# CACAO Training part I

TAMU 2013 BICH489-501/2

# Tues Aug 27, 2013

- What are we doing this semester?
- Big picture
- What is CACAO?
- Housekeeping

#### Overview of the semester

- Training
  - Today: Training
  - Sept 3: Practice papers
- Annotation
  - Sep 8– Nov 17: 5 rounds of finding papers, reading, doing annotation
    - Class time for meeting with your team and getting in-person help!
- Conclusion
  - Nov 19-26:Overview of what we did. Prizes?

## Big picture

- This class is about understanding how scientists
  - Infer the function of genes
  - Share knowledge with the larger community
- We will learn this by hands-on experience through CACAO
- Warnings:
  - This will be confusing at first. Stick with it!
  - Budget your time. This is not a class where you can cram at the end.

### What is CACAO?

- Community Assessment of Community
  Annotation with Ontologies (CACAO)
  - Annotation of gene function
  - Competition
    - Within this class
    - Between TAMU and other schools (BTHO everyone!)
    - Rules next week

#### **Annotation**

 Annotation: a note that is made while reading any form of text

- For genome biology,
  - Nucleotide level: Where the genes are in the genome
  - Protein level: What their functions are

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# Why do functional annotation?

#### Allow us to:

- Infer the function of genes
  - Related by common descent
  - Related by similar expression patterns
  - Related by phylogenetic profiles
  - ...

#### Allow us to:

- Understand the capabilities of organisms' genomes
- Understand patterns of gene expression
  - In different environments
  - In different tissues
  - In disease states
  - ...

# Where do functional annotations come from?

Journal home > Archive > Letters to Nature > Abstract

#### Letters to Nature

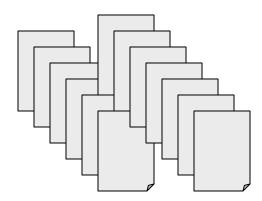
Nature 425, 628-633 (9 October 2003) | doi: 10.1038/nature02030

#### Basal body dysfunction is a likely cause of pleiotropic Bardet–Biedl syndrome

Stephen J. Ansley<sup>1,8</sup>, Jose L. Badano<sup>1,8</sup>, Oliver E. Blacque<sup>1,8</sup>, Josephine Hilf, Bethan E. Hoskins<sup>1,4</sup>, Germen C. Leitkn<sup>1,4</sup>, Jun Chil Kim<sup>1</sup>, Alson J. Ross<sup>2</sup>, Erica R. Echers<sup>3</sup>, Tanya M. Tesovich<sup>1</sup>, Allan K. Mah<sup>3</sup>, Robert C. Johnsen<sup>3</sup>, John C. Cavender<sup>2</sup>, Richard Alan Lewis<sup>3,6</sup>, Michel R. Leroux<sup>3</sup>, Philip L. Beales<sup>4</sup> and fitchols Katanis<sup>1,1,4</sup>

Bardet – Biedl syndrome (BBS) is a genetically heterogeneous disorder characterized primarily by retinal dystrophy, obesity, polydactyly, renol malformations and learning disabilities. Although five BBS genes have been cloned 1-2, 2-3, 4-5, 6, the molecular basis of this syndrome remains elusive. Here we show that BBS is probably caused by a defect at the basal body of ciliated cells. We have cloned a new BBS gene, BBSS, which encodes a protein with a prokaryotic domain, pilf; involved in pilus formation and twitching mobility. In one family, a protein probable swith randomization of left – right body axis symmetry, a known defect of the nodal cilium. We have also found that BBSB localizes specifically to ciliated structures, such as the connecting cilium of the retina and columnar processes of the control of the connecting cilium of the retina and columnar columnary of the connecting cilium of the retina and columnary columnary of the connecting cilium of the retina and columnary columnary of the connecting cilium of the retina and columnary columnary of the connecting cilium of the retina and columnary columnary of the connecting cilium of the retina and columnary columnary of the retination of the columnary of the columnary

