

Introduction to Pathway Modeling

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GLADSTONE INSTITUTES
SCIENCE OVERCOMING DISEASE



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What are biological pathways?

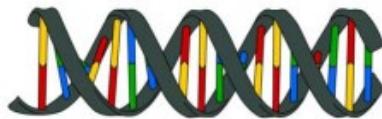
According to the NIH, “A biological pathway is a series of **interactions** among **molecules** in a cell that leads to a certain product or a change in the cell.”



What are biological pathways?

molecules

DNA



Genomics-25,000 genes



What are biological pathways?

interactions

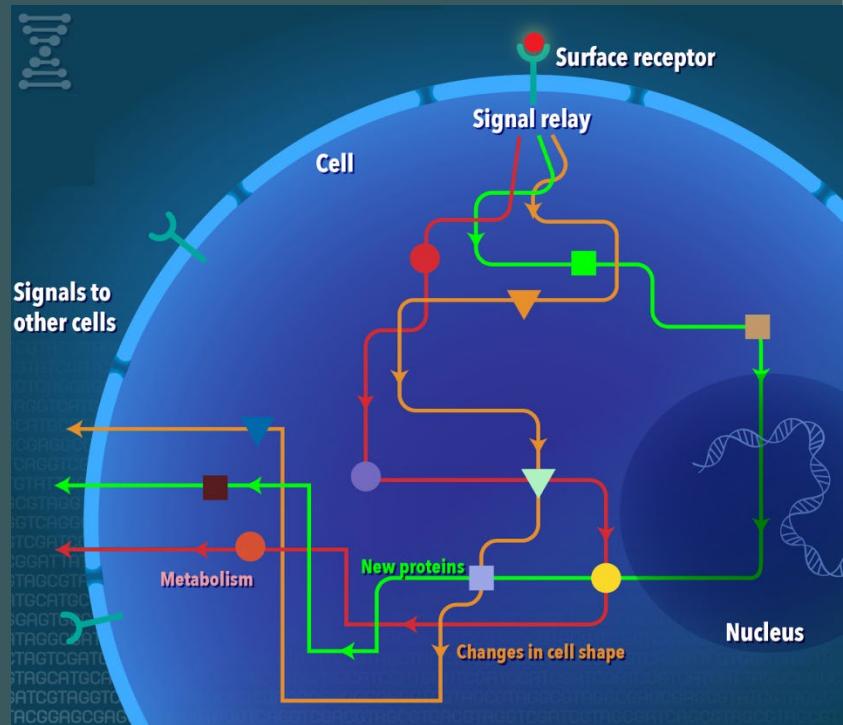
Complex Assembly:





What are biological pathways?

- Signaling pathways
- Metabolic pathways
- Gene regulation pathways



Images:
https://www.genome.gov/images/content/biologic_pathways.jpg

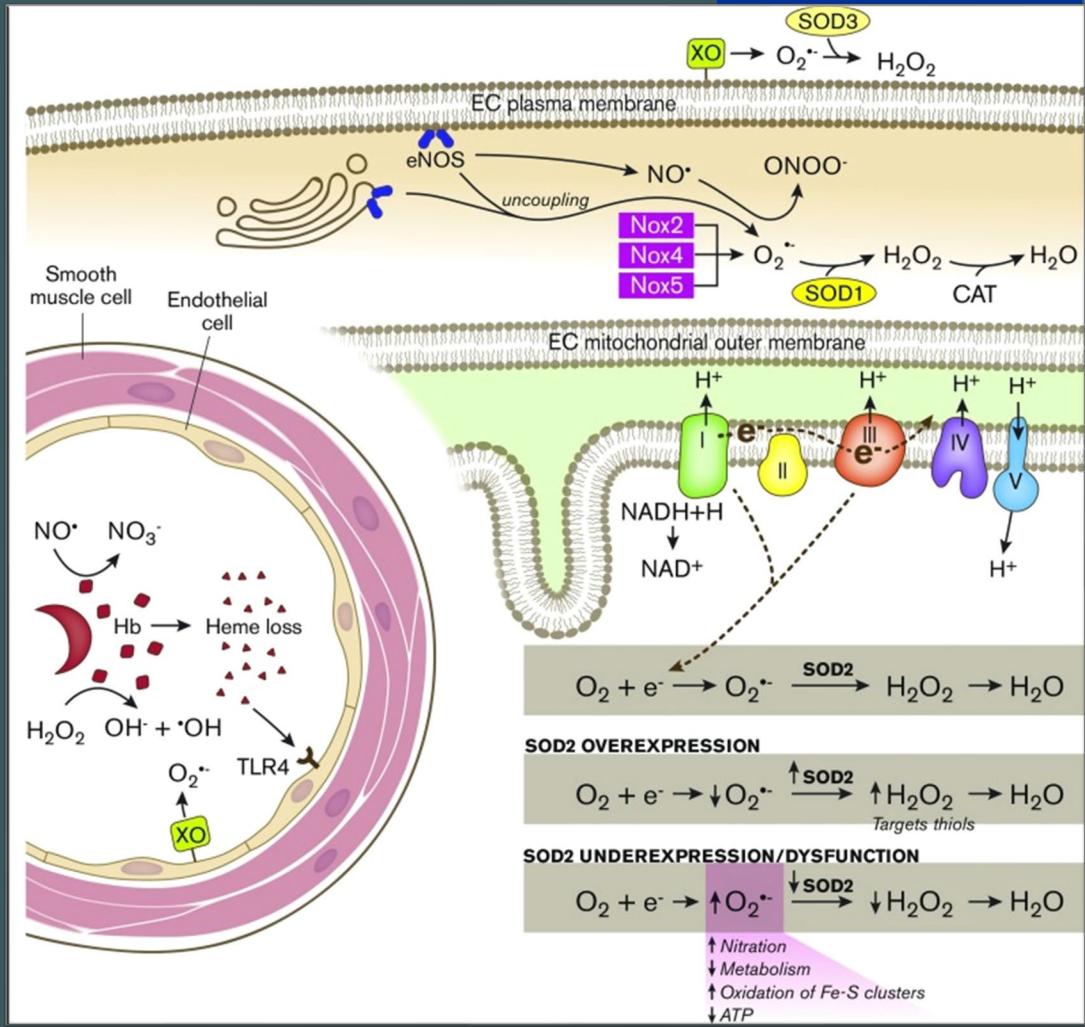
Images:
http://www.genome.gov/multimedia/illustrations/Biological_Pathways.pdf/



Why pathways?

- Intuitive representation of complex information
→ Over 1000 published each month
- Proteins / genes / metabolites
- Interactions / reactions
- Subcellular location

Dosunmu-Ogunbi et al, *Blood Advances*, 2019



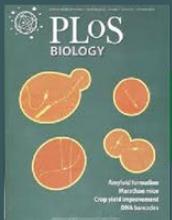
Why pathway models?



