

V9 funktionelle Annotation

- Analyse von Gen-Expression
- Funktionelle Annotation: Gene Ontology (GO)
- Signifikanz der Annotation: Hypergeometrischer Test
- Annotationsanalysen z.B. mit NIH-Tool DAVID
- Ähnlichkeit von GO-Termen automatisch bestimmen
- OMIM-Datenbank

Ausgangslage

Daten aus Microarray-Analyse wurden ursprünglich als sehr „verrauscht“ angesehen.

Mittlerweile wurden jedoch sowohl die experimentellen Schritte wie auch die Datenauswertung gründlich verfeinert.

Microarray-Analyse ist daher heute eine (zwar teure, aber zuverlässige) Routine-Methode, die in allen großen Firmen verwendet wird.

Heute wird die MA-Analyse zunehmend durch RNA-seq ersetzt.

Die Datenaufbereitung kann in beiden Fällen folgende Schritte enthalten:
Normalisierung, Logarithmierung, Clustering, evtl. Ko-Expressionsanalyse,
Annotation der Genfunktion (Inhalt von V9).

Sehr wichtig ist es immer, die Signifikanz der Ergebnisse zu bewerten.

Gentleman et al. Genome Biology 5, R80 (2004)

Beispiel: differentielle Gen-Expression für ALL-Patienten

Input:

Genexpressionsdaten für 128 Patienten mit akuter lymphatischer Leukämie (ALL).

Alle ALL-Patienten haben chromosomal Veränderungen.

Der Therapieerfolg ist jedoch sehr unterschiedlich.

Hintergrundinformation:

- Eine Gruppe von Patienten (ALL1/AF4) hat eine genetische Translokation zwischen den Chromosomen 4 und 11.
- Eine zweite Gruppe von Patienten (BCR/ABL) hat eine genetische Translokation zwischen den Chromosomen 9 und 22.
- Die Krankheitsursachen + optimale Therapie können für die beiden Gruppen verschieden sein.

Ziel:

Identifiziere Gene, die zwischen den beiden Gruppen differentiell exprimiert werden.

Beispiel für die Anwendung der Bioconductor-Software (siehe Ref unten, bisher 6700 mal zitiert).

Gentleman et al. Genome Biology 5, R80 (2004)

Auswahl der differentiell exprimierten Gene

```
> f <- factor(as.character(eset$mol))
> design <- model.matrix(~f)
> fit <- lmFit(eset, design)
> fit <- eBayes(fit)
> topTable(fit, coef = 2)
```

Bioconductor
Kommandos

	ID	M	A	t	p-value	B
1016	1914_at	-3.1	4.6	-27	5.9e-27	56
7884	37809_at	-4.0	4.9	-20	1.3e-20	44
6939	36873_at	-3.4	4.3	-20	1.8e-20	44
10865	40763_at	-3.1	3.5	-17	7.2e-18	39
4250	34210_at	3.6	8.4	15	3.5e-16	35
11556	41448_at	-2.5	3.7	-15	1.8e-15	34
3389	33358_at	-2.3	5.2	-13	3.3e-13	29
8054	37978_at	-1.0	6.9	-10	6.5e-10	22
10579	40480_s_at	1.8	7.8	10	9.1e-10	21
330	1307_at	1.6	4.6	10	1.4e-09	21

Figure I

Limma analysis of the ALL data. The leftmost numbers are row indices, ID is the Affymetrix HGU95av2 accession number, M is the log ratio of expression, A is the log average expression, and B is the log odds of differential expression.

Differential expression (D.E.) = $\log(R) / \log(G)$

Log ratio M : $2^M = \log(R) / \log(G)$; M = 1 \rightarrow zweifach D.E.

Wie signifikant ist dies? \rightarrow bewerte mit statistischem Test.

Vergleiche Gen-Expression in den beiden Gruppen.

Fokussiere auf Gene mit stark unterschiedlicher Expression.

Wähle z.B. alle Gene mit p-Wert < 0.05 aus.

Es bleiben 165 Gene übrig.

Gentleman et al. Genome Biology 5, R80 (2004)

Differentielle Gen-Expression als Heatmap visualisieren

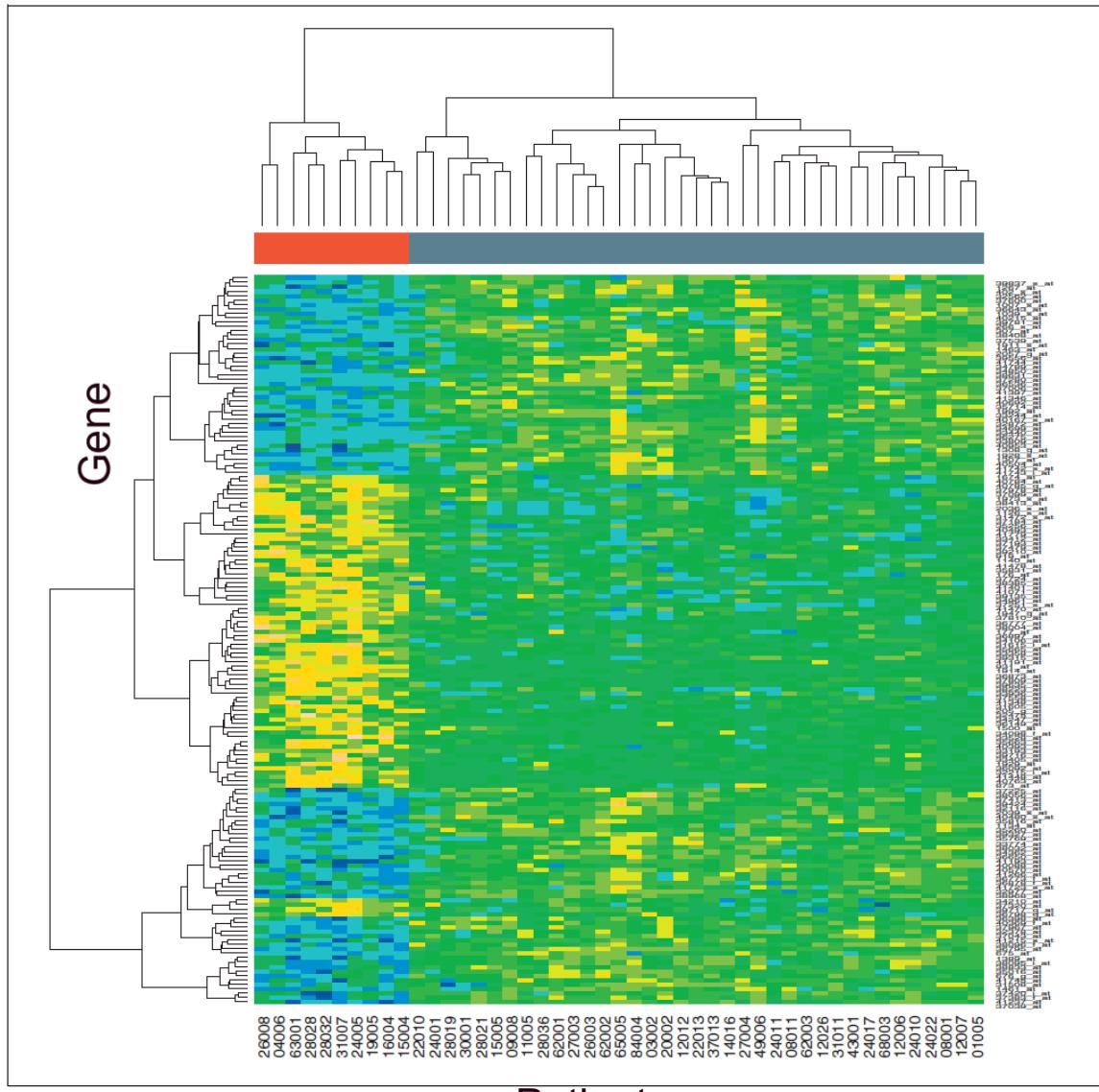


Figure 2
Heat map (produced by the Bioconductor function heatmap()) of the ALL leukemia data.

Mit einem Abstandsmaß und einem Cluster-Algorithmus werden die Ähnlichkeiten zwischen den Patienten (x-Achse) und den einzelnen Gene (y-Achse) erfasst.

Die beiden Patienten-Gruppen haben deutlich unterschiedliche Expressionsprofile (rot/grau).

