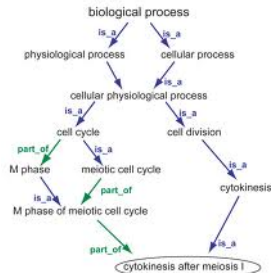


Annotation and enrichment analyses



Annotation packages, databases and ontologies

An **annotation database** (e.g., OMIM) annotates genes with functions or properties.

An **ontology** (e.g., GO) does this, and is additionally hierarchical.

A Bioconductor **annotation package** maps probes on an array to external IDs (Entrez gene), and to annotation databases and ontologies.

Enrichment analysis

Basic problem:

"Out of X total (genes/proteins) in a category, Y were (differentially expressed/selected). How significant is that?"

Answer: Fisher's Exact test, using hypergeometric distribution.

Fisher's exact test

Example. Are my differentially expressed (DE) genes enriched for angiogenesis (Ang) related genes?

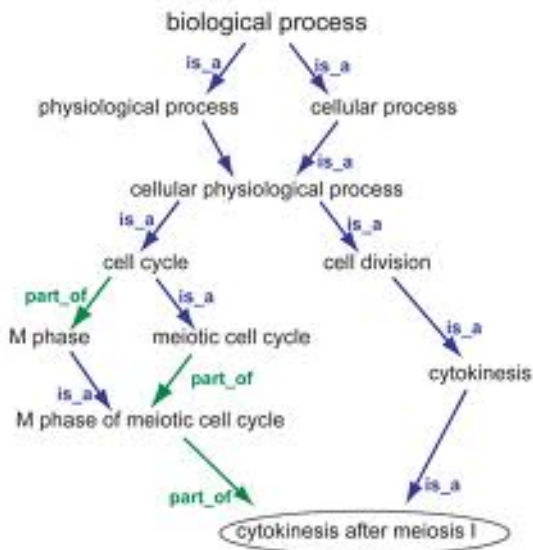
Requires a **contingency table**:

	Ang	Not Ang	Total
DE	35	165	200
Not DE	150	12000	12150
Total	185	12165	12350

Overview: enrichment analysis

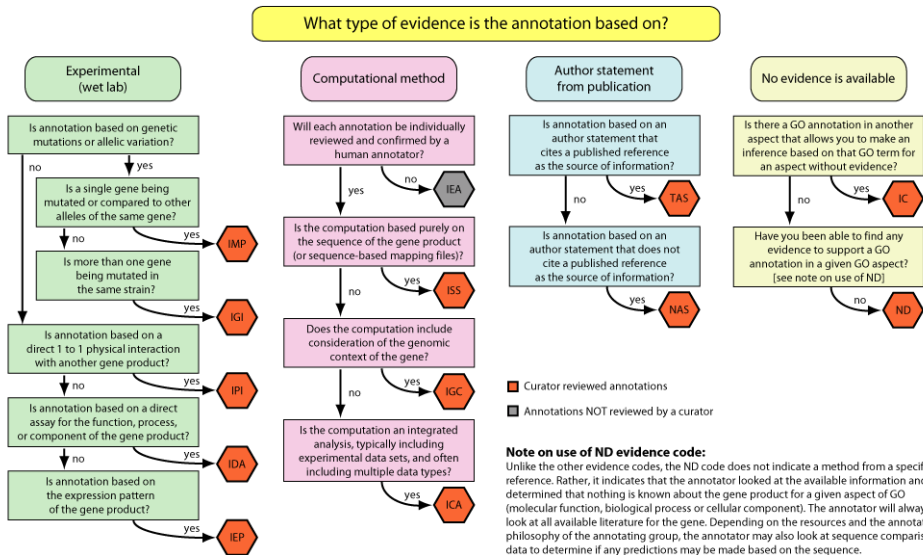
1. Map probes to annotations (possibly via intermediate IDs)
2. **Count annotations:**
 - within selected (DE) probes
 - in all probes tested
3. Get significance via Fisher's exact (or other) test
4. Map annotation IDs to annotation names
5. Wrap it all up in a data frame
6. Perform multiple hypothesis correction

Gene Ontology



Gene Ontology Evidence Codes

GO Evidence Code Decision Tree



DAVID: Easy GOEA

<http://david.abcc.ncifcrf.gov>

The screenshot shows the DAVID Functional Annotation Tool interface. The browser address bar displays david.abcc.ncifcrf.gov/tools.jsp. The page title is "DAVID: Functional Annotation Tool". The interface is divided into a left sidebar and a main content area.

Left Sidebar:

- Upload | List | Background
- Upload Gene List
- Demolist 1 Demolist 2
- Upload Help
- Step 1: Enter Gene List
- A Paste a list
- Clear
- Or
- B Choose From a File
- Choose File No file chosen
- Multi-List File ?
- Step 2: Select Identifier
- AFFYMETRIX_3PRIME_UT_ID
- Step 3: List Type
- Gene List
- Background
- Step 4: Submit List
- Submit List

Main Content Area:

Analysis Wizard

[Tell us how you like the tool](#)
[Contact us for questions](#)

← Step 1. Submit your gene list through left panel.

An example:

Copy/paste IDs to "box A" -> Select Identifier as "Affy_ID" -> List Type as "Gene List" -> Click "Submit" button

1007_s_at
1053_at
117_at
121_at
1255_g_at
1394_at
1316_at
1320_at
1405_i_at
1431_at
1438_at
1487_at
1494_f_at
1598_g_at

DAVID: Easy GOEA

The screenshot shows the DAVID Functional Annotation Tool interface. The browser address bar displays `david.abcc.ncifcrf.gov/tools.jsp`. The page has a dark background with an orange sidebar on the left and a main content area on the right.

DAVID: Functional Annotation Tool

Gene List Manager

Select to limit annotations by one or more species [Help](#)

Use All Species - Homo sapiens(14)

Select Species

List Manager [Help](#)

List_1

Select List to:

Use Rename Remove Combine Show Gene List

Analysis Wizard

[Tell us how you like the tool](#)
[Contact us for questions](#)

☒ **Step 1. Successfully submitted gene list**

Current Gene List: List_1
Current Background: Homo sapiens

Step 2. Analyze above gene list with one of DAVID tools

[Which DAVID tools to use?](#)

[Functional Annotation Tool](#)

- [Functional Annotation Clustering](#)
- [Functional Annotation Chart](#)
- [Functional Annotation Table](#)

[Gene Functional Classification Tool](#)

[Gene ID Conversion Tool](#)

[Gene Name Batch Viewer](#)

DAVID: Easy GOEA