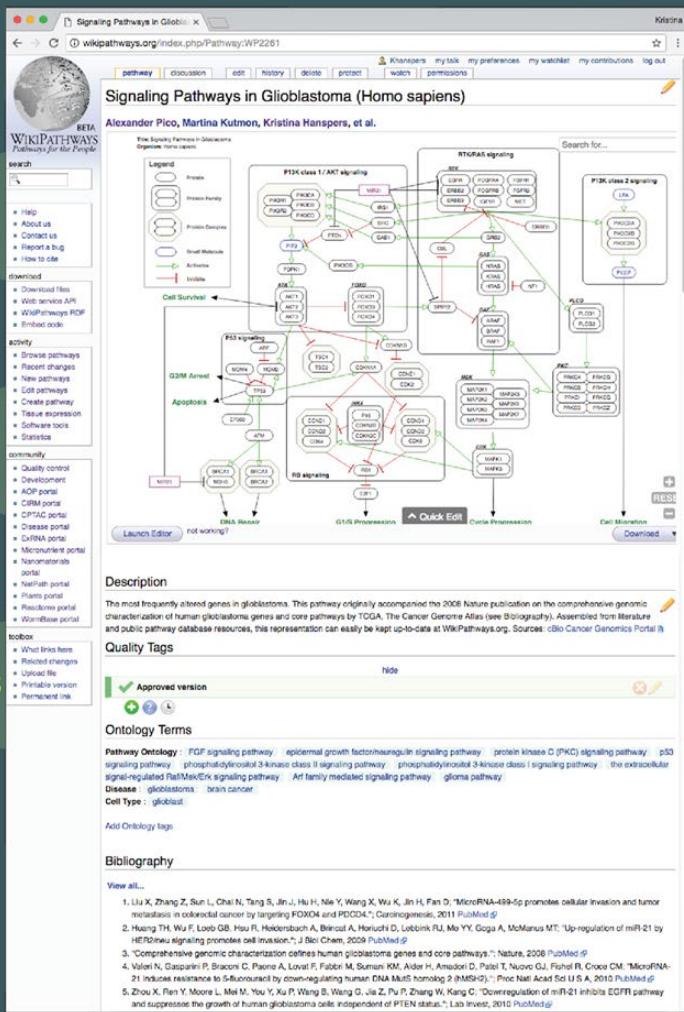
Literature



Experimental data



Static figures



Authorship

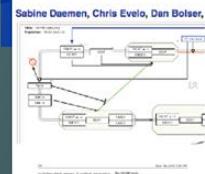
Figure images

Annotated models

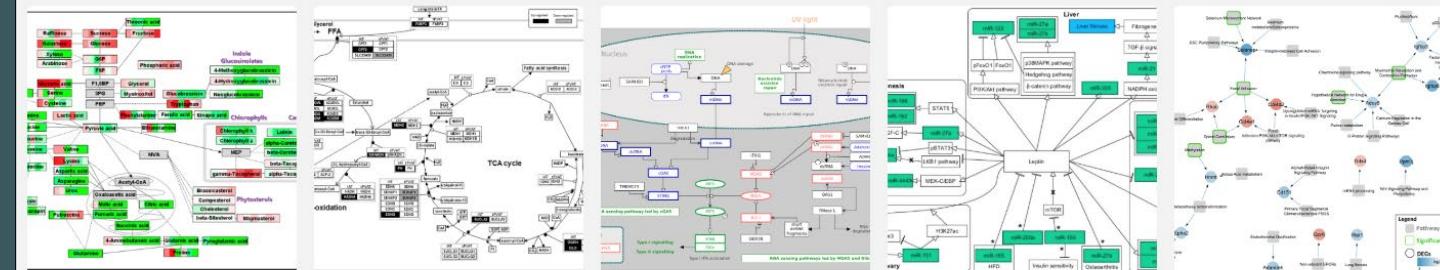
Visualization & analysis

Collaboration

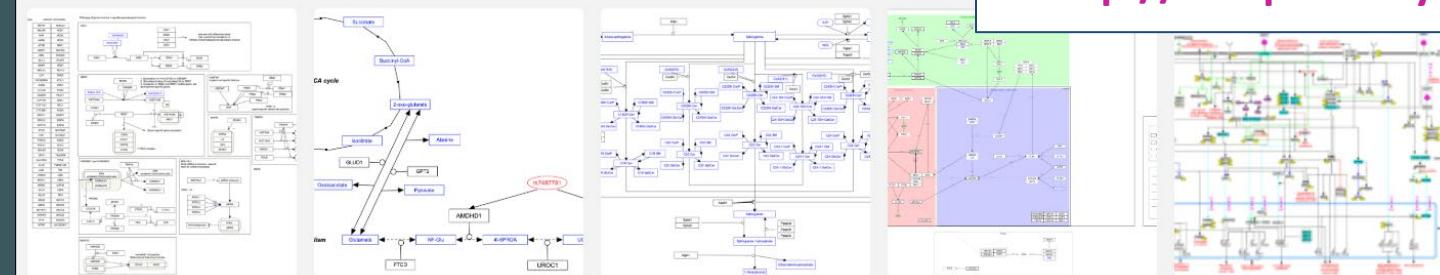
Distributed resource



October 2019



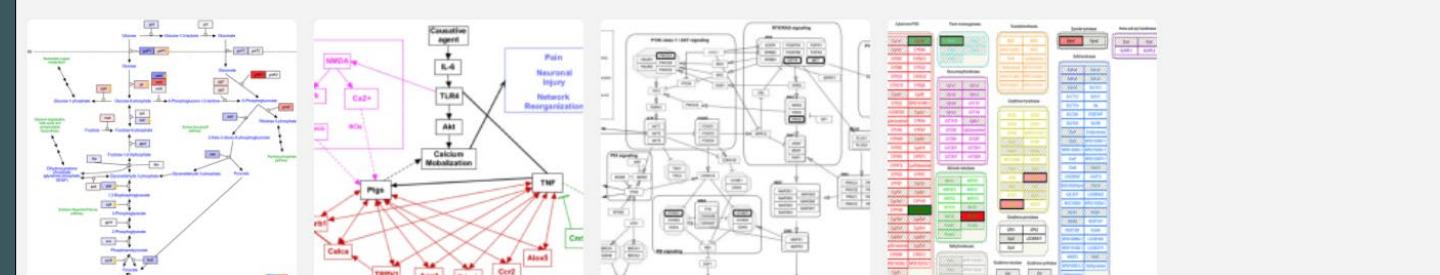
September 2019



WikiPathways Publications

<http://wikopathways.tumblr.com/>

August 2019





What pathway tools will we use?