#### Literature



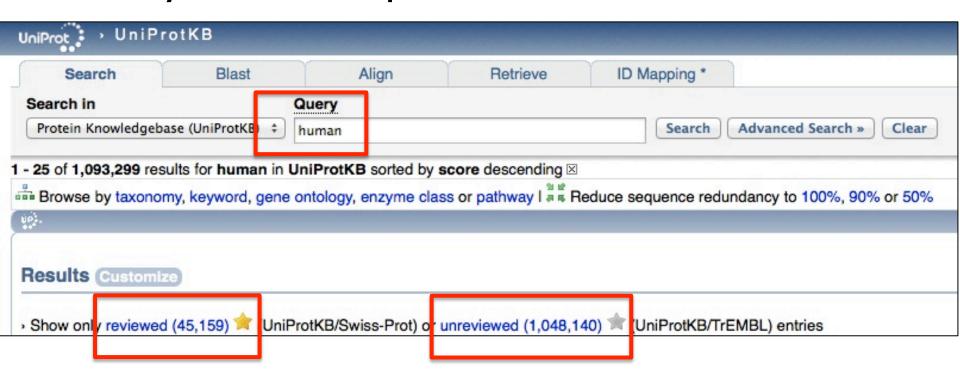
**Datasets** 



Biocurators (rate limiting)

### Databases need help!

- >21 million peer-reviewed articles in PubMed
- Many millions of proteins recorded in UniProt



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### What to annotate

- You can start with a paper
  - Find the proteins discussed
  - Start with a GO term
- You can start with a protein
  - Find papers about the protein
- Either way, don't get stuck on what you started with
  - Your first paper may not have experiments about function
  - Reading about your initial protein may lead you to better information about other proteins

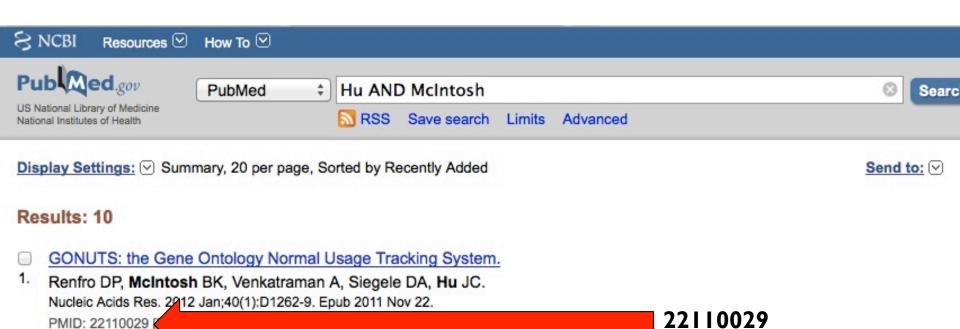
# Starting with a paper

- Need a scientific paper with experimental data
  - No review articles, no books, no textbooks, no wikipedia articles, no class notes...
  - BUT you should start with those!
  - DON'T start with a random PubMed search

# Starting with a paper

- Need a scientific paper with experimental data
  - You will need the PMID number

Related citations



# Getting the full text

- The abstract is not enough
  - But may be enough to reject a paper!!!
- Some papers are open access
  - Pubmed Central
  - Journal sites
    - TAMU pays for lots of subscriptions
    - Access from off campus via <a href="http://library.tamu.edu">http://library.tamu.edu</a>
      - E-journals
      - Citation search takes PMIDs

Good science ≠ good for annotation

# Second Extracellular Loop of Human Glucagon-like Peptide-1 Receptor (GLP-1R) Differentially Regulates Orthosteric but Not Allosteric Agonist Binding and Function\*5

Received for publication, September 30, 2011, and in revised form, November 29, 2011 Published, JBC Papers in Press, December 6, 2011, DOI 10.1074/jbc.M111.309369

Cassandra Koole<sup>‡</sup>, Denise Wootten<sup>‡</sup>, John Simms<sup>‡</sup>, Emilia E. Savage<sup>‡</sup>, Laurence J. Miller<sup>§</sup>, Arthur Christopoulos<sup>‡1</sup>, and Patrick M. Sexton<sup>‡2</sup>

From the <sup>‡</sup>Drug Discovery Biology, Monash Institute of Pharmaceutical Sciences and Department of Pharmacology, Monash University, Parkville, Victoria 3052, Australia and the <sup>§</sup>Department of Molecular Pharmacology and Experimental Therapeutics, Mayo Clinic, Scottsdale, Arizona 85259

Background: The ECL2 of the GLP-1R is critical for GLP-1 peptide-mediated selective signaling.

**Results:** Mutation of most ECL2 residues to alanine results in changes in binding and/or efficacy of oxyntomodulin and exendin-4 but not allosteric agonists.

**Conclusion:** ECL2 of the GLP-1R has ligand-specific as well as general effects on peptide agonist-mediated receptor activation. **Significance:** This work provides insight into control of family B GPCR activation transition.