Gelb: stark hochreguliert
Blau: stark runterreguliert

Gentleman et al. Genome Biology 5, R80 (2004)

Zuordnung von Gen-Funktion

Bioconductor Kommandos						
	ID	M	A	t	p-value	B
1016	1914_at	-3.1	4.6	-27	5.9e-27	56
7884	37809_at	-4.0	4.9	-20	1.3e-20	44
6939	36873_at	-3.4	4.3	-20	1.8e-20	44
10865	40763_at	-3.1	3.5	-17	7.2e-18	39
4250	34210_at	3.6	8.4	15	3.5e-16	35
11556	41448_at	-2.5	3.7	-15	1.8e-15	34
3389	33358_at	-2.3	5.2	-13	3.3e-13	29
8054	37978_at	-1.0	6.9	-10	6.5e-10	22
10579	40480_s_at	1.8	7.8	10	9.1e-10	21
330	1307_at	1.6	4.6	10	1.4e-09	21

Figure 1

Limma analysis of the ALL data. The leftmost numbers are row indices, ID is the Affymetrix HGU95av2 accession number, M is the log ratio of expression, A is the log average expression, and B is the log odds of differential expression.

Links gezeigt ist dieselbe Tabelle wie zwei Folien zuvor.

Nun interessiert uns, welche Funktionen diese Gene in der Zelle ausüben.

Verwende dazu Informationen aus der Gene Ontology über diese Gene.

Gentleman et al. Genome Biology 5, R80 (2004)

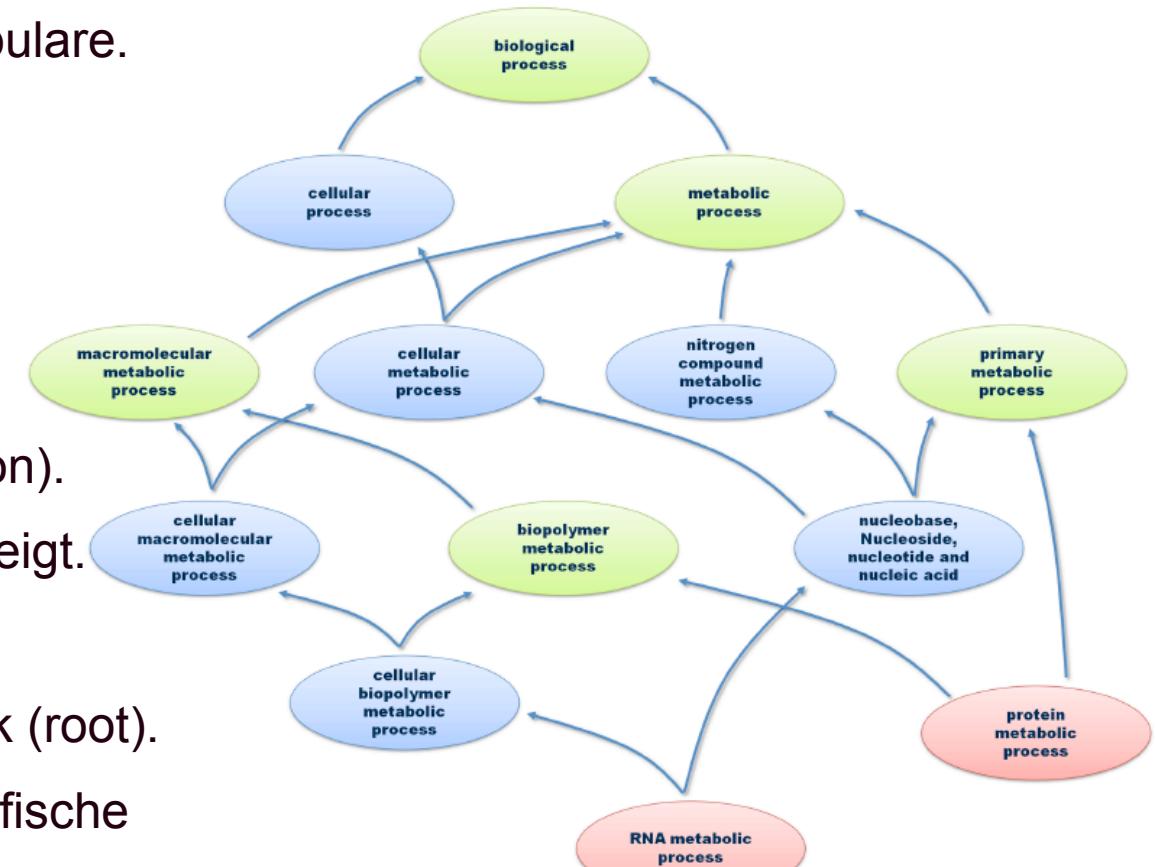
Die Gene Ontology (GO)

Ontologien sind strukturierte Vokabulare.

Die Gene Ontology hat 3 Bereiche:

- biologischer Prozess (BP)
- molekulare Funktion (MF)
- zelluläre Komponente (Lokalisation).

Hier ist ein Teil des BP-Baums gezeigt.



Oben ist der allgemeinste Ausdruck (root).

Rot: Blätter des Baums (sehr spezifische GO-Terme)

Grün: gemeinsame Vorgänger.

Blau: andere Knoten.

Linien: „Y ist in X enthalten“-Beziehungen

Dissertation Andreas Schlicker (UdS, 2010)

Gene Ontology (GO) - Konsortium

Berkeley Bioinformatics Open-source Project (BBOP)

British Heart Foundation - University College London (BHF-UCL)

dictyBase

EcoliWiki

FlyBase

GeneDB

UniProtKB-Gene Ontology Annotation @ EBI (UniProtKB-GOA)

GO Editorial Office at the European Bioinformatics Institute

Gramene

Institute of Genome Sciences, Univ. of Maryland

J Craig Venter Institute

Mouse Genome Informatics (MGI)

Rat Genome Database (RGD)

Reactome

Saccharomyces Genome Database (SGD)

The Arabidopsis Information Resource (TAIR)

WormBase

The Zebrafish Information Network (ZFIN)

Woher stammen die Gene Ontology Annotationen?

Evidence code	Evidence code description	Source of evidence	Manually checked	Current number of annotations*
IDA	Inferred from direct assay	Experimental	Yes	71,050
IEP	Inferred from expression pattern	Experimental	Yes	4,598
IGI	Inferred from genetic interaction	Experimental	Yes	8,311
IMP	Inferred from mutant phenotype	Experimental	Yes	61,549
IPI	Inferred from physical interaction	Experimental	Yes	17,043
ISS	Inferred from sequence or structural similarity	Computational	Yes	196,643
RCA	Inferred from reviewed computational analysis	Computational	Yes	103,792
IGC	Inferred from genomic context	Computational	Yes	4
IEA	Inferred from electronic annotation	Computational	No	15,687,382
IC	Inferred by curator	Indirectly derived from experimental or computational evidence made by a curator	Yes	5,167
TAS	Traceable author statement	Indirectly derived from experimental or computational evidence made by the author of the published article	Yes	44,564
NAS	Non-traceable author statement	No 'source of evidence' statement given	Yes	25,656
ND	No biological data available	No information available	Yes	132,192
NR	Not recorded	Unknown	Yes	1,185

*October 2007 release

Rhee et al. Nature Reviews Genetics 9, 509-515 (2008)

Woher stammen die Gene Ontology Annotationen?

Species (NCBI taxon ID)	Genes* with experimental annotations [‡]	Total annotated genes*	Percentage of genes* with at least one experimental annotation	Total genes*	Percentage annotated [§]	Percentage known in genome
<i>Schizosaccharomyces pombe</i> (4896)	4,482	4,930	90.9%	4,930	100%	90.9%
<i>Saccharomyces cerevisiae</i> (4932)	4,947	5,794	85.4%	5,794	100%	85.4%
Mouse (10090)	10,621	18,386	57.8%	27,289	67.4%	38.9%
<i>Caenorhabditis elegans</i> (6239)	4,614	14,154	32.6%	20,163	70.2%	22.9%
Human [¶] (9606)	4,780	17,021	28.1%	20,887	81.5%	22.9%
<i>Arabidopsis thaliana</i> [#] (3702)	5,530	26,637	20.8%	27,029	98.5%	20.5%
Rat (10116)	3,566	17,243	20.7%	17,993	95.8%	19.8%
Fruitfly (7227)**	2,790	9,563	29.2%	14,141	67.6%	19.7%
<i>Candida albicans</i> (5476)	806	3,756	21.4%	6,166	60.9%	13.0%
<i>Pseudomonas aeruginosa</i> PAO1 (208964)	491	2,506	19.6%	5,568	45.0%	8.82%
Slime mold (44689)	797	6,892	11.6%	13,625	50.6%	5.9%
<i>Trypanosoma brucei</i> (5691)	449	3,914	11.5%	9,154	42.8%	4.92%
Zebrafish (7955)	1,235	13,574	5.8%	21,322	63.7%	3.7%
<i>Plasmodium falciparum</i> (5833)	188	3,243	5.8%	5,420	59.8%	3.47%
Rice (39947)	654	29,877	2.2%	41,908	71.3%	1.57%
Chicken [¶] (9031)	75	6,063	1.2%	16,737	36.2%	0.4%
Cow [¶] (9913)	96	8,536	1.1%	21,756	39.2%	0.4%

*Total genes in genomes include only those that encode proteins. These numbers were obtained from the databases that contribute annotations to GO and are listed on the GO annotations download page (<http://www.geneontology.org/GO.current.annotations.shtml>). [‡]Experimental annotations include those only with the following evidence codes: IDA (inferred from direct assay), IEP (inferred from expression pattern), IGI (inferred from genetic interaction), IMP (inferred from mutant phenotype) and IPI (inferred from physical interaction). [§]Percentage annotated is determined by dividing the number of genes annotated by total genes. ^{||}Percentage known in genome is determined by multiplying the percentage of experimentally derived annotations by the percentage of the genome annotated. This is an approximation of the extent of knowledge about the portion of the genome that encodes proteins in an organism with a complete genome sequence that is captured by annotation. [#]Numbers are from the GO annotation project at the European Bioinformatics Institute, human data last updated 14 September 2007, cow data last updated 17 January 2007, chicken data last updated 10 July 2007. [¶]Numbers are from The Arabidopsis Information Resource (TAIR), last updated 14 December 2007. ^{**}Numbers are based on release 5.4 of the *Drosophila melanogaster* genome and GO annotations from FlyBase release FB2007_03 (dated 11 January 2007). NCBI, National Center for Biotechnology Information.