Editing Tools: WikiPathways and PathVisio



For drawing and editing pathways

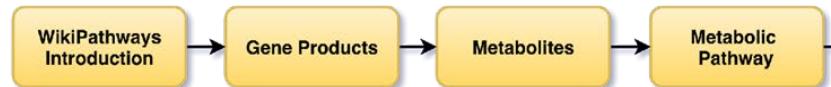


Pathway database



WikiPathways Academy

Biology 101



Walk along a Pathway



Pathway Building Blocks



My First Metabolic Pathway

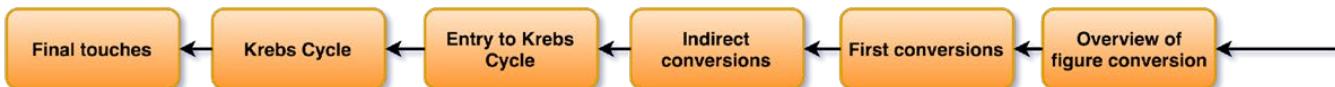


WikiPathways



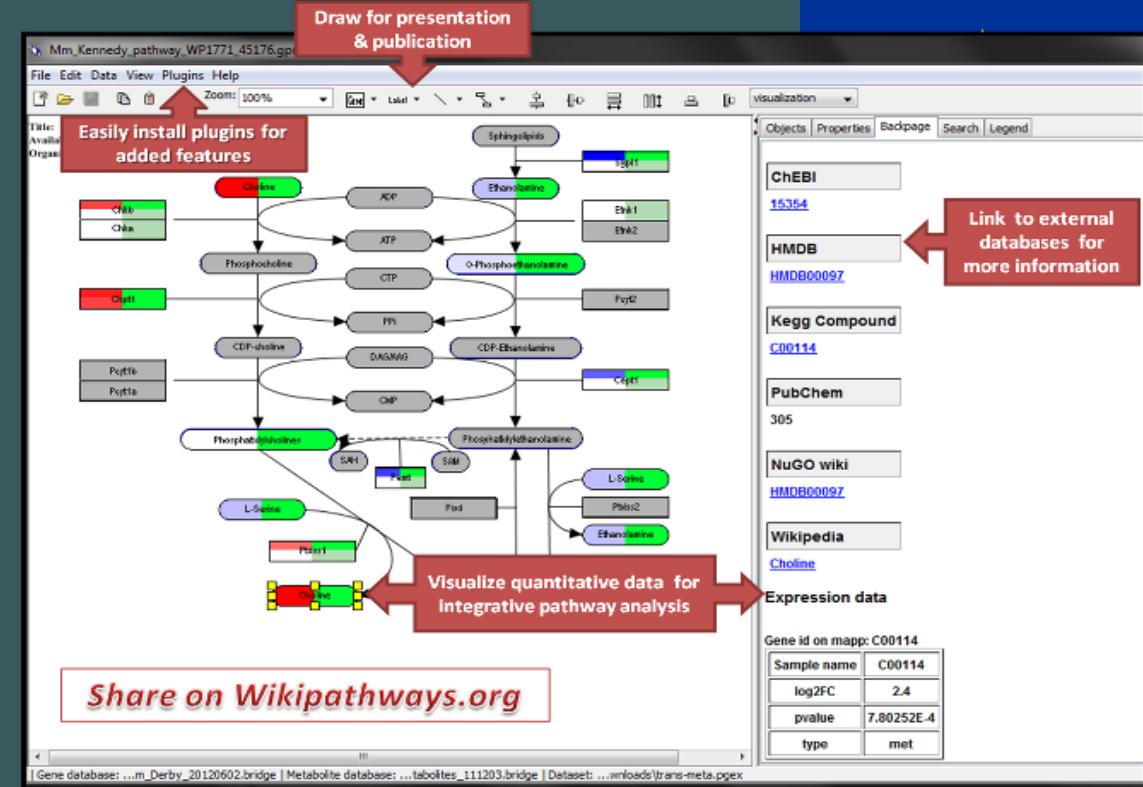
academy.wikipathways.org

Setting Pathway Information Free



PathVisio

PathVisio 3.3.0
www.pathvisio.org

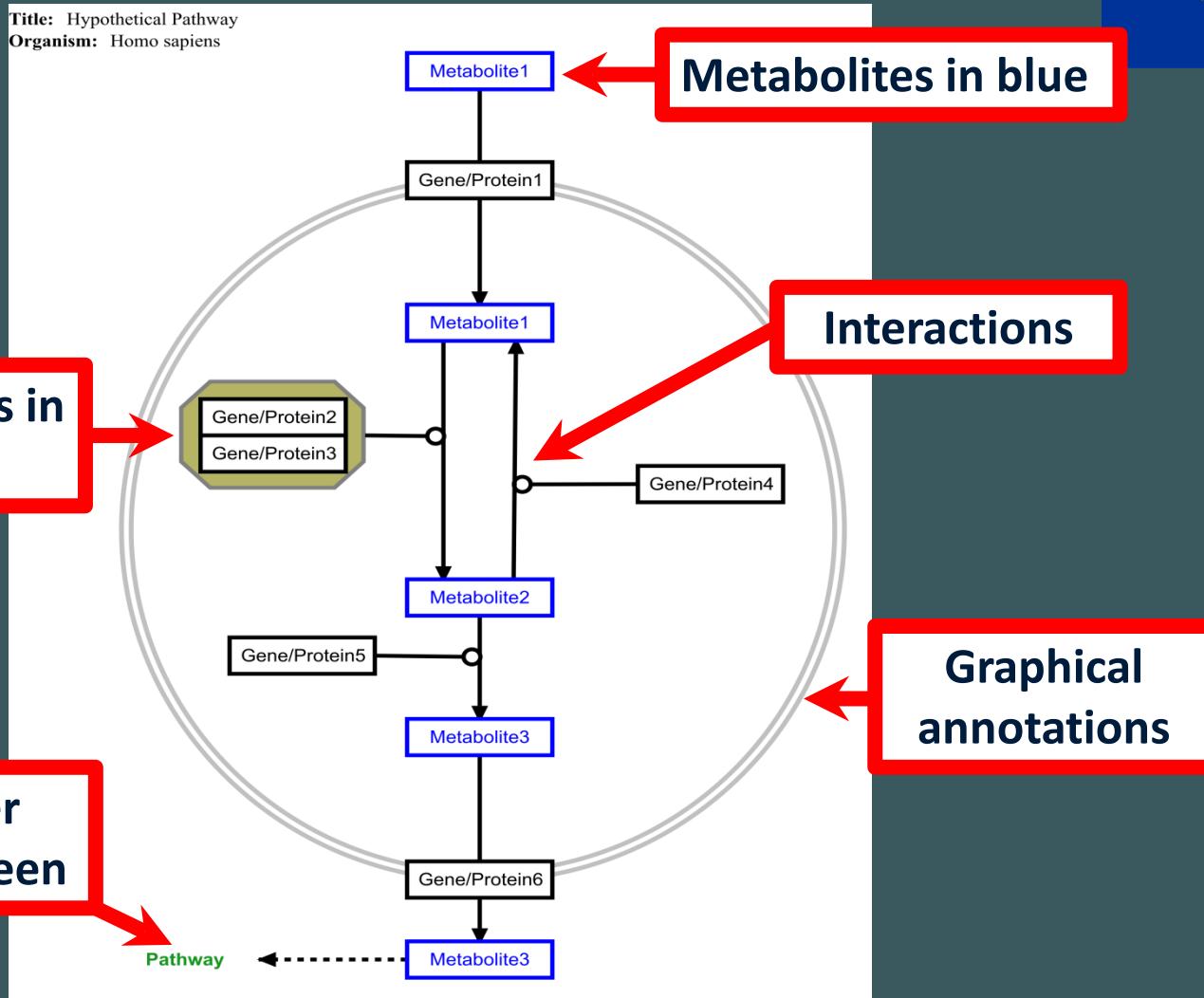


- Java standalone desktop application
- Draw and annotate biological pathways
- Visualize your experimental data on pathways
- Find pathways regulated in your dataset

Pathway Models in PathVisio



Title: Hypothetical Pathway
Organism: Homo sapiens





Pathway Models in PathVisio

More than just images - **fully annotated models**

Adenine
D-Glucose
Acetyl-CoA

p53
BRCA1
MAPK4

DataNode properties

Comments		Literature	Annotation
<input type="text"/> Search <input type="button" value="Search"/>			
Manual entry			
Text label	Adenine		
Biological Type	Metabolite		
Identifier	HMDB00034		
Database	HMDB		
Description Adenine is a purine base. Adenine is found in both DNA and RNA forms adenosine, a nucleoside, when attached to ribose triphosphate (ATP), a nucleotide, when three phosphate groups are added. Adenine is involved in metabolism as one of the basic methods of transferring energy in the cell. Adenine is also involved in serious hereditary disorders, such as Lesch-Nyhan syndrome, which causes neurological deficit, renal disease, self-mutilation and death. Adenine is found in urine and plasma. (OMIM 300322, 229600, 603027, 17052198, 17520339)			

DataNode properties

Comments		Literature	Annotation
<input type="text"/> Search <input type="button" value="Search"/>			
Manual entry			
Text label	p53		
Biological Type	GeneProduct		
Identifier	ENSG00000141510		
Database	Ensembl		
Description p53 is a gene located on chromosome 17. It encodes the tumor suppressor protein p53, which plays a key role in regulating the cell cycle and preventing uncontrolled cell growth. Mutations in the p53 gene have been implicated in many types of cancer.			

links to other databases

e!Ensembl
Home > Human
Location: 17:7,512,445-7,531,642 Gene: TP53

Gene: TP53

- Gene summary
- Splice variants (3)
- Supporting evidence
- Sequence
- External references (7)
- Regulation
- Comparative Genomics
 - Genomic alignments (36)

Gene: TP53 (ENSG00000141510)

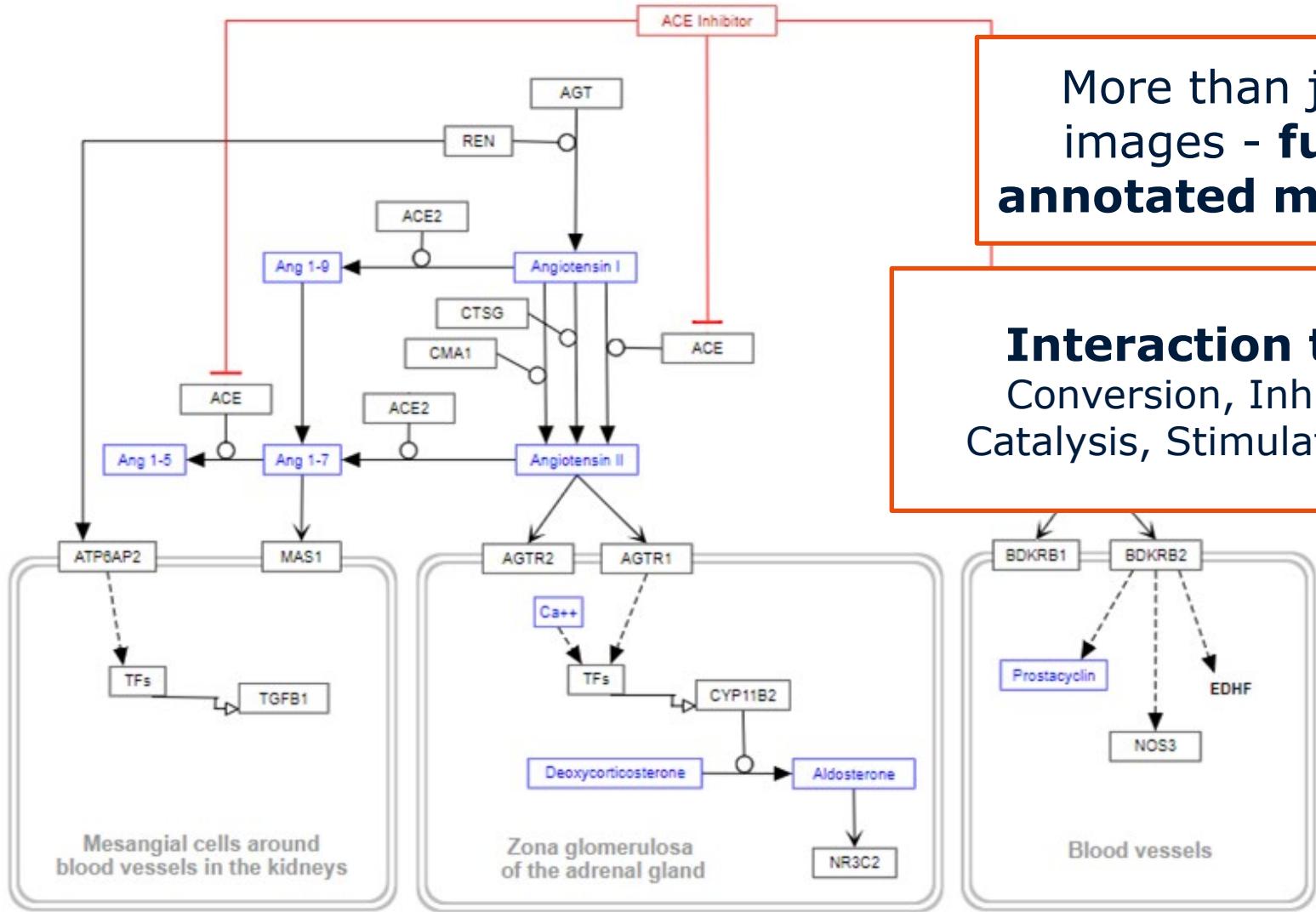
Cellular tumor antigen p53 (Tumor suppressor p53)(Phosphoprotein)