Good science ≠ good for annotation

# Robust design and optimization of retroaldol enzymes

Eric A. Althoff,<sup>1,2</sup> Ling Wang,<sup>1</sup> Lin Jiang,<sup>1,3</sup> Lars Giger,<sup>4</sup> Jonathan K. Lassila,<sup>5</sup> Zhizhi Wang,<sup>1</sup> Matthew Smith,<sup>1</sup> Sanjay Hari,<sup>1</sup> Peter Kast,<sup>4</sup> Daniel Herschlag,<sup>5</sup> Donald Hilvert,<sup>4</sup> and David Baker<sup>1</sup>\*

<sup>&</sup>lt;sup>1</sup>Department of Biochemistry, University of Washington and HHMI, Seattle, Washington 98195

<sup>&</sup>lt;sup>2</sup>Arzeda Corp., Seattle, Washington 98102

<sup>&</sup>lt;sup>3</sup>Department of Biological Chemistry, UCLA, Los Angeles, California 90095

<sup>&</sup>lt;sup>4</sup>Laboratory of Organic Chemistry, ETH Zurich, 8093 Zurich, Switzerland

<sup>&</sup>lt;sup>5</sup>Department of Biochemistry, Stanford University, Stanford, California 94305

Good science ≠ good for annotation

Short Article



# Vitamin C Enhances the Generation of Mouse and Human Induced Pluripotent Stem Cells

Miguel Angel Esteban,<sup>1,6</sup> Tao Wang,<sup>1,6</sup> Baoming Qin,<sup>1,6</sup> Jiayin Yang,<sup>1</sup> Dajiang Qin,<sup>1</sup> Jinglei Cai,<sup>1</sup> Wen Li,<sup>1</sup> Zhihui Weng,<sup>1</sup> Jiekai Chen,<sup>1</sup> Su Ni,<sup>1</sup> Keshi Chen,<sup>1</sup> Yuan Li,<sup>1</sup> Xiaopeng Liu,<sup>1</sup> Jianyong Xu,<sup>1</sup> Shiqiang Zhang,<sup>1</sup> Feng Li,<sup>1</sup> Wenzhi He,<sup>1</sup> Krystyna Labuda,<sup>2</sup> Yancheng Song,<sup>3</sup> Anja Peterbauer,<sup>4</sup> Susanne Wolbank,<sup>2</sup> Heinz Redl,<sup>2</sup> Mei Zhong,<sup>5</sup> Daozhang Cai,<sup>3</sup> Lingwen Zeng,<sup>1</sup> and Duanqing Pei<sup>1,\*</sup>

<sup>1</sup>Stem Cell and Cancer Biology Group, Key Laboratory of Regenerative Biology, South China Institute for Stem Cell Biology and Regenerative Medicine, Guangzhou Institutes of Biomedicine and Health, Chinese Academy of Sciences, Guangzhou 510663, China

21 udwig Roltzmann Institute for Clinical and Experimental Traumatology, Austrian Cluster for Tissue Regeneration, Vienna 1200, Austria

Good science ≠ good for annotation

**10624** • The Journal of Neuroscience, August 11, 2010 • 30(32):10624 – 10638

Neurobiology of Disease

# Excess Phosphoinositide 3-Kinase Subunit Synthesis and Activity as a Novel Therapeutic Target in Fragile X Syndrome

Christina Gross,<sup>1</sup> Mika Nakamoto,<sup>2\*</sup> Xiaodi Yao,<sup>1\*</sup> Chi-Bun Chan,<sup>3</sup> So Y. Yim,<sup>1</sup> Keqiang Ye,<sup>3</sup> Stephen T. Warren,<sup>2,4,5</sup> and Gary J. Bassell<sup>1,6</sup>

Departments of <sup>1</sup>Cell Biology, <sup>2</sup>Human Genetics, <sup>3</sup>Pathology and Laboratory Medicine, <sup>4</sup>Biochemistry, <sup>5</sup>Pediatrics, and <sup>6</sup>Neurology, Emory University School of Medicine, Atlanta, Georgia 30322

# Suggested papers

- We will post some suggested papers to use as starting points
  - Not required
  - Use if you are struggling or try these first before tackling more complicated/interesting things

# Finding proteins

- Search UniProt for something interesting
- Look in UniProt for the protein(s) in the paper you are reading.