Rhee et al. Nature Reviews Genetics 9, 509-515 (2008)

Format des GO flat files

Column	Content	Required?	Cardinality	Example
1	DB	required	1	UniProtKB
2	DB Object ID	required	1	P12345
3	DB Object Symbol	required	1	PHO3
4	Qualifier	optional	0 or greater	NOT
5	GO ID	required	1	GO:0003993
6	DB:Reference (DB:Reference)	required	1 or greater	PMID:2676709
7	Evidence Code	required	1	IMP
8	With (or) From	optional	0 or greater	GO:0000346
9	Aspect	required	1	F
10	DB Object Name	optional	0 or 1	Toll-like receptor 4
11	DB Object Synonym (Synonym)	optional	0 or greater	hToll Tollbooth
12	DB Object Type	required	1	protein
13	Taxon(taxon)	required	1 or 2	taxon:9606
14	Date	required	1	20090118
15	Assigned By	required	1	SGD
16	Annotation Extension	optional	0 or greater	part_of(CL:0000576)
17	Gene Product Form ID	optional	0 or 1	UniProtKB:P12345-2

Beispiel: GO-Annotation für humanes BRCA1-Gen

BRCA1

Breast cancer type 1 susceptibility protein

protein from *Homo sapiens* (human)

Term associations → Gene product information → Peptide Sequence → Sequence information →

Term Associations

Download all association information in: [gene association format](#) [RDF-XML](#)

▼ Filter associations displayed [?](#)

Filter Associations

Ontology	Evidence Code
All biological process cellular component molecular function	All IC IDA IEA

[Set filters](#) [Remove all filters](#)

1 [2](#) [View all results](#)

Select all Clear all [Perform an action with this page's selected terms...](#) [Go!](#)

Accession, Term	Ontology	Qualifier	Evidence	Reference	Assigned by
162 gene products view in tree biological process GO:0030521 : androgen receptor signaling pathway		NAS		PMID:15572661	UniProtKB
6411 gene products view in tree biological process GO:0006915 : apoptosis		TAS		PMID:10918303	UniProtKB
1997 gene products view in tree biological process GO:0007420 : brain development		IEA With Ensembl:ENSRNOP00000028109		GO REF:0000019	Ensembl (via UniProtKB)
10144 gene products view in tree biological process GO:0007049 : cell cycle		IEA With SP KW:KW-0131		GO REF:0000004	UniProtKB
26 gene products view in tree biological process		IDA		PMID:10868478	UniProtKB

Einzelne
GO-Terme, mit
denen das
Brustkrebs
-Gen
BRCA1
annotiert
ist.

Signifikanz von GO-Annotationen

Sehr **allgemeine GO-Terme** wie z.B. “cellular metabolic process“ werden vielen Genen im Genom zugeordnet.

Sehr **spezielle Terme** gehören jeweils nur zu wenigen Genen.

Man muss also vergleichen, wie **signifikant** das Auftreten jedes GO-Terms in einer Testmenge an Genen im Vergleich zu einer zufällig ausgewählten Menge an Genen derselben Größe ist.

Dazu verwendet man meist den **hypergeometrischen Test**.

Vorbemerkung

Zieht man aus einer Urne mit n Kugeln insgesamt k Kugeln **ohne Beachtung der Reihenfolge**, so gibt es hierfür genau

$$\frac{n \cdot (n-1) \cdot (n-2) \cdot \dots \cdot (n-k+1)}{k!} = \frac{n!}{k! \cdot (n-k)!} = \binom{n}{k} \text{ Möglichkeiten}$$

Hypergeometrischer Test

$$\text{p-Wert} = \sum_{i=k_\pi}^{\min(n, K_\pi)} \frac{\binom{K_\pi}{i} \binom{N-K_\pi}{n-i}}{\binom{N}{n}}$$

Der hypergeometrische Test ist ein statistischer Test, der z.B. überprüft, ob in einer vorgegebenen Testmenge an Genen eine biologische Annotation π gegenüber dem gesamten Genom statistisch signifikant angereichert ist.

- Sei N die Anzahl an Genen im Genom.
- Sei n die Anzahl an Genen in der Testmenge.
- Sei K_π die Anzahl an Genen im Genom mit der Annotation π .
- Sei k_π die Anzahl an Genen in der Testmenge mit der Annotation π .

Der hypergeometrische p-Wert drückt die Wahrscheinlichkeit aus, dass k_π oder mehr **zufällig** aus dem Genom ausgewählte Gene auch die Annotation π haben.

Hypergeometrischer Test

Wähle $i = k_{\pi}$ Gene mit
Annotation π aus dem Genom.
Davon gibt es genau K_{π} .

$$p\text{-Wert} = \sum_{i=k_{\pi}}^{\min(n, K_{\pi})} \frac{\binom{K_{\pi}}{i} \binom{N-K_{\pi}}{n-i}}{\binom{N}{n}}$$

Die anderen $n - i$ Gene in der Testmenge haben dann nicht die Annotation π . Davon gibt es im Genom genau $N - K_{\pi}$.

Die Summe läuft von mindestens k_{π} Elementen bis zur maximal möglichen Anzahl an Elementen.

Eine Obergrenze ist durch die Anzahl an Genen mit Annotation π im Genom gegeben (K_{π}).

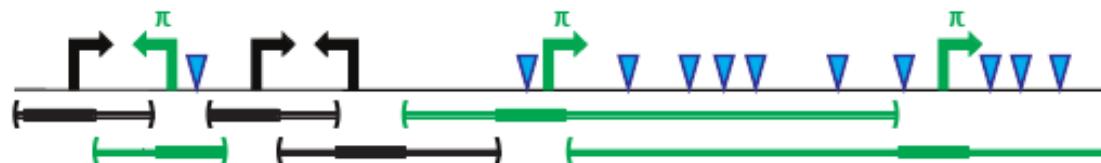
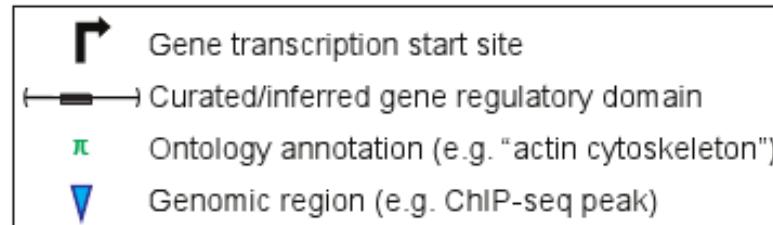
Die andere Obergrenze ist die Zahl der Gene in der Testmenge (n).

Korrigiert für die kombinatorische Vielfalt an Möglichkeiten um n Elemente aus einer Menge mit N Elementen auszuwählen.

N.B. dies gilt für den Fall, dass die Reihenfolge der Elemente egal ist.

Beispiel

$$p\text{-Wert} = \sum_{i=k_\pi}^{\min(n, K_\pi)} \frac{\binom{K_\pi}{i} \binom{N-K_\pi}{n-i}}{\binom{N}{n}}$$



Frage: ist die Annotation π in der Testmenge von 3 Genen signifikant angereichert?

Ja! $p = 0.05$ ist (knapp) signifikant.

<http://great.stanford.edu/>

Anwendung auf ALL-Beispiel

```
> ll <- mget(geneNames(esetSel),  
             hgu95av2LOCUSID)  
> ll <- unique(unlist(ll))  
> mf <- as.data.frame(GOHyperG(ll))[, 1:3]  
> mf <- mf[mf$pvalue < 0.01, ]  
> mf
```

Bioconductor
Kommandos

	p-values	goCounts	intCounts
GO:0045012	1.1e-08	12	6
GO:0030106	6.4e-03	8	2
GO:0004888	7.0e-03	523	16
GO:0004872	9.7e-03	789	21

```
> as.character(unlist(mget(row.names(mf),  
                      GOTERM)))
```

- [1] "MHC class II receptor activity"
- [2] "MHC class I receptor activity"
- [3] "transmembrane receptor activity"
- [4] "receptor activity"

Figure 3

Hypergeometric analysis of molecular function enrichment of genes selected in the analysis described in Figure 1.

