

PomBase advanced search and FungiDB cytoscape

In this exercise we will identify fungal genes that when mutated increase resistance to the antifungal amphotericin, cross reference the gene list with the metabolic pathways in FungiDB, and review the available transcriptomic data in hypoxia conditions using Paint function in CytoScape.

- Go to <https://www.pombase.org/query>
- Using the phenotype filter, search for “resistance to amphotericin B” and click submit, this should match 8 genes
- Click on the hyperlinked result at the bottom of the advanced search, and then on the ‘visualize’ button

The screenshot shows the PomBase advanced search interface. At the top, there is a blue button labeled '<- Advanced search' and a row of social media icons (email, Twitter, Facebook, GitHub, Google+, LinkedIn) followed by a link 'Contact curators ...'. Below this, a box indicates 'Result: 8 genes'. The main content area is titled 'Results for: resistance to amphotericin B (FYPO:0000070) [single allele genotypes]'. It features four blue buttons: 'Visualise', 'Slim', 'Select subset', and 'Download ...'. Below the buttons is a table with three columns: 'Gene name', 'Systematic ID', and 'Product'. The first row of the table shows the gene 'ada2' with systematic ID 'SPCC24B10.08c' and product 'SAGA complex subunit Ada2'.

Gene name	Systematic ID	Product
ada2	SPCC24B10.08c	SAGA complex subunit Ada2

- “Sort” on GO Process

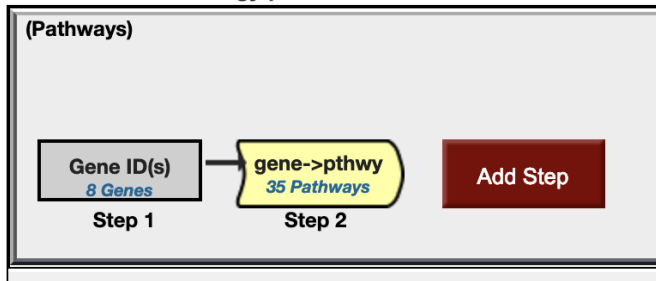
The screenshot shows the PomBase visualization interface for the 8 genes. The title is 'Visualising 8 genes'. Below the title, it says 'Visualisation for: resistance to amphotericin B (FYPO:0000070) [single allele genotypes]'. On the left side, there is a list of filters with checkboxes and 'sort' links: 'Deletion viability: [checked] [sort]', 'Budding yeast ortholog: [checked] [sort]', 'Human ortholog: [checked] [sort]', 'Transmembrane domain: [checked] [sort]', 'GO process: [checked] [sort]', 'GO component: [checked] [sort]', 'GO function: [checked] [sort]', 'Characterisation status: [checked] [sort]', 'Taxonomic distribution: [checked] [sort]', and 'Protein length: [checked] [sort]'. Below the filters are links for 'Documentation' and 'Download image ...'. The main visualization area shows a horizontal bar chart with 10 categories: 'Deletion viability', 'Budding yeast ortholog', 'Human ortholog', 'Transmembrane domain', 'GO process', 'GO component', 'GO function', 'Characterisation status', 'Taxonomic distribution', and 'Protein length'. Each category has a bar with colored segments representing different values. A legend on the right side explains the colors: 'Deletion viability' (blue for variable, green for viable), 'Budding yeast ortholog' (green for present), 'Human ortholog' (red for absent, green for present), 'Transmembrane domain' (black for present, white for absent), and 'GO process' (red for gene expression, yellow for membrane organization, black for lipid metabolic process).

Q1: Which GO Process is common to 5 genes?

Q2: Which one gene does not have a reported human ortholog?

- “Finish” visualization and click the button to “download...” the systematic IDs of the genes
- Go to fungidb and perform a GeneID search (<https://fungidb.org/fungidb/showQuestion.do?questionFullName=GeneQuestions.GeneByLocusTag>)
- Add a step to “transform to pathway” (use default settings)

Hide search strategy panel



- Open the Metacyc “Superpathway of ergosterol biosynthesis I”
- Evaluate transcriptomic evidence in hypoxia conditions for *A. fumigatus* using microarray paint operator in Cytoscape

Search My Strategies My Basket (0) My Data Sets BETA Tools Data Summary Downloads Community Analyze My Experiment

.1 Metabolic pathways

▼ Cytoscape Drawing

NOTE Click on nodes for more info. Nodes highlighted in Identifiers which were used to map your results to this pa Here is the list of all Identifiers returned by your search re

File Layout Paint Enzymes

Experiment Selector

Paint

expand all | collapse all

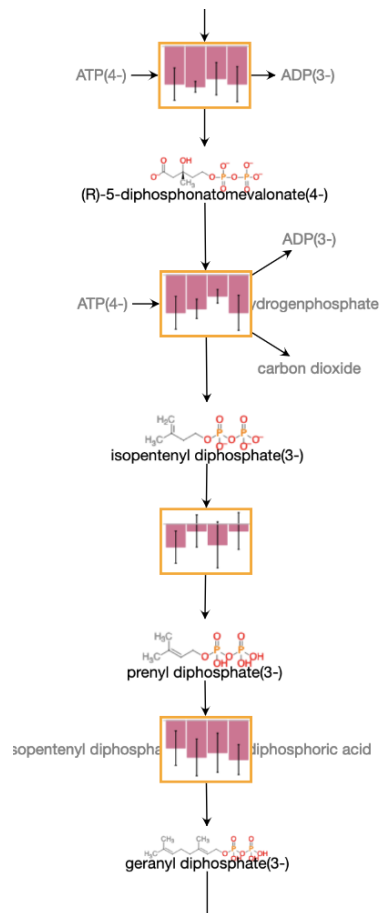
Search for Experiments

▼ Transcriptomics

▼ DNA Microarray

- A. fumigatus Af293 Aspergillus fumigatus hypoxia response transcriptome (Barker et al.)
- C. albicans SC5314 Antifungal Benzimidazole Derivative Response (Rupp)
- C. gattii WM276 Comparison of gene expression in Cryptococcus gattii clinical isolates (Ma et al.)
- C. immitis RS Gene expression changes during mycelia differentiation in Coccidioides immitis (Viriyakosol et al.)
- C. neoformans var. grubii H99 Flucytosine Susceptibility (Bahn)
- F. graminearum PH-1 Fusarium graminearum transcriptome during symptomless and symptomatic wheat infection (Brown et al.)

Q3: Are genes involved in ergosterol biosynthesis typically downregulated or upregulated in hypoxia?



- Ergosterol biosynthesis is a very oxygen intensive process, so during hypoxic conditions this pathway is downregulated
- Amphotericin kills cells by binding ergosterol in the cell membrane and form pores through which ion can leak out of the cell.
- When ergosterol biosynthesis genes are knocked out, cells become resistant to amphotericin