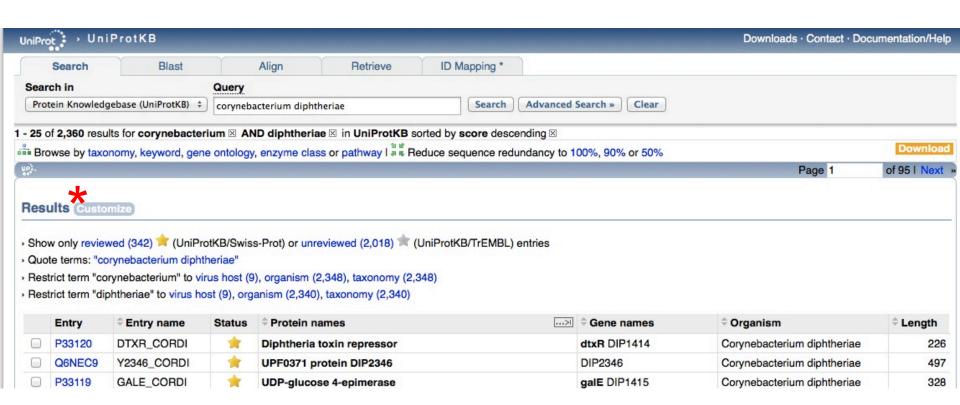
No matter what, you will need to find the protein's accession on UniProt (http://uniprot.org)

Use that accession to make a page for that protein on GONUTS (http://gowiki.tamu.edu)

Add your GO annotations to the protein's page on GONUTS

# UniProt (http://www.uniprot.org)

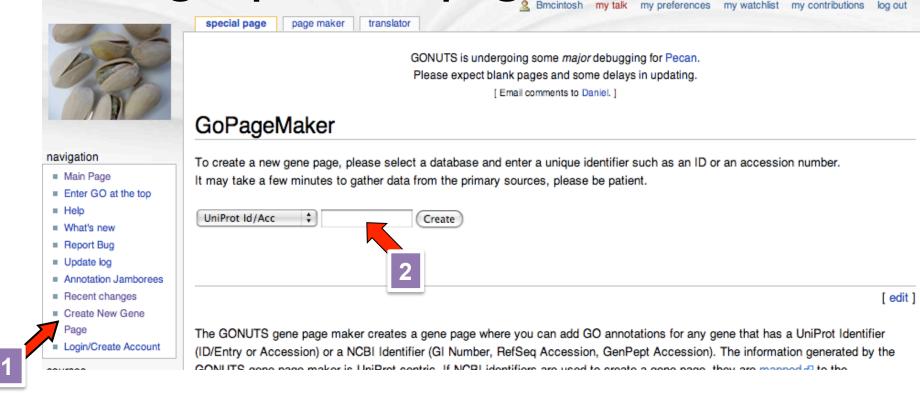
- If you have a paper, look for an accession
- Otherwise, search by name/keyword



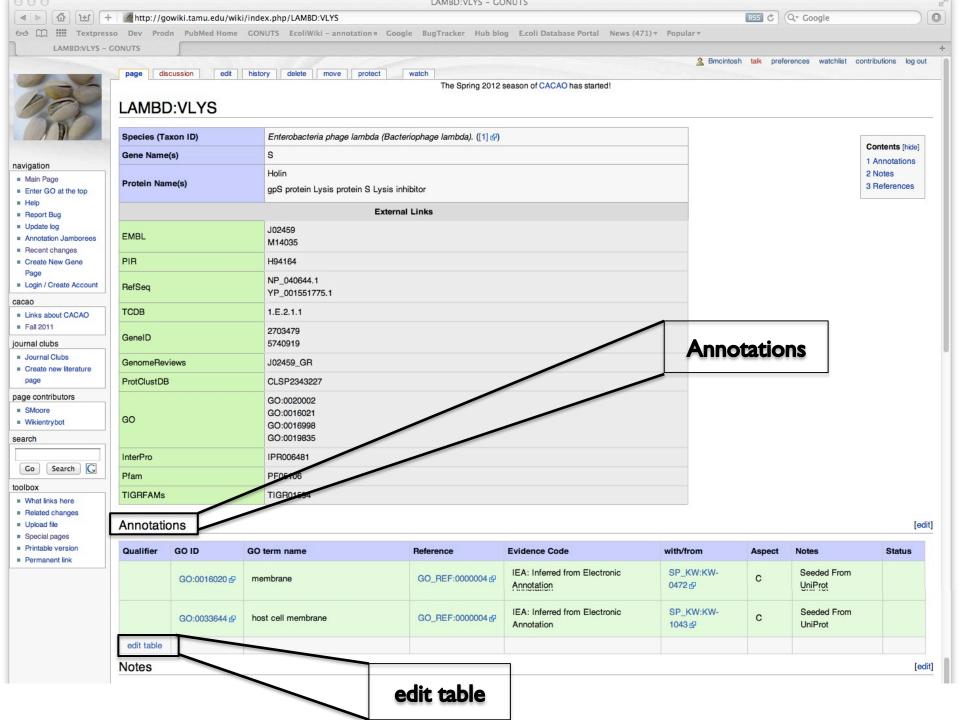
### Make sure you have the right protein

- Right species/strain
- Not a fragment
- Sometimes UniProt has multiple entries for the same protein
  - Gold star = SwissProt = reviewed
  - Blank star = TrEMBL = computational entry
- Sometimes the protein you want is not in UniProt
  - May want to find another paper/protein
- Ask for help
  - OK to email the UniProt help desk
  - check your reasoning with us!