Die signifikanteste Anreicherung ergibt sich für MHC Klasse 2 Rezeptoraktivität.
6 von 12 Genen im Genom mit dieser Annotation sind in den 2 ALL-Klassen differentiell exprimiert.

Gentleman et al. Genome Biology 5, R80 (2004)

NIH Tool David: Tool für Annotation der Genfunktion

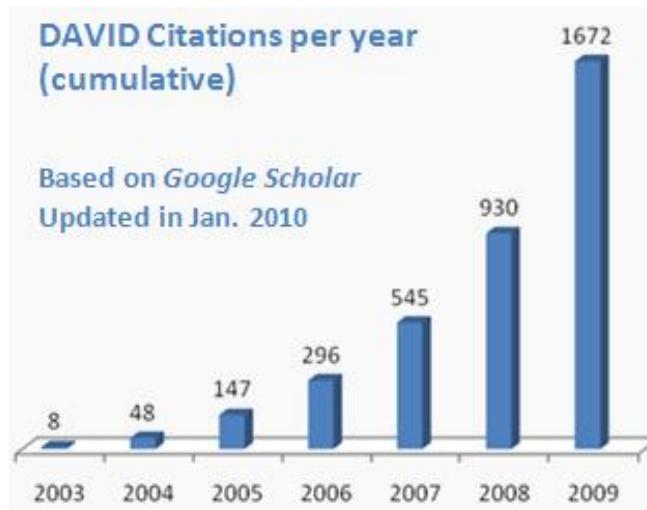
PROTOCOL

Systematic and integrative analysis of large gene lists using DAVID bioinformatics resources

Da Wei Huang^{1,2}, Brad T Sherman^{1,2} & Richard A Lempicki¹

¹Laboratory of Immunopathogenesis and Bioinformatics, Clinical Services Program, SAIC-Frederick Inc., National Cancer Institute at Frederick, Frederick, Maryland 21702, USA. ²These authors contributed equally to this work. Correspondence should be addressed to R.A.L. (rlempicki@mail.nih.gov) or D.W.H. (huangdawei@mail.nih.gov)

Published online 18 December 2008; doi:10.1038/nprot.2008.211



NIH Tool David

The screenshot shows the DAVID 2006 Functional Annotation Bioinformatics homepage. At the top, there is a navigation bar with links to Home, Start Analysis, Shortcut to DAVID Tools, Technical Center, Archives, Term of Service, DAVID Forum, Credits, and About Us. A red arrow points to the "Shortcut to DAVID Tools" link. Below the navigation bar, there is a banner for "DAVID Bioinformatic Resources 2006" from the National Institute of Allergy and Infectious Diseases (NIAID), NIH. To the left, there is a sidebar with sections for Functional Annotation, Gene Functional Classification, Gene ID Conversion, and a link to List Gene Names in Batch. The main content area features a "Welcome to DAVID Bioinformatic Resources" section with a brief description of the tool's purpose and a list of features. To the right, there are sections for "What's New in DAVID 2006?", "DAVID Bioinformatic Forum", and "Statistics About DAVID". The bottom of the page includes standard browser controls like Back, Forward, Stop, Refresh, and Home, along with a search bar and a toolbar.

DAVID Bioinformatic Resources 2006
National Institute of Allergy and Infectious Diseases (NIAID), NIH

Home Start Analysis **Shortcut to DAVID Tools** Technical Center Archives Term of Service DAVID Forum Credits About Us

Welcome to DAVID Bioinformatic Resources

The Database for Annotation, Visualization and Integrated Discovery (**DAVID**) 2006 is an expanded version of our original web-accessible programs of DAVID 2.1, 2.0 & 1.0. DAVID provides a comprehensive set of functional annotation tools for investigators to understand biological meaning behind large list of genes. For any given gene list, DAVID tools are able to:

- Identify enriched biological themes, particularly GO terms
- Discover enriched functional-related gene groups
- Visualize genes on BioCarta & KEGG pathway maps
- Search for other functionally related genes not in the list
- List interacting proteins
- Explore gene names in batch
- Link gene-disease associations
- Highlight protein functional domains and motifs
- Redirect to related literatures

What's New in DAVID 2006?

- [Functional Annotation Clustering](#)
- [Pre-built Affy gene backgrounds](#)
- [User's customized gene background](#)
- [Updated annotation databases](#)
- [Enhanced calculating speed](#)

DAVID Bioinformatic Forum

- [Technical notes & help](#)
- [Ask questions & get answers](#)
- [Share experiences](#)
- [Comments and feedback](#)
- [Bug report](#)

Statistics About DAVID

Submit gene list or use built-in demo_lists

The screenshot shows a Microsoft Internet Explorer window displaying the DAVID 2006 functional annotation result summary. The title bar reads "DAVID 2006: functional annotation result summary - Microsoft Internet Explorer". The address bar shows the URL "http://david.abcc.ncifcrf.gov/tools.jsp". The main content area features the DAVID Bioinformatic Resources logo and the "Analysis Wizard" title. A blue arrow points from the text "Step 1. Submit your gene list through left panel." to the "Upload Gene List" section on the left. This section includes links for "Demolist 1" and "Demolist 2", a "Upload Help" link (which has a red arrow pointing to it), and a "Step 1: Enter Gene List" area with options to "Paste a list" (with a text input field and "Clear" button) or "Choose From a File" (with a "Browse..." button). Below this is "Step 2: Select Identifier" (set to "AFFY_ID"), "Step 3: List Type" (radio buttons for "Gene List" and "Background" with "Gene List" selected), and "Step 4: Submit List" (a "Submit List" button). To the right of the left panel, there is an "Analysis Wizard" section with a "Tell us how you like the tool" link and a "Contact us for questions" link. Below these are some sample gene IDs: 1007_s_at, 1053_at, 117_at, 121_at, 1255_g_at, 1294_at, 1316_at, 1320_at, 1405_i_at, 1431_at, 1438_at, 1487_at, 1494_f_at, and 1598_g_at.

Select the DAVID Gene Functional Classification Tool

The screenshot shows the DAVID 2006 functional annotation result summary page in Microsoft Internet Explorer. The title bar reads "DAVID 2006: functional annotation result summary - Microsoft Internet Explorer". The main content area is titled "Analysis Wizard" and displays the DAVID Bioinformatic Resources 2006, NIAID/NIH logo. On the left, there is a "Gene List Manager" sidebar with tabs for "Upload", "List", and "Background". Under "List", a dropdown menu shows "HOMO SAPIENS(403)" and "SYNTHETIC CONSTRUCT(0)". A "Select" button is below the dropdown. Below the dropdown is a "List Manager" section with a "Demo_List_2" entry. Under "Select List to:", there are buttons for "Use", "Rename", "Remove", and "Combine". A "Show Gene List" button is also present. The main right-hand panel is titled "Analysis Wizard" and contains the following steps:

- Step 1. Successfully submitted gene list**
 - Current Gene List: Demo_List_2
 - Current Background: HOMO SAPIENS
- Step 2. Analyze above gene list with one of DAVID tools**
 - A blue downward arrow points to the following list:
 - [Functional Annotation Tool](#)
 - [Functional Annotation Clustering](#)
 - [Functional Annotation Chart](#)
 - [Functional Annotation Table](#)
 - [Gene Functional Classification Tool](#) (highlighted with a red arrow)
 - [Gene ID Conversion Tool](#)
 - [Show Gene List Tool](#)

On the right side of the main panel, there are links: "Tell us how you like the tool" and "Contact us for questions". At the bottom right, there is a link "Which DAVID tools to use?".

Select the DAVID Gene Functional Classification Tool

DAVID 2006: Gene Functional Classification - Microsoft Internet Explorer

File Edit View Favorites Tools Help

Address http://david.abcc.ncifcrf.gov/gene2gene.jsp

Home Start Analysis Shortcut to DAVID Tools Technical Center Archives Term of Service DAVID Forum Credits About Us

Upload List Background

Gene List Manager

Select to limit annotations by one or more species

Help

- Use All Species -
HOMO SAPIENS(403)
SYNTHETIC CONSTRUCTS

Select

List Manager Help

Demo_List_2

Select List to:

Use Rename
Remove Combine
Show Gene List new!

