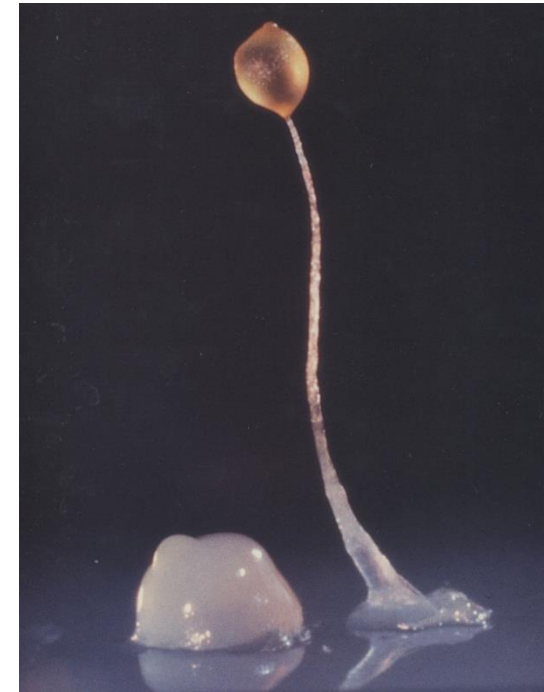
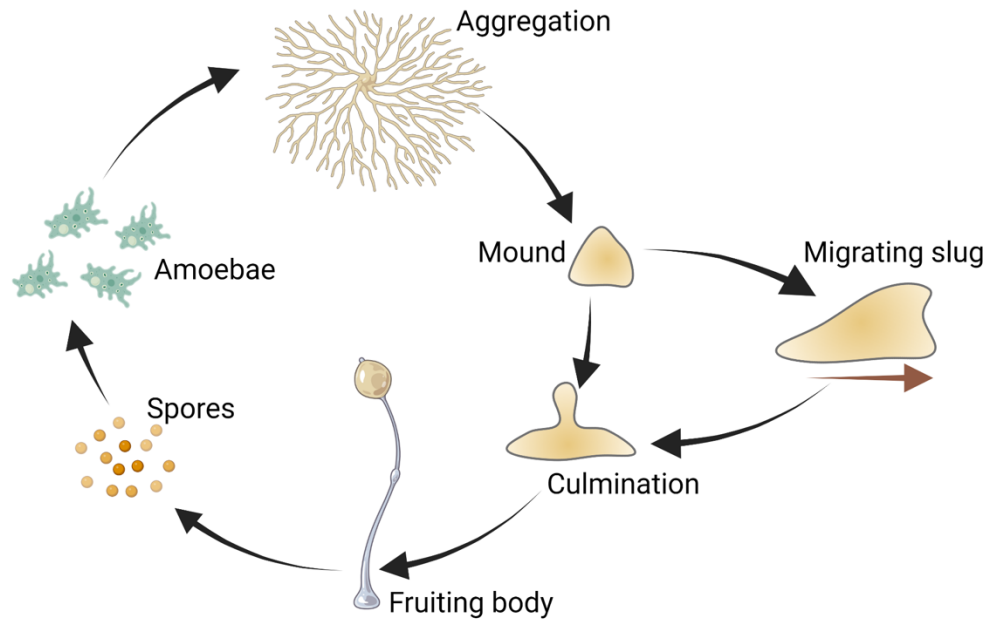


# Gene Ontology Enrichment Exercise

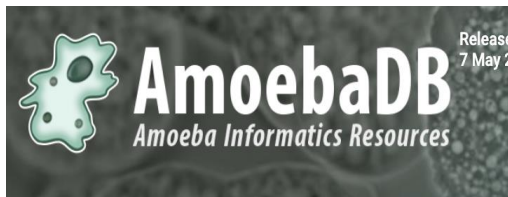
# Start with a set of regulated genes

- *Dictyostelium* (slime mold) is a model system for developmental biology
- Its life cycle transitions from single cells to multicellular structure



# Find RNA-Seq experiments in AmoebaDB.org

Choose Differential Expression (**DE**) in *D. discoideum* developmental program



**Search for...**

rna

Genes

- Gene models
  - Gene Model Characteristics
  - Unannotated Intron Junctions
- Transcriptomics
  - Microarray Evidence
  - RNA-Seq Evidence

## Identify Genes based on RNA-Seq Evidence

**Legend:** **DE** Differential Expression **FC** Fold Change **P** Percentile **SA** SenseAntisense

Filter Data Sets: 19 rows

Organism	Data Set	Choose a Search
<i>Acanthamoeba castellanii</i> str. Neff	Trophozoite transcriptome of <i>A. castellanii</i> (Wojtkawska et al.)	P
<i>Acanthamoeba castellanii</i> str. Neff	Transcriptome during encystment of <i>A. castellanii</i> Neff (Bernard C. et al. 2022)	DE FC P
<i>Dictyostelium discoideum</i> AX4	<i>D. discoideum</i> developmental program transcriptome (Parikh et al. 2010) <b>NEW</b>	DE FC P
<i>Dictyostelium discoideum</i> AX4	Intracellular <i>M. marinum</i> infection of <i>D. discoideum</i> (Kjellin et al. 2019)	DE FC P
<i>Dictyostelium discoideum</i> AX4	Intracellular <i>L. pneumophila</i> infection of <i>D. discoideum</i> (Kjellin et al. 2019)	DE FC P SA
<i>Dictyostelium purpureum</i> QSDP1	<i>D. purpureum</i> developmental program transcriptome (Parikh et al. 2010) <b>NEW</b>	DE FC P

# This experiment is a 24 hour timecourse of Dicty development

- Find genes that are upregulated in late stages (16-24 hours)
- Use default settings of 2-fold and p-value of 0.1
- How many genes do you find?