# Making a protein page in GONUTS



- GoPageMaker will:
  - Check if the page exists in GONUTS & take you there if it does.
  - Make a page if it does not exist in GONUTS already & pull all of the annotations from UniProt into a table that you can edit.
- Make as many protein pages as you would like!



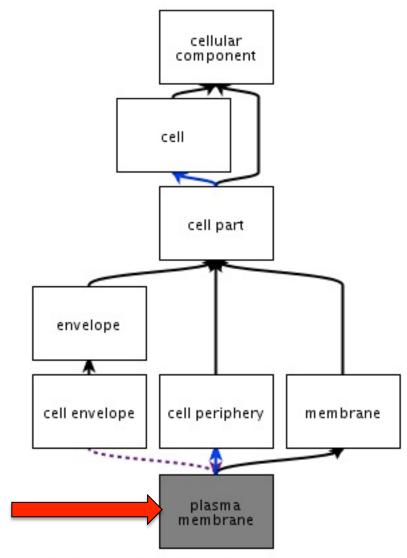
### **Functional Annotation**

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# GO (Gene Ontology) Annotations

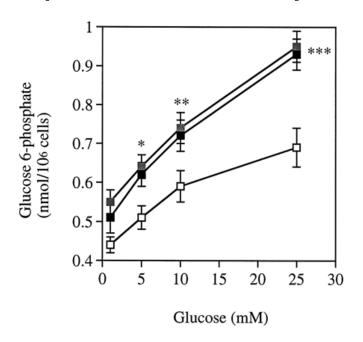
- 3 aspects (ontologies):
  - Molecular Function
  - Biological Process
  - Cellular Component
- Controlled vocabulary
  - ID number for computers
  - Name and definition for humans
- Relationships



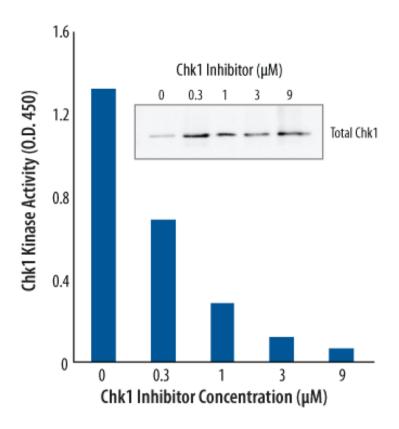
GO:0005886

### Molecular Function

 activities or what a protein can do by itself



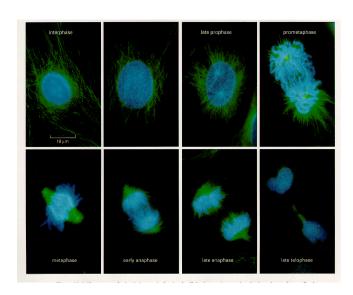
GO:0004347 hexokinase activity



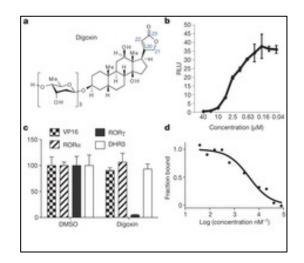
GO:0016301 Kinase activity

# **Biological Process**

- a commonly recognized series of events
  - Including, but not just biochemical pathways



GO:0051301 cell division



GO:0006351 transcription, DNA dependent



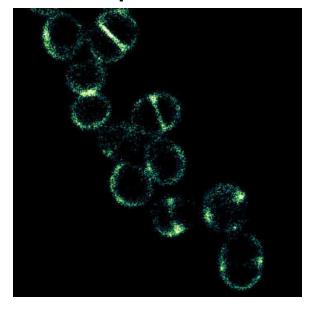
GO:0009405 pathogenesis

# Cellular Component

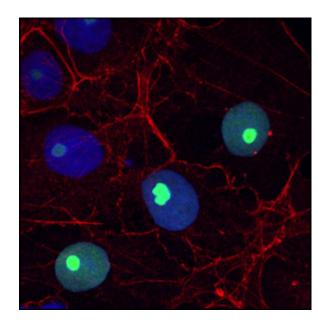
- where a gene product acts
  - Subcellular location
  - Multicomponent complex