Gene Functional Classification

Current Gene List: Demo_List_2

Current Background: HOMO SAPIENS

394 DAVID IDs

Options Classification Stringency Medium

Rerun using options Create Sublist Heatmap

16 Clusters

Download File

	Gene Group 1	Enrichment Score: 3.37	RG	T	
1	34375_at, 875_g_at	chemokine (c-c motif) ligand 2			
2	40385_at	chemokine (c-c motif) ligand 20			
3	36103_at	chemokine (c-c motif) ligand 3			
4	36674_at	chemokine (c-c motif) ligand 4			
5	408_at	chemokine (c-x-c motif) ligand 1 (melanoma growth stimulating activity, alpha)			
6	1369_s_at, 35372_r_at	interleukin 8			

	Gene Group 2	Enrichment Score: 2.89	RG	T	
1	1857_at	smad, mothers against dpp homolog 7 (drosophila)			
2	39421_at	runt-related transcription factor 1 (acute myeloid leukemia 1; aml1 oncogene)			
3	36999_at	jumonji, at rich interactive domain 1a (rbbp2-like)			
4	1994_at	activating transcription factor 2			
5	1895_at, 32583_at	v-jun sarcoma virus 17 oncogene homolog (avian)			
6	35768_at	ring finger protein 40			
7	36226_r_at	splicing factor proline/glutamine-rich (polypyrimidine tract binding protein associated)			
8	789_at	early growth response 1			

Select the DAVID Gene Functional Annotation Tool

The screenshot shows the DAVID 2006 functional annotation result summary page. At the top, the title "DAVID 2006: functional annotation result summary - Microsoft Internet Explorer" is displayed. Below the title, the DAVID logo and the text "Functional Annotation Tool" and "DAVID Bioinformatic Resources 2006, NIAID/NIH" are visible. A navigation menu at the top includes links for Home, Start Analysis, Shortcut to DAVID Tools, Technical Center, Archives, Term of Service, DAVID Forum, Credits, and About Us.

The main content area is titled "Annotation Summary Results". It displays the current gene list ("Demo_List_2") and background ("HOMO SAPIENS"). There are 394 DAVID IDs. A "Check Defaults" checkbox is checked, and a "Clear All" button is available. On the left, there is a "Gene List Manager" section where users can select species (e.g., HOMO SAPIENS, SYNTHETIC CONSTRUCT), manage lists (e.g., Demo_List_2), and perform actions like Use, Rename, Remove, Combine, or Show Gene List.

Below the manager, a "Combined View for Selected Annotation" section offers three options: "Functional Annotation Clustering" (highlighted with a red arrow), "Functional Annotation Chart", and "Functional Annotation Table".

Funktionelles Clustering von angereicherten GO-Termen

Options		Classification Stringency	Custom		
		Rerun using options	Create Sublist	Heatmap	Cluster Comparison
		Functional Group 1	2.9E-4	RG	T
1	<input type="checkbox"/> 31506_s_at, 31793_at	defensin, alpha 1			
2	<input type="checkbox"/> 34546_at	defensin, alpha 4, corticostatin			
3	<input type="checkbox"/> 34623_at	defensin, alpha 5, paneth cell-specific			
		Functional Group 2	7.0E-4	RG	T
1	<input type="checkbox"/> 35566_f_at	immunoglobulin heavy constant gamma 1 (g1m marker)			
2	<input type="checkbox"/> 35566_f_at	immunoglobulin heavy locus			
3	<input type="checkbox"/> 1355_g_at	neurotrophic tyrosine kinase, receptor, type 2			
4	<input type="checkbox"/> 1786_at	c-mer proto-oncogene tyrosine kinase			
5	<input type="checkbox"/> 1901_s_at	v-erb-b2 erythroblastic leukemia viral oncogene homolog 2, neuro/glioblastoma derived oncogene homolog (avian)			
6	<input type="checkbox"/> 1112_g_at	neural cell adhesion molecule 1			
7	<input type="checkbox"/> 32469_at	carcinoembryonic antigen-related cell adhesion molecule 3			
8	<input type="checkbox"/> 35038_at	myosin binding protein c, cardiac			
9	<input type="checkbox"/> 35090_g_at, 35091_at	neuregulin 2			
10	<input type="checkbox"/> 37968_at	natural cytotoxicity triggering receptor 3			
11	<input type="checkbox"/> 33530_at	carcinoembryonic antigen-related cell adhesion molecule 8			
12	<input type="checkbox"/> 35956_s_at	pregnancy specific beta-1-glycoprotein 4			
13	<input type="checkbox"/> 31987_at	kin of irre like (drosophila)			
14	<input type="checkbox"/> 35956_s_at	pregnancy specific beta-1-glycoprotein 2			
		Functional Group 3	2.7E-3	RG	T
1	<input type="checkbox"/> 37454_at	chemokine (c-c motif) ligand 13			
2	<input type="checkbox"/> 36703_at	chemokine (c-c motif) ligand 25			
3	<input type="checkbox"/> 1403_s_at	chemokine (c-c motif) ligand 5			
		Functional Group 4	3.5E-3	RG	T
1	<input type="checkbox"/> 31687_f_at	hemoglobin, beta			
2	<input type="checkbox"/> 33516_at	hemoglobin, delta			
3	<input type="checkbox"/> 31525_s_at	hemoglobin, alpha 1			
		Functional Group 5	4.1E-3	RG	T
1	<input type="checkbox"/> 40317_at	amiloride-sensitive cation channel 1, neuronal (neenerin)			

XXXX_at sind die Kürzel für einzelne Proben auf Affymetrix-Microarray-Chip

Huang et al. Genome Biology 2007 8:R183

Messe funktionelle Ähnlichkeit von GO-Termen

Die **Wahrscheinlichkeit eines Knoten** t kann man auf 2 Arten ausdrücken:

Wieviele Gene besitzen die

Anzahl an GO-Termen im bei t

Annotation t relativ zur Häufigkeit

startenden Unterbaum relativ zu der

der Wurzel?

Anzahl an GO-Termen im Gesamtbaum.

$$p_{anno}(t) = \frac{occur(t)}{occur(root)}$$

$$p_{graph}(t) = \frac{D(t)}{D(root)}$$

Die Wahrscheinlichkeit hat Werte zwischen 0 und 1 und nimmt zwischen den Blättern bis zur Wurzel monoton zu.

Aus der Wahrscheinlichkeit p berechnet man den **Informationsgehalt** jedes Knotens:

$$IC(t) = -\log p(t)$$

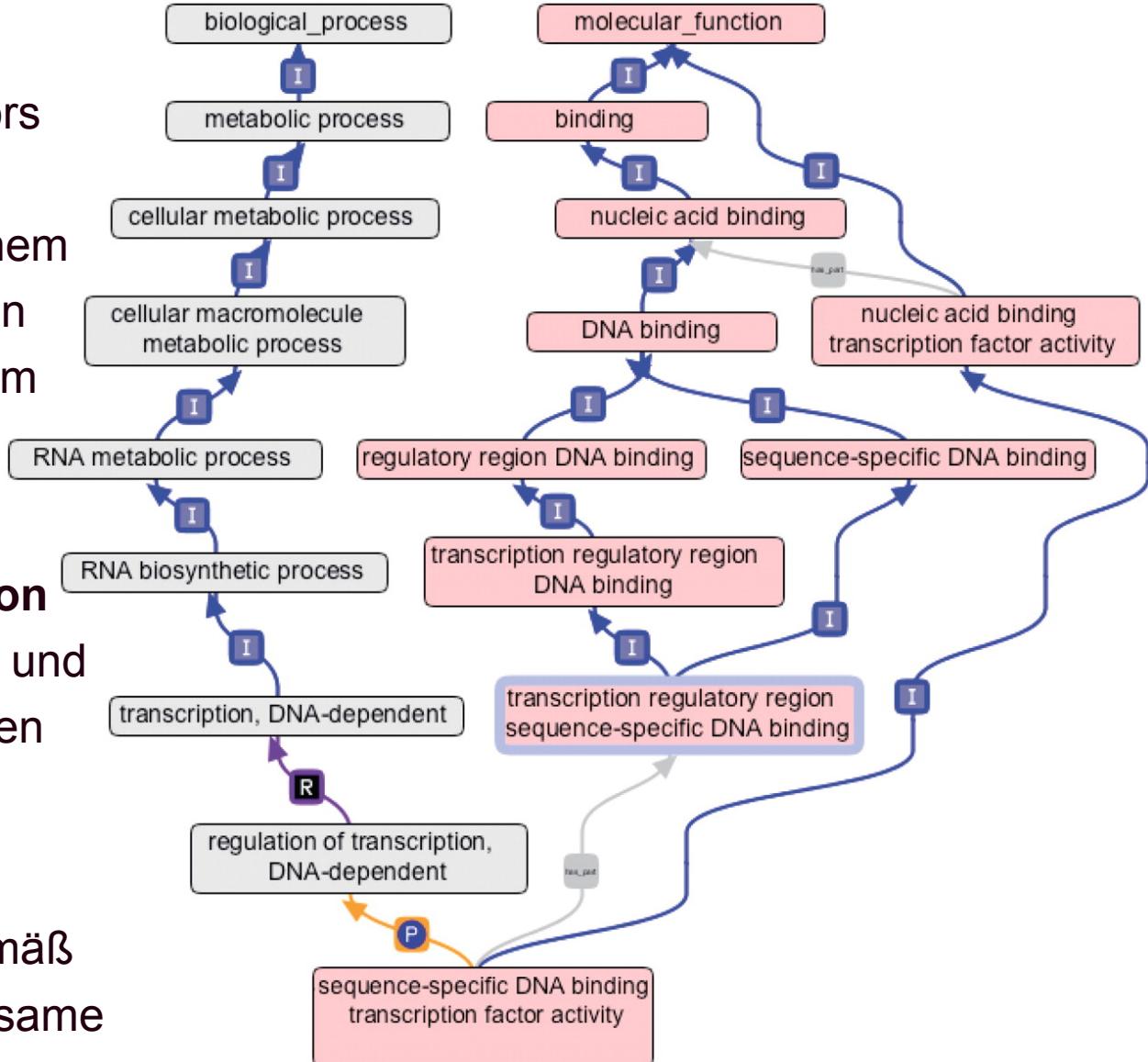
Je seltener ein Knoten ist, desto höher sein Informationsgehalt.