Location Chromosome 17: 7,512,445-7,531,642

Transcripts There are 3 transcripts in this gene: TP53-201, TP53-202, TP53-203

TP53-201	ENST00000370000
TP53-202	ENST00000370001
TP53-203	ENST00000370002

Pathway Models in PathVisio



More than just images - **fully annotated models**

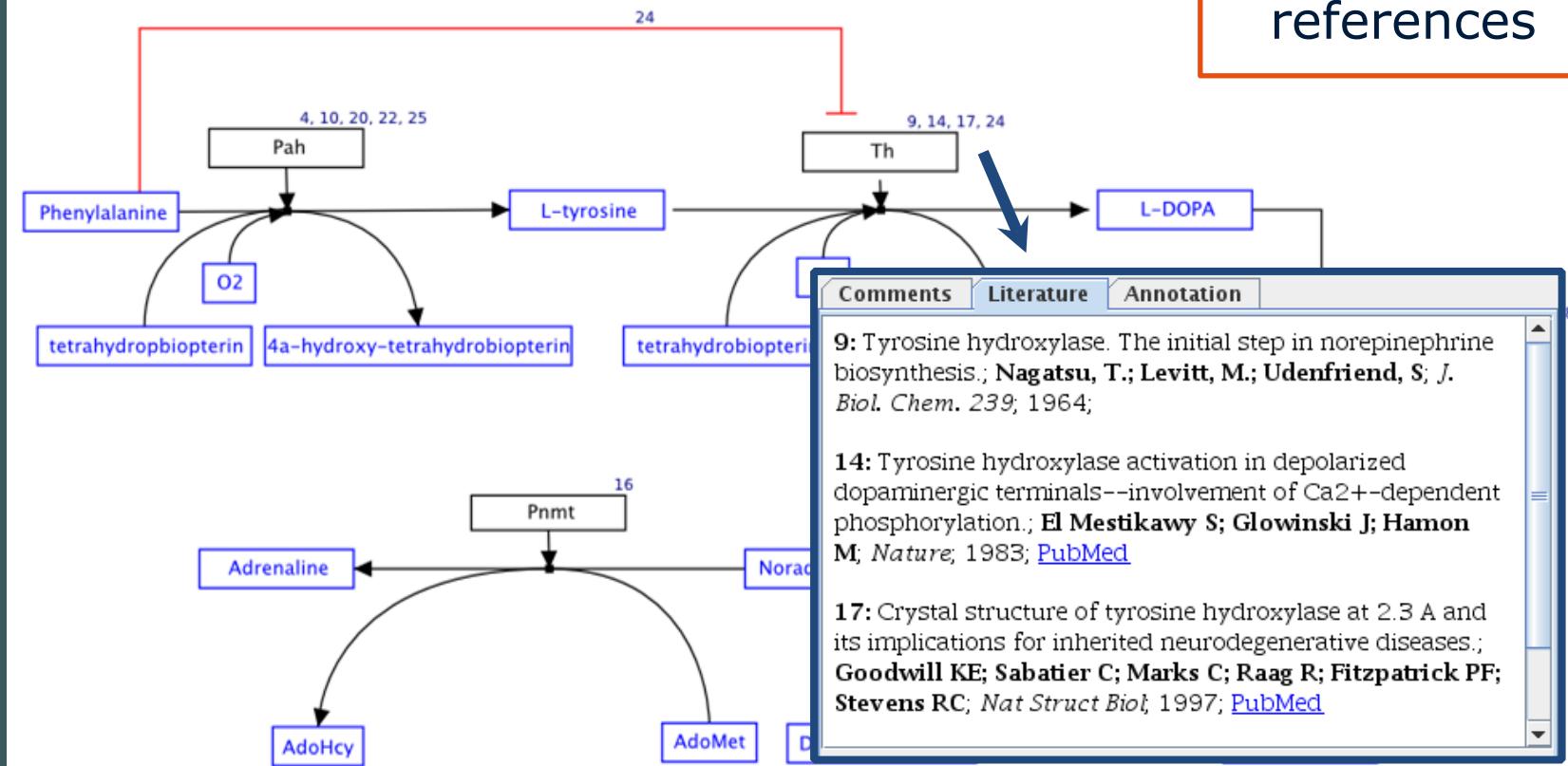
Interaction types
Conversion, Inhibition,
Catalysis, Stimulation, etc.



Pathway Models in PathVisio

More than just
images - **fully
annotated models**

literature
references





Upload to WikiPathways

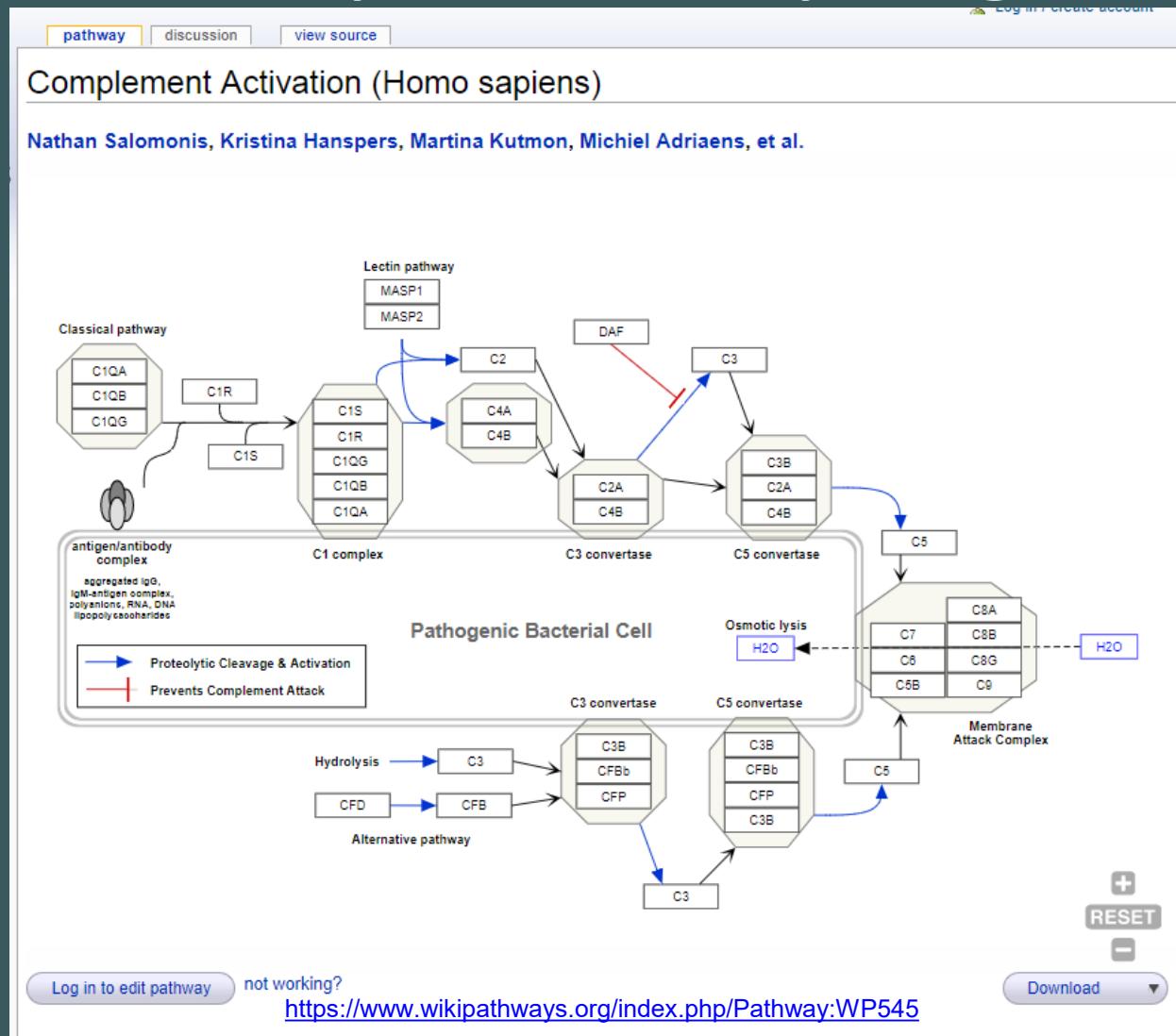
- New pathways can be directly uploaded to WikiPathways through the WikiPathways plugin
- Plugin also allows curators to search for, load and update existing pathways