GO:0005739 mitochondrion



GO:0009274 peptidoglycan-based cell wall



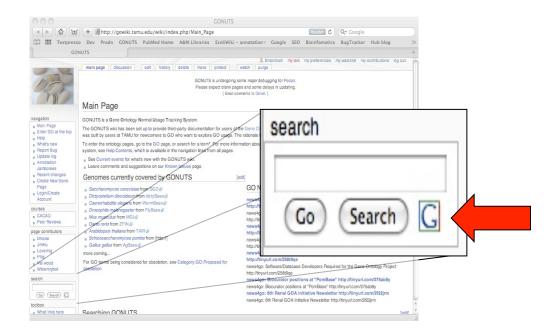
GO:0005840 ribosome

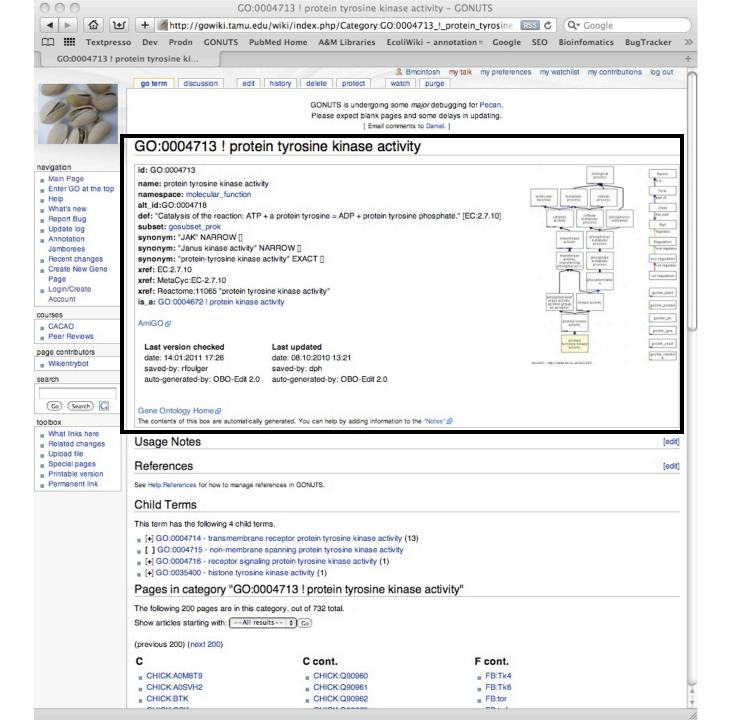
# Finding GO terms

GONUTS: <a href="http://gowiki.tamu.edu">http://gowiki.tamu.edu</a>

QuickGO: <a href="http://www.ebi.ac.uk/QuickGO">http://www.ebi.ac.uk/QuickGO</a>

• AmiGO: <a href="http://amigo.geneontology.org">http://amigo.geneontology.org</a>





#### GO:0004713! protein tyrosine kinase activity

id: GO:0004713

name: protein tyrosine kinase activity

namespace: molecular\_function

alt id:GO:0004718

def: "Catalysis of the reaction: ATP + a protein tyrosine = ADP + protein tyrosine phosphate." [EC:2.7.10]

subset: gosubset\_prok

synonym: "JAK" NARROW []

synonym: "Janus kinase activity" NARROW []

synonym: "protein-tyrosine kinase activity" EXACT []

xref: EC:2.7.10

xref: MetaCyc:EC-2.7.10

xref: Reactome:11065 "protein tyrosine kinase activity"