Messe funktionelle Ähnlichkeit von GO-Termen

Die Menge an gemeinsamen Vorgängern (common ancestors (CA)) zweier Knoten t_1 und t_2 enthält alle Knoten, die auf einem Pfad von t_1 zum Wurzel-Knoten **UND** auf einem Pfad von t_2 zum Wurzelknoten liegen.

Der **most informative common ancestor** (MICA) der Terme t_1 und t_2 ist der Term mit dem höchsten Informationsgehalt in CA.

Normalerweise ist das der gemäß dem Abstand nächste gemeinsame Vorgänger.



Nucl. Acids Res. (2012) 40 (D1):
D559-D564

Messe funktionelle Ähnlichkeit von GO-Termen

Schlicker et al. definierten aus dem Abstand zum **most informative common ancestor** (MICA) die Ähnlichkeit der Terme t_1 und t_2

$$sim_{Rel}(t_1, t_2) = \frac{2 \cdot IC(MICA)}{IC(t_1) + IC(t_2)} \cdot (1 - p(MICA))$$

Der hintere Faktor gewichtet
die Ähnlichkeit mit der
Häufigkeit $p(MICA)$. Dies ergab
Vorteile in der Praxis.

Messe funktionelle Ähnlichkeit von GO-Termen

Zwei Gene oder zwei Mengen an Genen A und B haben jedoch meist jeweils mehr als eine GO-Annotation. Betrachte daher die Ähnlichkeit aller Terme i und j :

$$s_{ij} = \text{sim}(GO_i^A, GO_j^B), \forall i \in 1, \dots, N, \forall j \in 1, \dots, M.$$

und wähle daraus in den Reihen und Spalten jeweils die Maxima

$$\text{rowScore}(A, B) = \frac{1}{N} \sum_{i=1}^N \max_{1 \leq j \leq M} s_{ij}, \quad \text{GOscore}_{\text{avg}}^{\text{BMA}}(A, B) = \frac{1}{2} \cdot (\text{rowScore}(A, B) + \text{columnScore}(A, B))$$

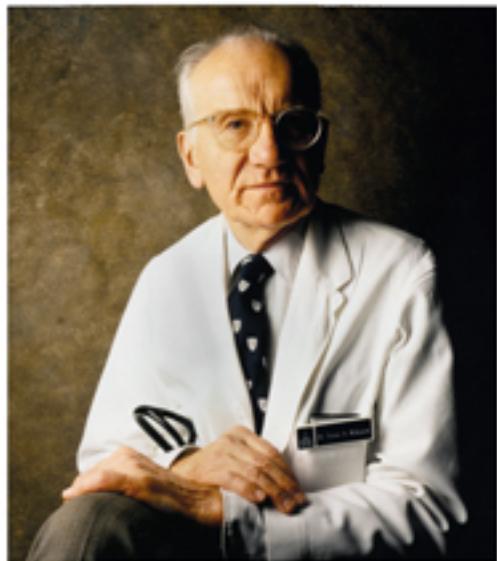
$$\text{columnScore}(A, B) = \frac{1}{M} \sum_{j=1}^M \max_{1 \leq i \leq N} s_{ij}. \quad \text{GOscore}_{\text{max}}^{\text{BMA}}(A, B) = \max(\text{rowScore}(A, B), \text{columnScore}(A, B))$$

Aus den Scores für den BP-Baum und den MF-Baum wird der *funsim*-Score berechnet.

$$\text{funsim}(A, B) = \frac{1}{2} \cdot \left[\left(\frac{\text{BPscore}}{\max(\text{BPscore})} \right)^2 + \left(\frac{\text{MFscore}}{\max(\text{MFscore})} \right)^2 \right]$$

Schlicker PhD dissertation (2010)

OMIM-Datenbank



Victor McKusick (1921-2008),
Johns Hopkins Universität,
- begründete das Gebiet
Medical genetics
- gründete die Datenbank
*Mendelian Inheritance in
Man*

OMIM®, Online Mendelian Inheritance in Man®.
OMIM is a comprehensive, authoritative, and
timely compendium of human genes and genetic
phenotypes.

NCBI All Databases PubMed Nucleotide Protein Genome Structure PMC OMIM

Join My NCBI [Sign In] [Register]

Search OMIM for Go Clear

Limits Preview/Index History Clipboard Details

Display Detailed Show 20 Send to

All: 1 OMIM UniSTS: 0 OMIM dbSNP: 0

MIM ID #211980 GeneTests, Links

LUNG CANCER

Other entities represented by this entry

ALVEOLAR CELL CARCINOMA, INCLUDED
ADENOCARCINOMA OF LUNG, INCLUDED
NONSMALL CELL LUNG CANCER, INCLUDED
LUNG CANCER, PROTECTION AGAINST, INCLUDED

Gene map locus: 17q21.1, 12p12.1, etc.

Clinical Synopsis

Text [Back to Top](#)

A number sign (#) is used with this entry because mutations in several different genes are associated with lung cancer. Both germline and somatic mutations have been identified in the EGFR ([131550](#)) and p53 ([TP53; 191170](#)) genes, and somatic mutations have been identified in the KRAS ([190070](#)), BRAF ([164757](#)), ERBB2 ([164870](#)), MET ([164860](#)), STK11 ([602216](#)), PIK3CA ([171834](#)), and PARK2 ([602544](#)) genes. Amplification of several genes,

Table of Contents

MIM #211980
Text
Description
Clinical Features
Inheritance
Population Genetics
Pathogenesis
Clinical Management
Mapping
Molecular Genetics
Cytogenetics
Clinical Synopsis
See Also
References
Contributors
Creation Date
Edit History

Links

Improving disease gene prioritization using the semantic similarity of Gene Ontology terms

Andreas Schlicker[†], Thomas Lengauer and Mario Albrecht*

Max Planck Institute for Informatics, Department of Computational Biology and Applied Algorithmics, Campus E1.4,
66123 Saarbrücken, Germany