developed by Sravanthi Sinah

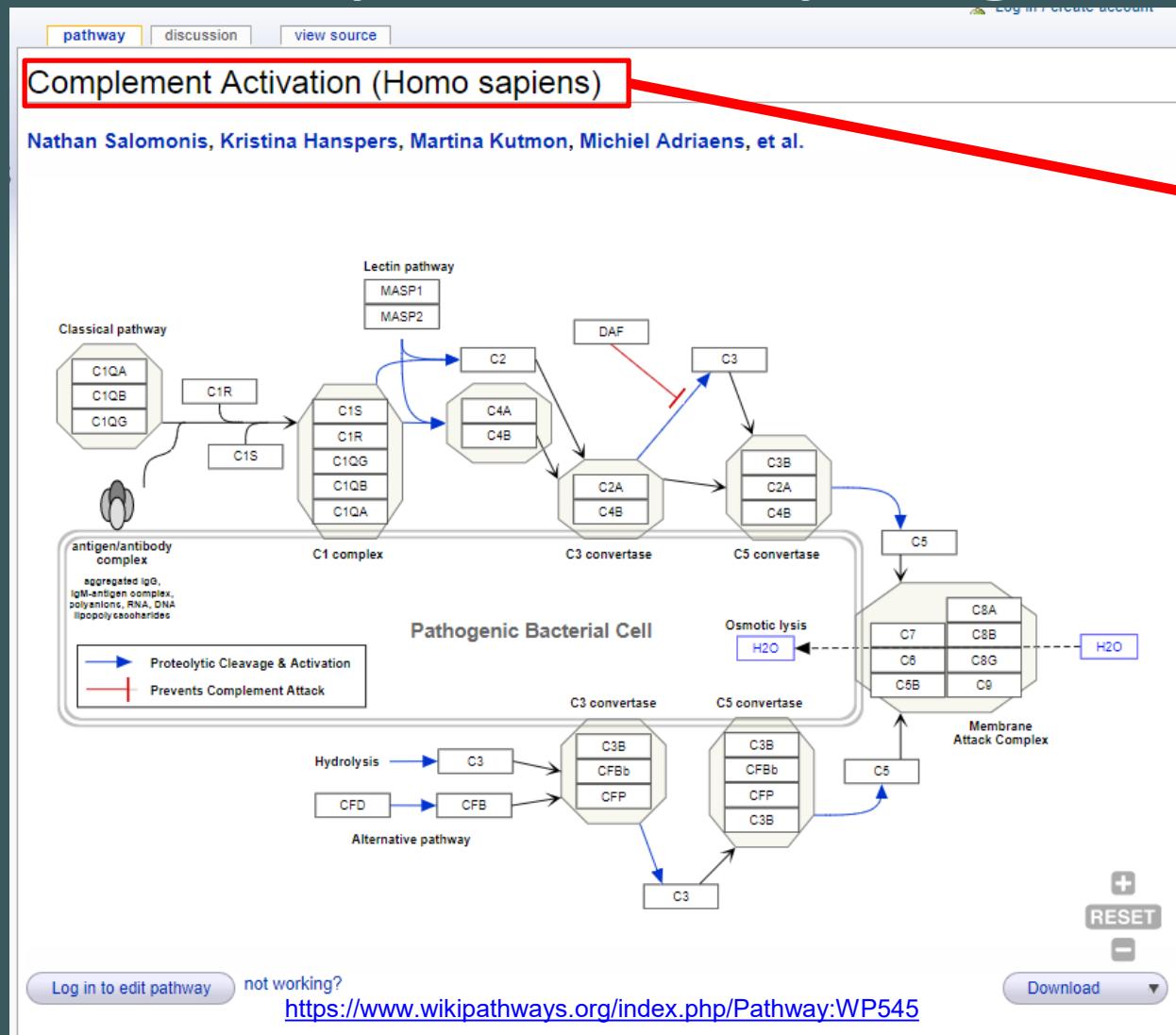


WikiPathways Pathway Pages





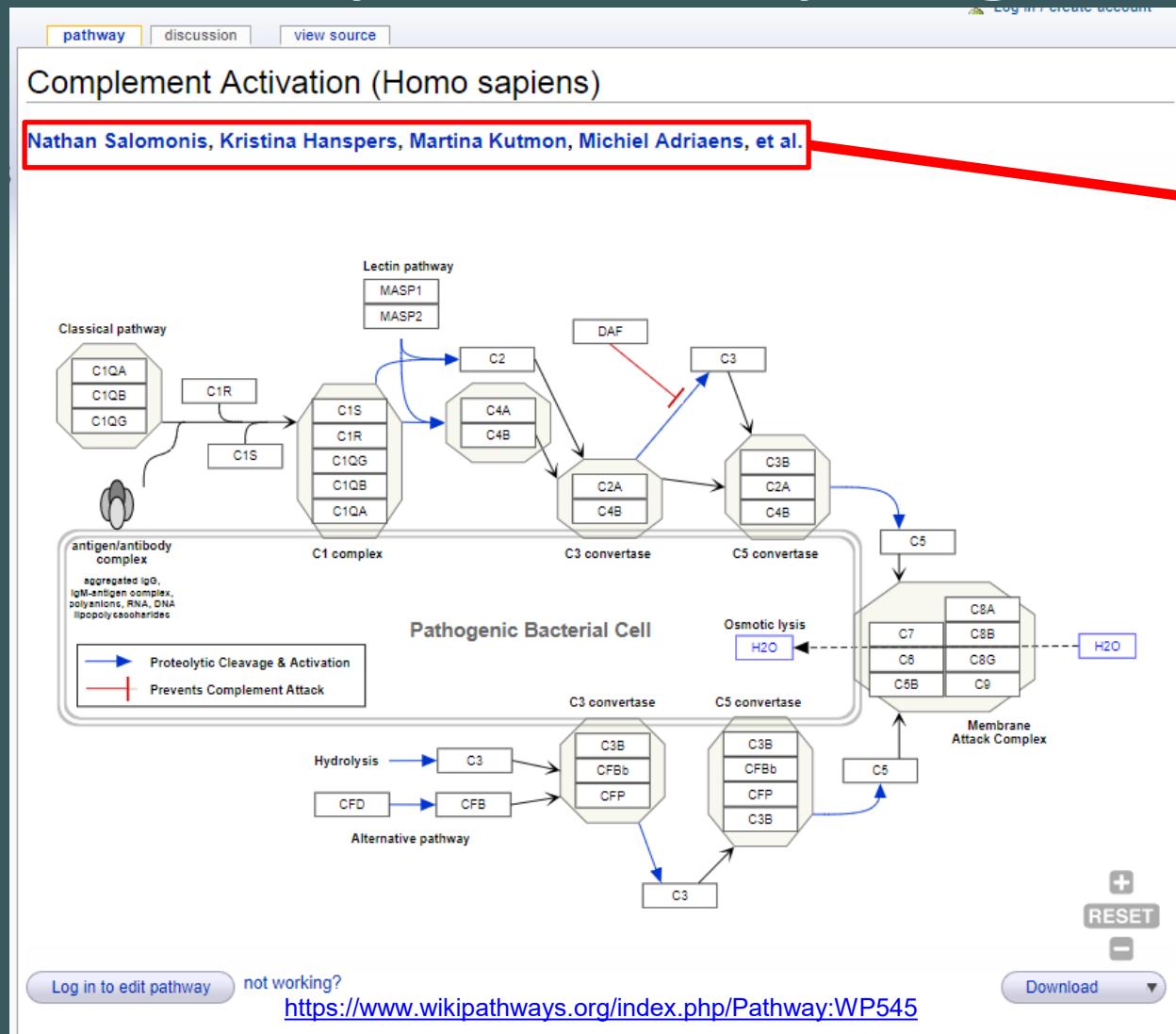
WikiPathways Pathway Pages



Title /
Organism



WikiPathways Pathway Pages



List of
Curators



WikiPathways Pathway Pages

pathway discussion view source Log in / create account

Complement Activation (Homo sapiens)

Nathan Salomonis, Kristina Hanspers, Martina Kutmon, Michiel Adriaens, et al.

The diagram illustrates the three main pathways of complement activation: Classical, Lectin, and Alternative. It shows the proteolytic cleavage of C1, C2, C3, and C5 into active fragments, forming complexes like C3 convertase and C5 convertase, which lead to osmotic lysis or the formation of the Membrane Attack Complex (MAC). The diagram also shows the role of DAF (decay accelerating factor) in preventing complement attack on self-tissue. A red box highlights the central part of the diagram, and a red arrow points from it to a callout box labeled "Interactive Pathway Viewer".

Classical pathway

Lectin pathway

Antigen/antibody complex: aggregated IgG, IgM-antigen complex, polyanions, RNA, DNA, lipopolysaccharides

Pathogenic Bacterial Cell

Proteolytic Cleavage & Activation

Prevents Complement Attack

Osmotic lysis: H₂O

Hydrolysis

CFD

Alternative pathway

Download

Log in to edit pathway not working? <https://www.wikipathways.org/index.php/Pathway:WP545>

RESET

Interactive
Pathway
Viewer



WikiPathways Pathway Pages

Description

The complement system is a biochemical cascade that helps, or complements, the ability of antibodies to clear pathogens from an organism. It is part of the immune system that is not adaptable and does not change over the course of an individual's lifetime. However, it can be recruited and brought into action.

Classical pathway of activation of the complement system is a group of blood proteins that mediate the specific antibody response. [source: Wikipedia]

The Classical pathway begins with circulating C1Q binding to an antigen on the surface of a pathogen, which goes on to activate and recruit 2 copies of C1. The activated C1 complex cleaves C2 and C4. Activated cleavage products C2A and C4B combine to form C3 convertase, which cleaves C3. The cleaved C3 fragments form C5 convertase, which cleaves C5. The cleavage product C5B joins C6, C7, C8 and multiple copies of C9 to form the Membrane Attack Complex, which embeds into the target cell, leading to osmotic lysis. The Decay accelerating factor (DAF) inhibits C3 convertase.

The Lectin pathway involves mannose-binding lectin (MBL) binding the surface of the pathogen instead of C1Q. MBL-associated serine proteases MASP1 and MASP2 act as substitutes for the C1 complex, leading to the formation of C3 convertase and the subsequent cascade.

The Alternative pathway relies on the spontaneous hydrolysis of C3 and the cleavage of factor B (CFB) by factor D (CFD), which form an alternative C3 convertase. Additional copies of the cleavage product C3B are recruited to the complex, resulting in an alternative C5 convertase, which cleaves C5 and contributes to the formation of the Membrane Attack Complex.

Pathway description

Quality Tags

[hide](#)

- Featured version**
- Approved version**
-

Ontology Terms

Pathway Ontology : [innate immune response pathway](#) [immune response pathway](#) [classical complement pathway](#)



WikiPathways Pathway Pages

Description

The complement system is a biochemical cascade that helps, or complements, the ability of antibodies to clear pathogens from an organism. It is part of the innate immune system that is not adaptable and does not change over the course of an individual's lifetime. However, it can be recruited and brought into action by antibodies.

