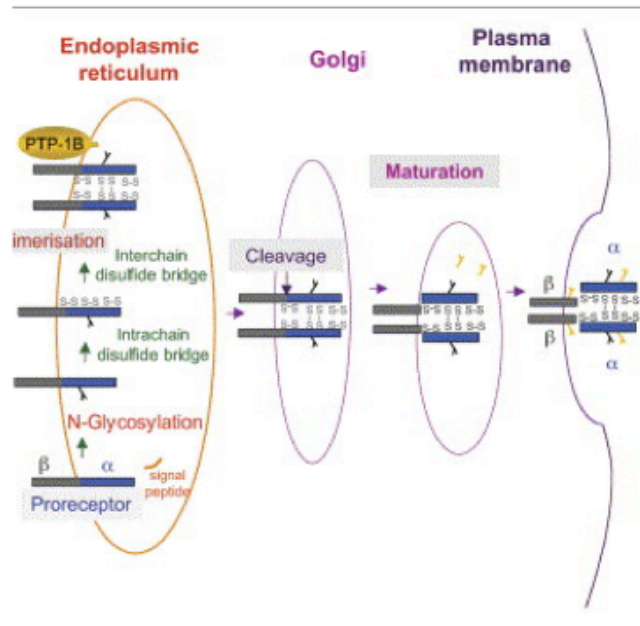
2. RECEPTOR EVENTS UPSTREAM OF LIGAND-RECEPTOR BINDING

2.1 INSULIN RECEPTOR SYNTHESIS

- ❖ 1/ Transcription of the insulin receptor gene
 - One (large) gene.
 - Both the alpha subunit (binds insulin) and the beta subunit (spans the membrane and contains Tyr kinase activity) are encoded by a single mRNA.
- ❖ 2/ Translation of the insulin receptor cDNA into protein

2.2 INSULIN RECEPTOR PROCESSING

- ❖ 3/ Processing of the proreceptor to form a functional receptor.



- Intramolecular disulfide bonds are formed rapidly (probably as a co-translation event).
 - interchain disulfide bond formation results in dimerization.
- The proreceptor dimer then undergoes proteolytic cleavage and carbohydrate maturation
- Appears at the plasma membrane as the mature tetrameric $\alpha_2\beta_2$ receptor.

Future Edits

- ❖ Put the proposals in place for receptors, non 1:1 receptor binding.
- ❖ Finalise how to handle intracellular signaling.
- ❖ Coordinate with Reactome to standardize which steps are included in the pathway.

Strategy: work through a pathway at a time.