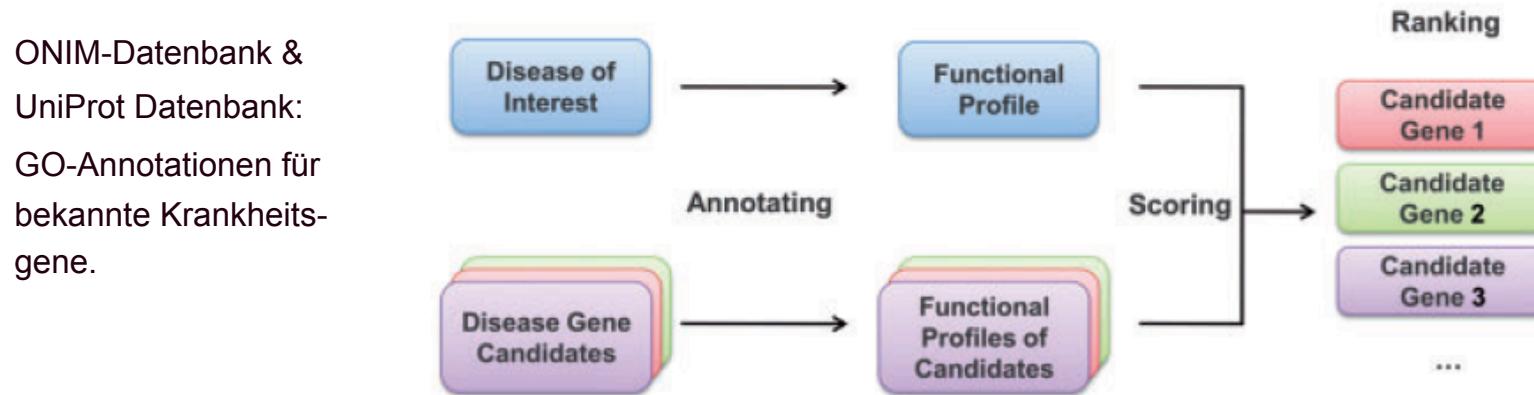
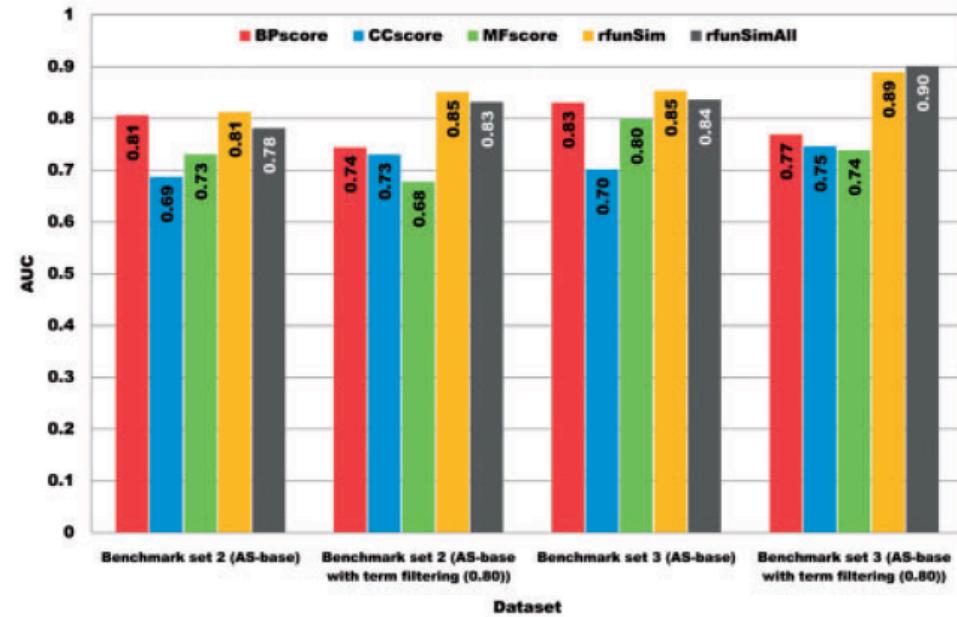


Fig. 1. Flow chart of the MedSim approach. First, the functional profiles of the disease of interest and the disease gene candidates are created using one of the annotation strategies. Afterwards, the functional profile of the disease is scored against each functional profile of a candidate, and the candidates are ranked according to this functional similarity score.

Schlicker et al. Bioinformatics 26, i561 (2010)

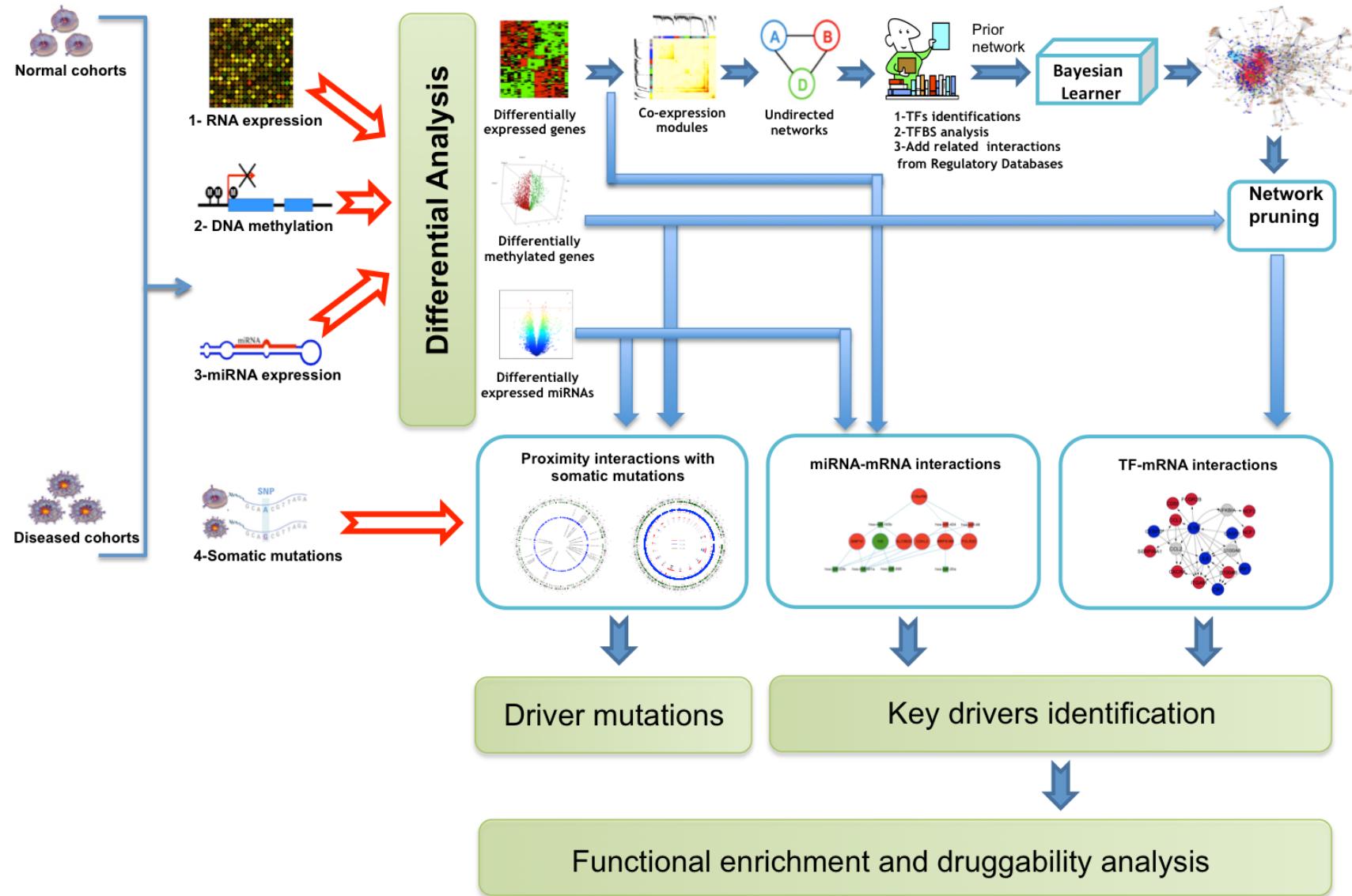
Die Methode liefert recht genaue Vorhersagen, mit welchen Krankheiten Gene in Verbindung stehen könnten.

Die Sensitivität, d.h. die Anzahl der korrekten Vorhersagen relativ zur Anzahl aller Vorhersagen, beträgt 73%.



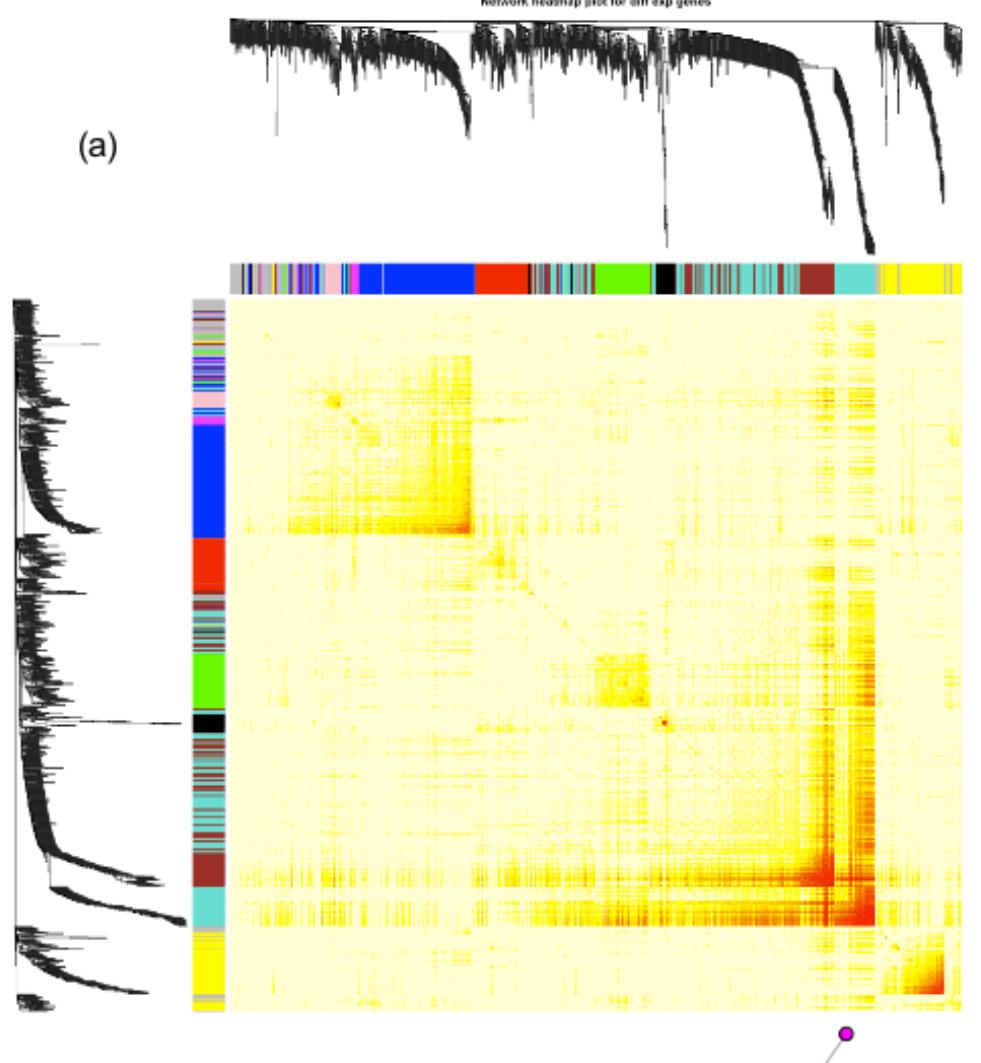
Schlicker et al. Bioinformatics 26, i561 (2010)

funktionelle Annotation von OMICS-Daten für Brustkrebs



Hamed et al. BMC Genomics (2015)