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Quality Tags

[hide](#)

- Featured version
 - Approved version
- [?](#) [L](#)

Quality
Tags

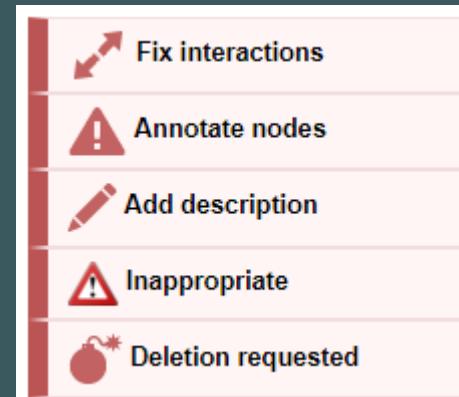
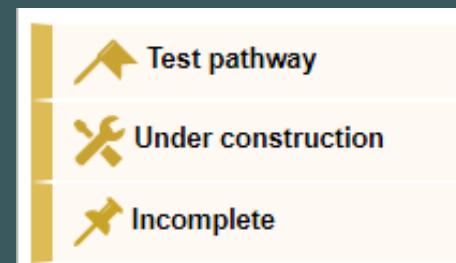
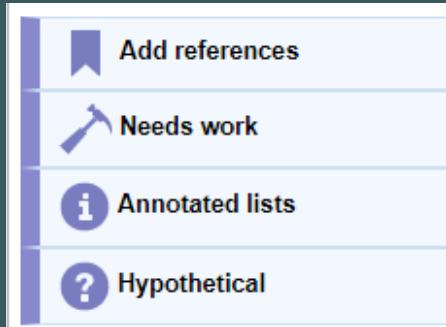
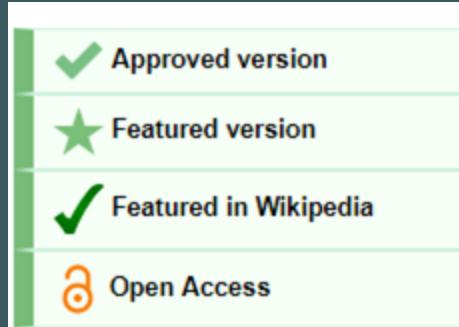
Ontology Terms

Pathway Ontology : [innate immune response pathway](#) [immune response pathway](#) [classical complement pathway](#)



Quality Tags

- Information about the state of the pathway
 - Is the pathway included in any collections?
 - How complete is the pathway?





WikiPathways Pathway Pages

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The Alternative pathway relies on the spontaneous hydrolysis of C3 and the cleavage of factor B (CFB) by factor D (CFD), which form an alternative C3 convertase. Additional copies of the cleavage product C3B are recruited to the complex, resulting in an alternative C5 convertase, which cleaves C5 and contributes to the formation of the Membrane Attack Complex.

Quality Tags

[hide](#)

- Featured version
- Approved version
-

Ontology Terms

Pathway Ontology : [innate immune response pathway](#) [immune response pathway](#) [classical complement pathway](#)

Ontology
Terms



Ontology Terms

- Controlled vocabularies to categorize the pathway
- Support of three ontologies
 - <http://bioportal.bioontology.org/>
 - Pathway, disease and cell type ontology



Ontology Terms

Ontology Terms

Pathway Ontology : innate immune response pathway immune response pathway classical complement pathway

[Hide Ontology Options](#)

Type Ontology term..

To add a tag, either select from the available ontology trees below or type a search term in the search box.

Pathway Ontology

- + classic metabolic pathway
- + disease pathway
- + drug pathway
- + regulatory pathway
- + signaling pathway

Disease

- + disease by infectious agent
- + disease of anatomical entity
- + disease of cellular proliferation
- + disease of mental health
- + disease of metabolism
- + genetic disease
- + physical disorder
- + syndrome

Cell Type

- + abnormal cell
- + cell in vitro
- + compound eye retinal cell
- + native cell
- + neural cell
- + neuron



WikiPathways Pathway Pages

Bibliography

1. Brook E, Herbert AP, Jenkins HT, Soares DC, Barlow PN; "Opportunities for new therapies based on the natural history of C1Q nephropathy".

History

[View all...](#)

[Compare selected versions](#)

Compare	Revision	Action	Time	User	Comment
<input checked="" type="radio"/>	89636 Featured Approved	view	06:10, 22 September 2016	Egonw	Connected lines.
<input checked="" type="radio"/> <input type="radio"/>	87260	view	14:39, 20 July 2016	MaintBot	missing graphids
<input type="radio"/> <input checked="" type="radio"/>	82142	view	01:23, 9 September 2015	AlexanderPico	fixed C1Q complex
<input type="radio"/> <input checked="" type="radio"/>	82141	view	01:19, 9 September 2015	AlexanderPico	Modified description
<input type="radio"/> <input checked="" type="radio"/>	82140	view	01:13, 9 September 2015	AlexanderPico	Modified title

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External references

DataNodes

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Name	Type	Database reference	Comment
C1QA	GeneProduct	712 (Entrez Gene)	update: C1QA HUMAN
C1QB	GeneProduct	713 (Entrez Gene)	
C1QG	GeneProduct	714 (Entrez Gene)	update C1QC HUMAN
C1R	GeneProduct	ENSG00000159403 (Ensembl)	
C1S	GeneProduct	716 (Entrez Gene)	

Annotated Interactions

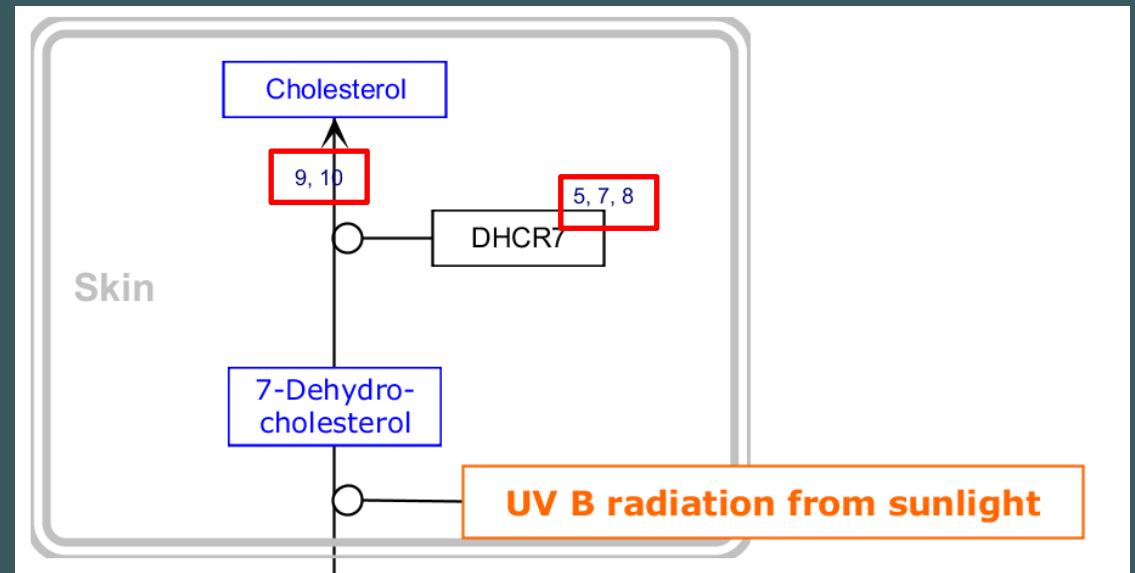
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Bibliography



Bibliography

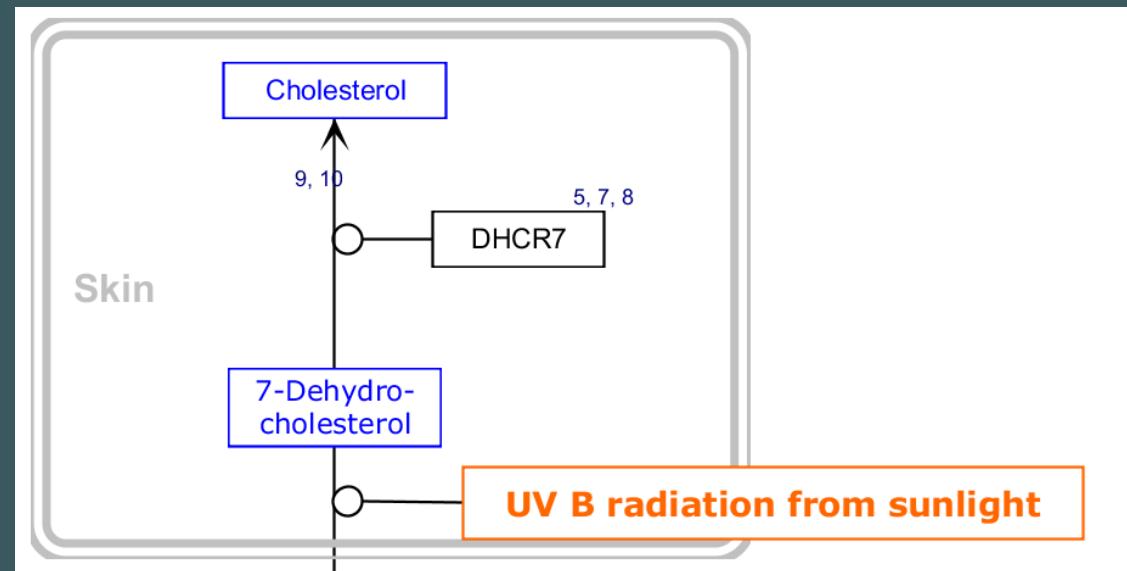
- Every element of the pathway can be linked to literature references including