- What happens to the results if you increase the stringency to 4-fold upregulated and p-value of 0.01?
- How does this affect the GO enrichment?

## ? Reference Sample

- ☐ 0 hr
- ☐ 4 hr
- ☐ 8 hr
- ☐ 12 hr
- ☒ 16 hr
- ☐ 20 hr
- ☐ 24 hr
- ☐ NC4 prespore
- ☐ NC4 prestalk

## ? Comparator Sample

- ☐ 0 hr
- ☐ 4 hr
- ☐ 8 hr
- ☐ 12 hr
- ☐ 16 hr
- ☐ 20 hr
- ☒ 24 hr
- ☐ NC4 prespore
- ☐ NC4 prestalk

## ? Direction

up-regulated ▼

## ? fold difference >=

2

## ? adjusted P value less than or equal to

0.1

Get Answer

# Analyze the results for GO Enrichment

Gene Results | Genome View | **Analyze Results**


Genes: 1,723 | Transcripts: 1,727 | ☐ Show Only One Transcript Per Gene | ☐ Show only the Genes in my basket.

Rows per page: 500


Download

Gene ID	Transcript ID	Organism	Product Description
DDB_G0287293	DDB_G0287293	Dictyostelium discoideum AX4	spore germination protein A
DDB_G0287561	DDB_G0287561	Dictyostelium discoideum AX4	NADH:flavin oxidoreductase/NADH oxidase domain-containing
DDB_G0285443	DDB_G0285443	Dictyostelium discoideum AX4	unspecified product
DDB_G0285569	DDB_G0285569	Dictyostelium discoideum AX4	unspecified product
DDB_G0274337	DDB_G0274337	Dictyostelium discoideum AX4	unspecified product

Analyze your Gene results with a tool below.



Gene Ontology Enrichment



Metabolic Pathway Enrichment

kinase  
phosphatase  
exported  
membrane

Word Enrichment

## Gene Ontology Enrichment

Find Gene Ontology terms that are enriched in your gene result. [Read More](#)

### Parameters

- Organism** ? Dictyostelium discoideum AX4
- Ontology** ?  
☒ Biological Process  
☐ Cellular Component  
☐ Molecular Function
- Evidence** ?  
☒ Computed  
☒ Curated  
[select all](#) | [clear all](#)
- Limit to GO Slim terms** ? ☒ No  
☐ Yes
- P-Value cutoff** ? 0.05 (0 - 1)

Submit

Choose the **Biological Process** ontology  
Use default settings for the GO parameters

# Visualize functional groups

- What functions are enriched in the gene set?
- Visualize related functions with **Revigo**

Analysis Results:

139 rows

[Open in Revigo](#) [Show Word Cloud](#) [Download](#)

GO ID	GO Term	Genes in the bkgd with this term	Genes in your result with this term	Percent of bkgd genes in your result	Fold enrichment	Odds ratio	P-value	Benjamini	Bonferroni
GO:0030198	extracellular matrix organization	40	28	70.0	5.00	14.63	9.73e-16	9.67e-13	1.93e-12
GO:0043062	extracellular structure organization	40	28	70.0	5.00	14.63	9.73e-16	9.67e-13	1.93e-12
GO:0045229	external encapsulating structure organization	51	31	60.8	4.34	9.73	1.12e-14	7.39e-12	2.22e-11
GO:0072330	monocarboxylic acid biosynthetic process	78	27	34.6	2.47	3.30	3.40e-6	1.69e-3	6.75e-3
GO:0006633	fatty acid biosynthetic process	74	25	33.8	2.41	3.18	1.27e-5	4.08e-3	2.52e-2
GO:0046394	carboxylic acid biosynthetic process	141	39	27.7	1.98	2.39	1.44e-5	4.08e-3	2.86e-2
GO:0016053	organic acid biosynthetic process	141	39	27.7	1.98	2.39	1.44e-5	4.08e-3	2.86e-2
GO:0042060	wound healing	24	12	50.0	3.57	6.19	2.85e-5	6.29e-3	5.66e-2
GO:001778	plasma membrane repair	24	12	50.0	3.57	6.19	2.85e-5	6.29e-3	5.66e-2
GO:0043436	oxoacid metabolic process	390	83	21.3	1.52	1.71	4.04e-5	6.92e-3	8.03e-2
GO:0009653	anatomical structure morphogenesis	205	50	24.4	1.74	2.02	4.18e-5	6.92e-3	8.31e-2