Analyse von Ko-Expression



Ko-Expression der 1317 differenziell exprimierten Gene (Krebs vs. Normal)

Hierarchisches Clustern

-> 10 Module mit 26 – 295 Genen

Hamed et al. BMC Genomics (2015)

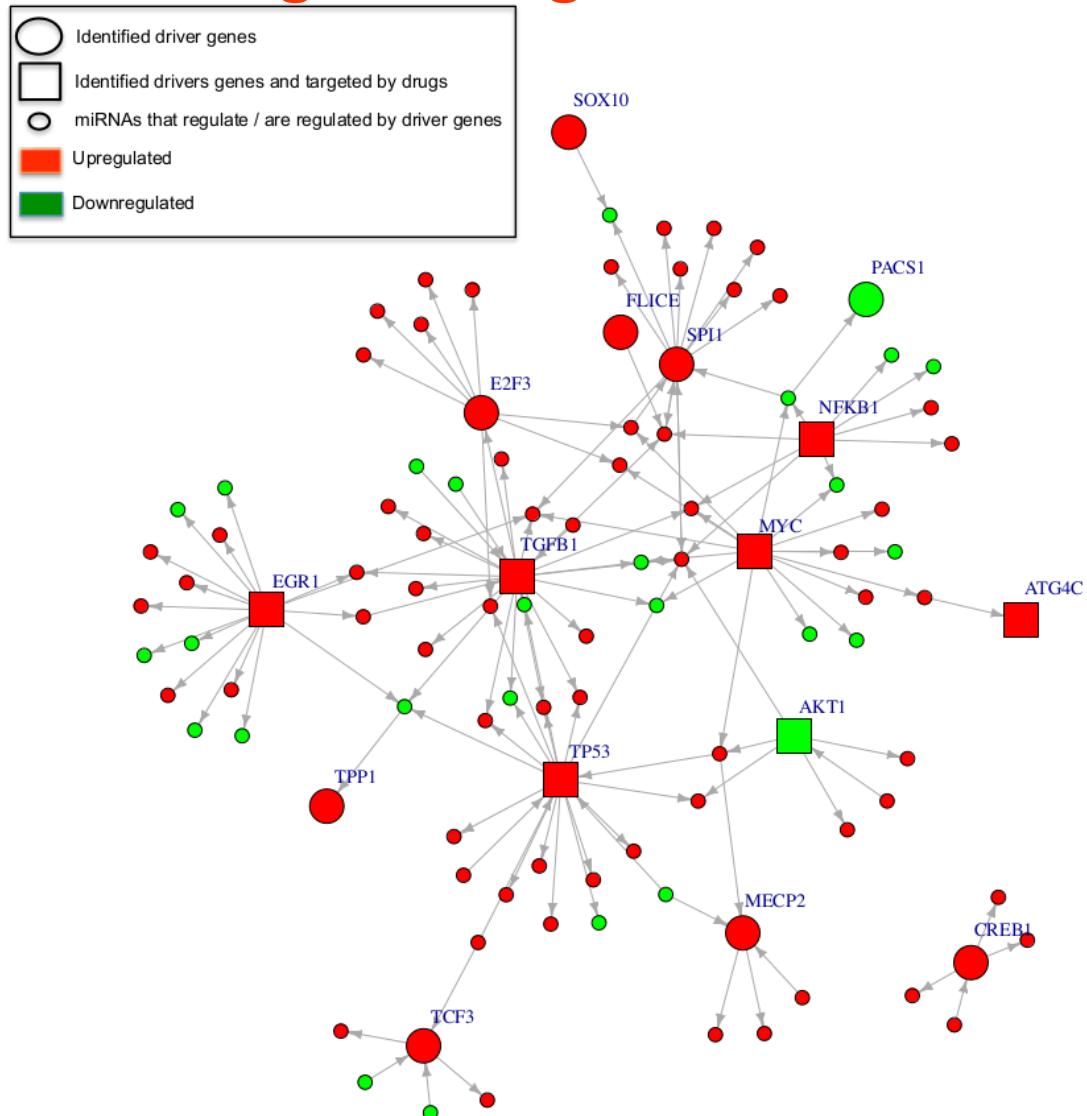
Gibt es angereicherte Genfunktionen in diesen Modulen?

	Module	Gene count	Top GO category	Top KEGG categories	Key driver count	Key drivers
TF- mRNA interactions	black	41	Regulation of transcription	Pathways in cancer, Renal cell carcinoma	5	SORBS3, ZNF43, ZNF681, RBMX, POU2F1
	blue	247	Nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	Cell cycle, Prostate cancer, Melanoma	9	AR , BRCA1 , ESR1 , JUN , MYB , RPN1, E2F1, E2F2, PPARD
	brown	195	Anatomical structure morphogenesis	Leukocyte transendothelial migration	5	TMOD3, CREB1, POU5F1, SP3, TERT
	green	110	Cellular macromolecule metabolic process	Endometrial cancer, Insulin signaling pathway	15	B4GALT7 , OS9 , CDC34 , MAN2C1, MYO1C, SH3GLB2, INPP5E, PLXNB1, USF2, PPP1R12C, CDK9, DAP, E4F1, E2F4, USF1
	grey	148	Anatomical structure development	Sulfur metabolism	18	AHCTF1 , NQO2 , FGFR2 , CCDC130 , ABCG4 , BIRC6 , CA6 , SP4, RNF2, SPRR1B, C16orf65, DNAJC5G, SNCAIP, GRIK5, SLC6A4, SMAD1, DAD1, POU4F2
	magenta	26	Regulation of metabolic process	p53 signaling pathway, Alzheimer's disease	3	ATF6 , NGEF, POGK
	pink	30	Transcription initiation from RNA polymerase II promoter	Basal transcription factors	4	CCDC92 , TMEM70, RNF139, E2F5
	red	93	Regulation of cellular process	Endometrial cancer, Neurotrophin signaling pathway	14	ATP1B1 , STAT3 , ABCB8 , MYC , TGFB1 , SP1 , TP53 , PCGF1, SUMF2, GTF3A, IPO13, GMPPA, HTR6, TGIF1
	turquoise	295	Regulation of cellular metabolic process	p53 signaling pathway, Pancreatic cancer, Apoptosis	2	UBL5, RNF111
	yellow	132	Immune system process	Chemokine signaling pathway, Natural killer cell mediated cytotoxicity	19	APOC1 , CD2 , CD79B , LRRK28 , DAPK1 , FAM124B, EML2, LAP3, TSPAN2, FCRL3, ELMO1, SLC7A7, RASSF5, SLC31A2, TRAF3IP3, GALNT12, ITGA4, SPI1, TFAP2A
	Total	1317				

Module hängen mit Prozessen zusammen, die bereits mit Brustkrebs in Verbindung gebracht werden (endometrical cancer, p53, Prostatakrebs ...)

Hamed et al. BMC Genomics (2015)

Ergänze regulatorische Information + driver genes



Differenziell experimentierte Gene eines Moduls

-> extrahiere regulatorische Interaktionen (TF -> Gen) aus den öffentlichen Datenbanken JASPAR, TRED, MSigDB

Driver genes sind Transkriptionsfaktoren, die möglichst viele Gene des Moduls regulieren.

31% der Driver genes kodieren für Proteine, die Targets für bekannte Krebs-Medikamente sind!

Hamed et al. BMC Genomics (2015)

Ausblick auf den 3. Teil der Vorlesung

- Protein-Protein-Interaktionsnetzwerke – Analyse mit Cytoscape
- metabolische Netzwerke – Simulation mit Copasi
- Ko-Expression / Go-Annotation – Prozessierung mit Bioconductor