is a: GO:0004672 ! protein kinase activity

#### AmiGO @

Last version checked

Last updated date: 14:01:2011 17:26

saved-by: rfoulger

saved-by: dph

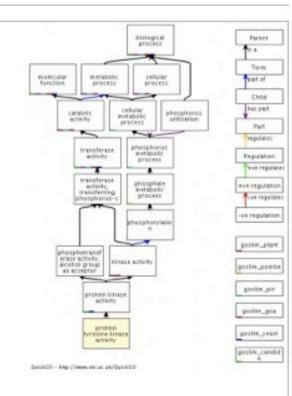
auto-generated-by: OBO-Edit 2.0

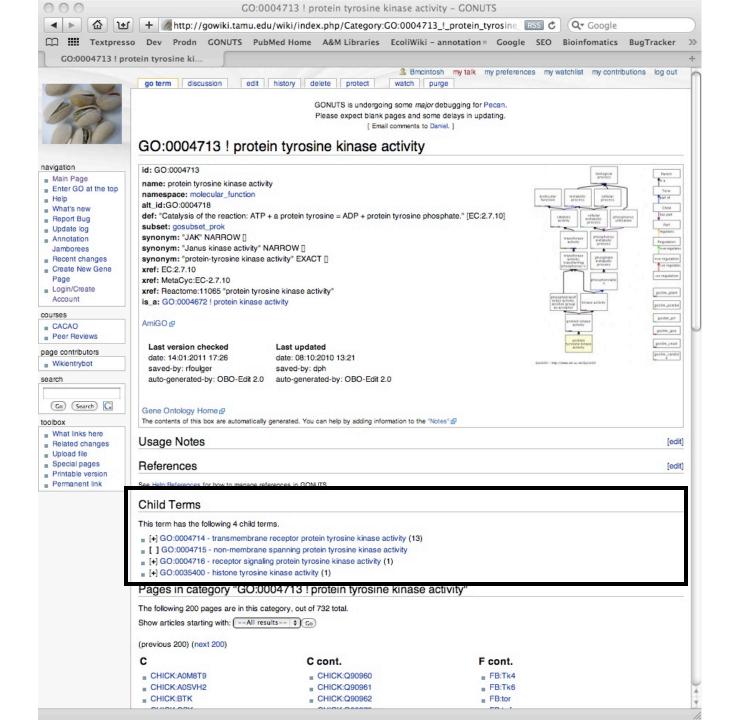
date: 08:10:2010 13:21

auto-generated-by: OBO-Edit 2.0

#### Gene Ontology Home @

The contents of this box are automatically generated. You can help by adding information to the "Notes" @





# Strategies

- Search for a keyword and browse the ontology for the right term
- Look at terms suggested by others for your protein
  - Computational with the IEA evidence code
  - Curators with TAS or IC
- Look at terms used for homologous proteins in model organisms

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#### Evidence Codes for CACAO

- Evidence codes describe the type of work or analysis done by the authors
  - IDA: Inferred from Direct Assay
  - IMP: Inferred from Mutant Phenotype
  - IGI: Inferred from Genetic Interaction
  - ISO: Inferred from Sequence Orthology
  - ISA: Inferred from Sequence Alignment
  - ISM: Inferred from Sequence Model
  - IGC: Inferred from Genomic Context
- Expert biocurators get to use others, but we restrict them for CACAO. If it's not one of these 7, your annotation is incorrect!!!
- http://gowiki.tamu.edu/wiki/index.php/evidence\_codes

#### Evidence Codes for CACAO

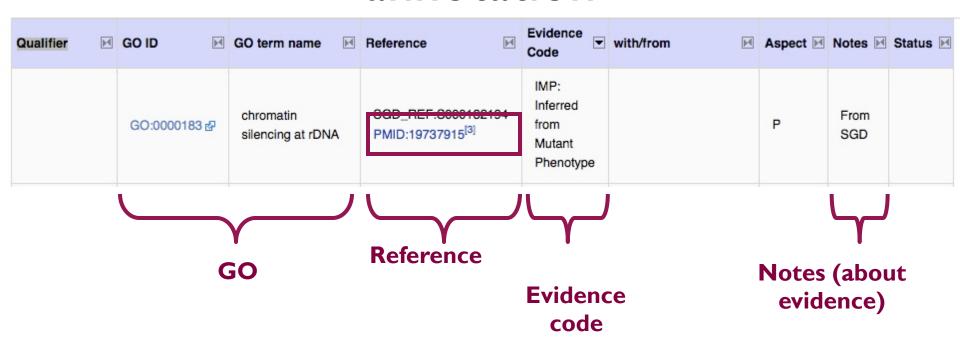
- Picking the right evidence code is important
- Use the evidence code decision tree
- Use the evidence code guidelines at the GO consortium website:
  - http://www.geneontology.org/GO.evidence.shtml
- Discuss!