Bibliography

- Every element of the pathway can be linked to literature references including
 - Data nodes
 - Interactions
 - Pathway





WikiPathways Pathway Pages

Bibliography

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History



WikiPathways Pathway Pages

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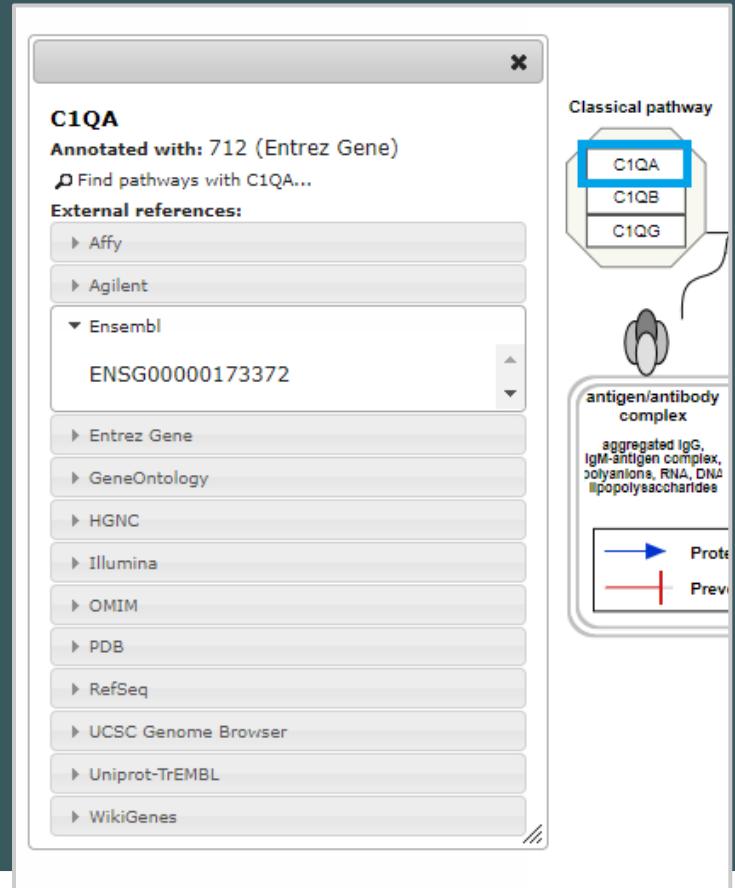
No annotated interactions

External
References

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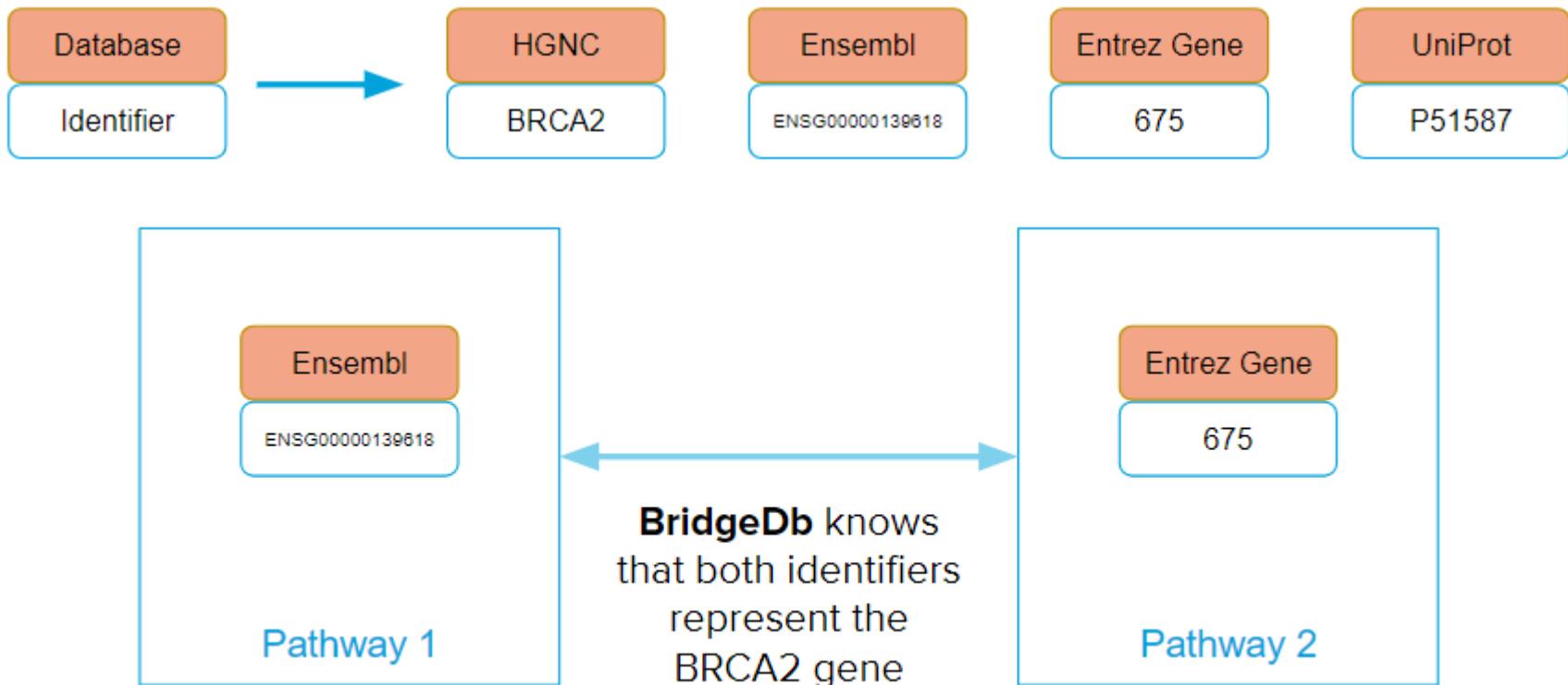


- Powered by BridgeDb
 - Mapping databases available for gene products, metabolites and interactions
 - Identifier mapping framework
 - www.bridgedb.org
- Pathway curators can use their identifier system of choice





External References





Community Portals

page discussion view source history

Welcome to WikiPathways BETA

WikiPathways is a database of biological pathways maintained by a community of experts.

Find Pathways

Search

You can search by:

- Pathway name (*Apoptosis*)
- Gene or protein name (*p53*)
- Any page content (*cancer*)

Get Pathways

Download

Multiple formats and methods

Edit Pathways

Learn

Play with the editor tools

community

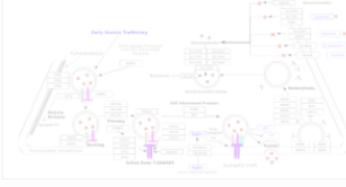
- Quality control
- Development
- WikiPathways Blog
- AOP portal
- CIRM portal
- CPTAC portal
- Renal Genomics portal
- Disease portal
- ExRNA portal
- Lipids portal
- Micronutrient portal
- Nanomaterials portal
- NetPath portal
- Plants portal
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- Reactome portal
- WormBase portal

community

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Today's Featured Pathway

Synaptic Vesicle Pathway (*Homo sapiens*)



Curator of the Week



Susan Coort (Maastricht University) ↗

Updates

- November 2019 Release: 172 edits by 19 contributors and 15 new pathways.
- October 2019 Release: 728 edits by 16 contributors and 5 new pathways
- September 2019 Release: 905 edits by 15 contributors and 5 new pathways
- August 2019 Release: 416 edits by 13 contributors and 8 new pathways
- July 2019 Release: 284 edits by 21 contributors and 18 new pathways
- June 2019 Release: 270 edits by 29 contributors and 4 new pathways
- May 2019 Release: 243 edits by 23 contributors and 14 new pathways
- April 2019 Release: 213 edits by 20 contributors and 17 new pathways

more...

Discussions

9 November 2019

Welcome to Wikopathways! (12) by Daniela Digles

more...

Forum

Re: [wp-discuss] I kindly request to help me in importing and mapping ID

Helis Academy

Interreg Vlaanderen-Nederland

Europes Fonds voor Regionale Ontwikkeling

Maastricht Universi

JOINT PROGRAMME

DISEASES



Community Portals

portal discussion view source history Log in / create account

Portal:RareDisease

Rare Disease pathways at WikiPathways

This portal highlights WikiPathways content related to rare genetic human diseases, and is designed as a central organizing point for exploring, curating and expanding the collection of rare disease pathways.

A rare or orphan disease is a disease which affects relatively few people. The exact definition varies between 1:1000 and 1:200.000. E.g. it can be 1:1500 (USA) , 1:2000 (EU) or 1:2500 (Japan). Rare diseases are mostly caused by genetic variation making them chronic and hard to cure. The severity depends on the affected gene and its physiological implications.