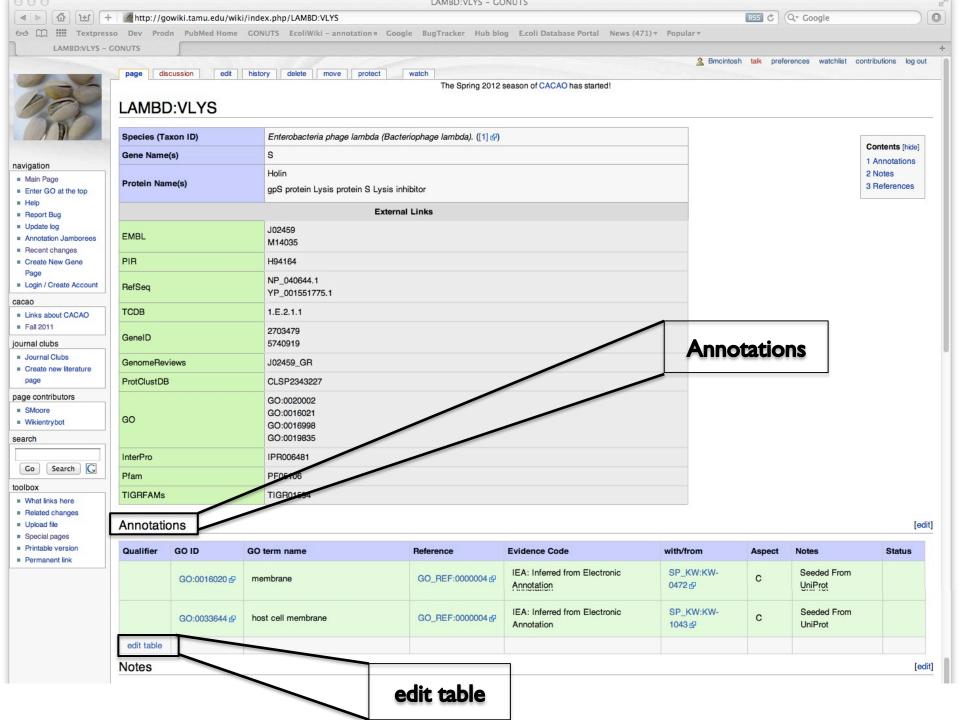
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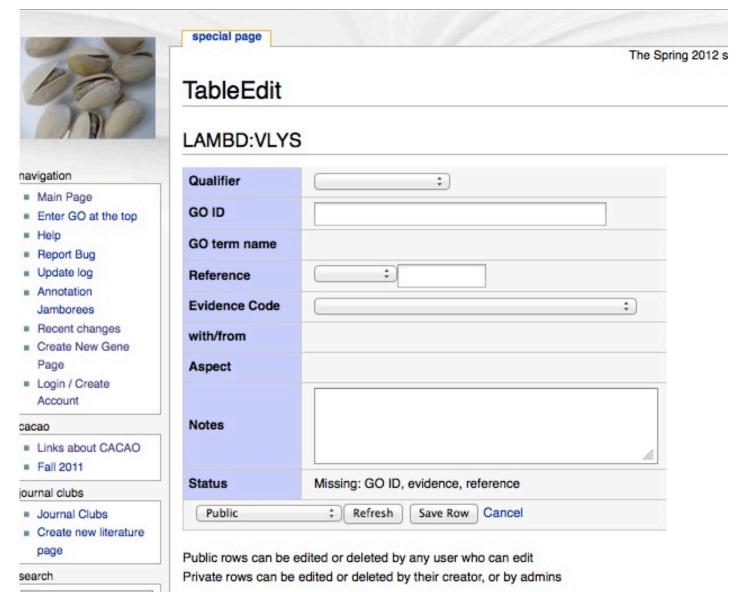
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# 4 REQUIRED parts of EVERY GO annotation

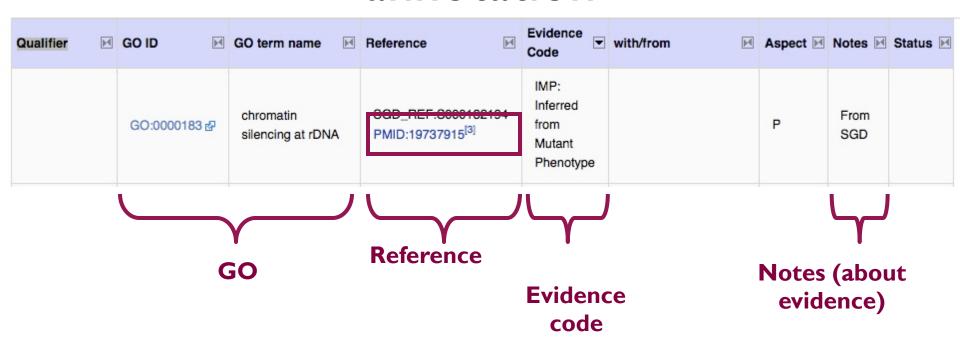




# Entering/editing annotations



# 4 REQUIRED parts of EVERY GO annotation



### 2 other parts that may be required...



## Example paper

http://www.ncbi.nlm.nih.gov/pubmed/8227000