This portal is funded by:

**EUROPEAN JOINT PROGRAMME
RARE DISEASES**

elixir

**Netherlands
RETT
EXPERTISE CENTRE**

Rare Disease Pathways

Featured Pathway

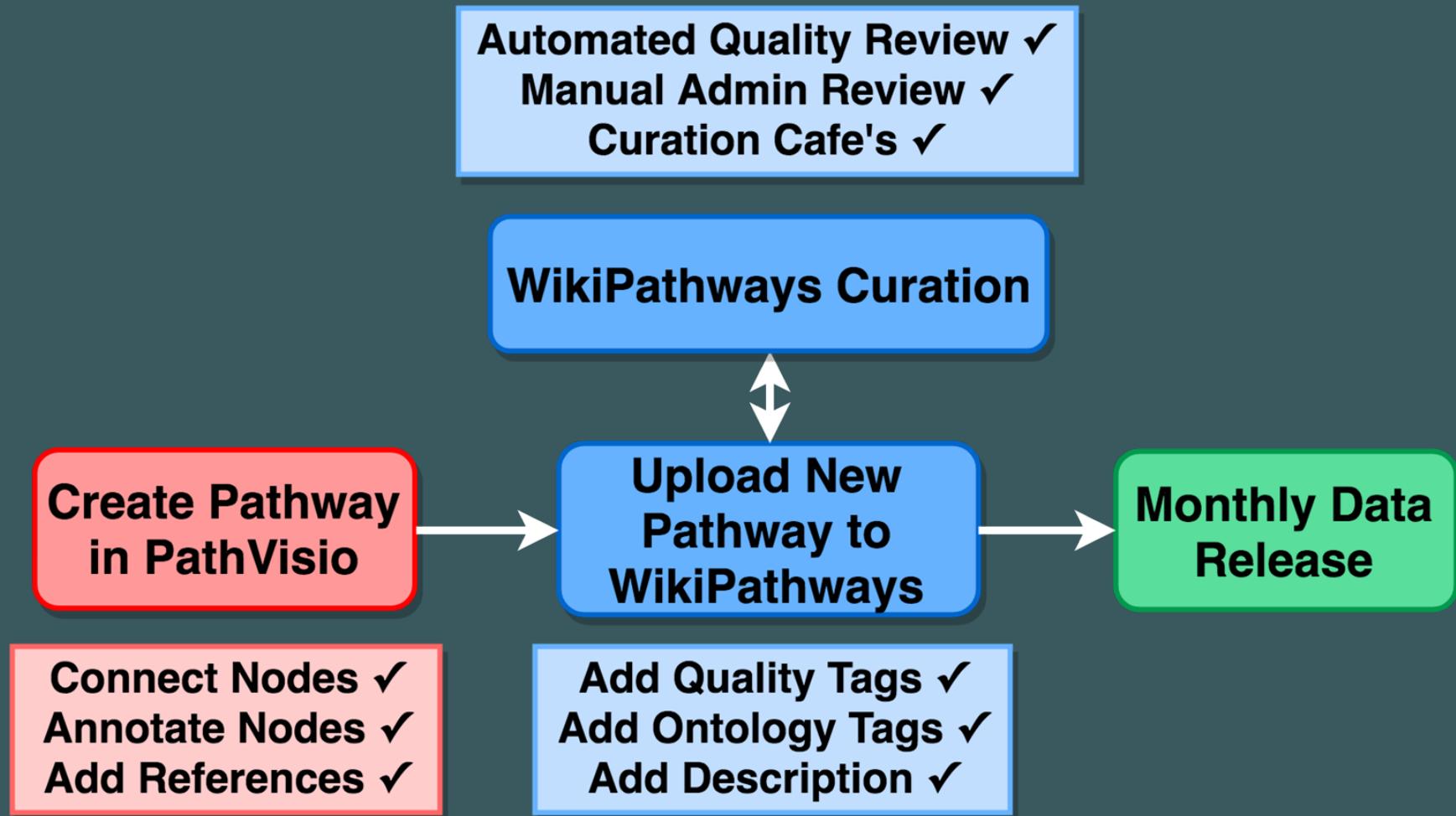
Explore rare disease pathways on WikiPathways

- MECP2 and Associated Rett Syndrome
- Amyotrophic lateral sclerosis (ALS)
- Fanconi Anemia Pathway
- The effect of progerin on the involved genes in Hutchinson-Gilford Progeria Syndrome
- Lamin A-processing pathway
- Viral Acute Myocarditis

Neurodegeneration with brain iron accumulation (NBIA) subtypes pathway (Homo sapiens)



Life-Cycle of a Pathway Model at WikiPathways



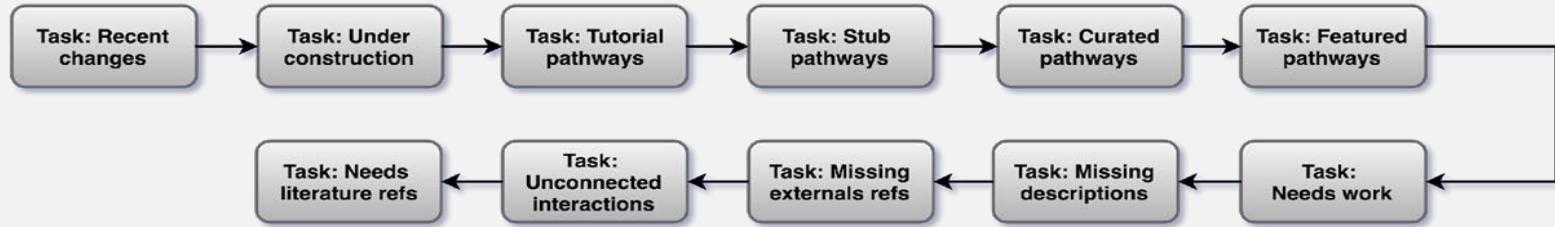


Quality Control Protocol

QA Team



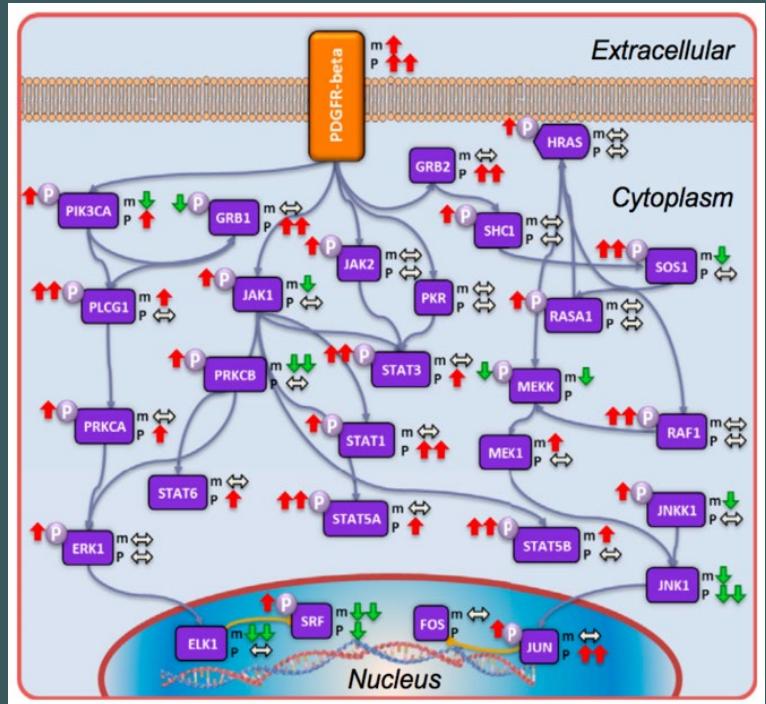
QA Protocol



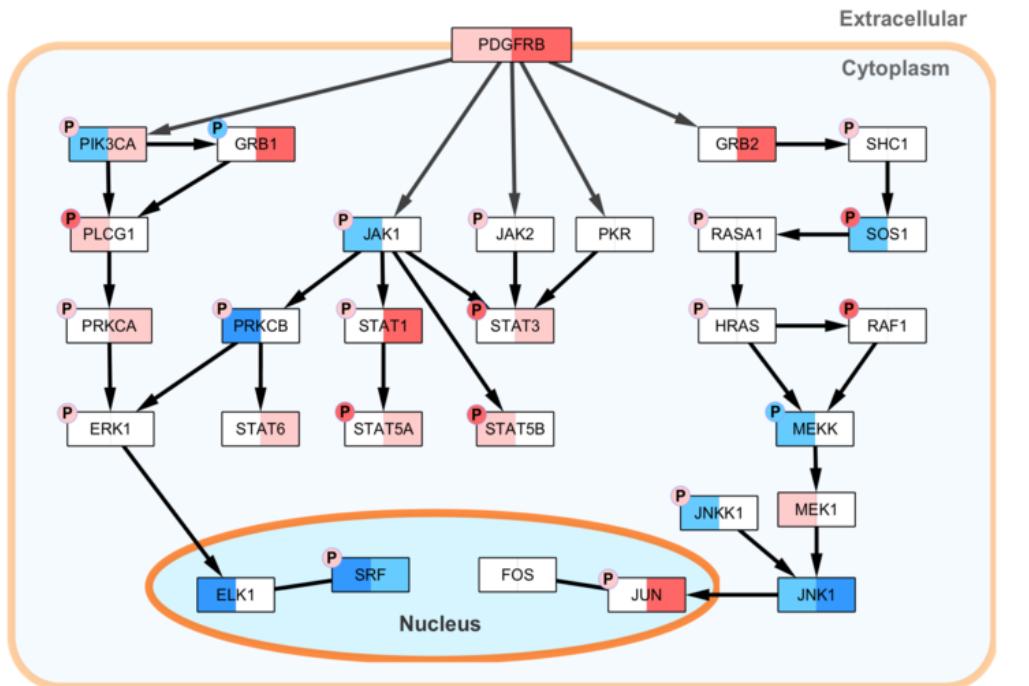
<https://wikopathways.github.io/academy/qaprotocol.html>



Machine readability



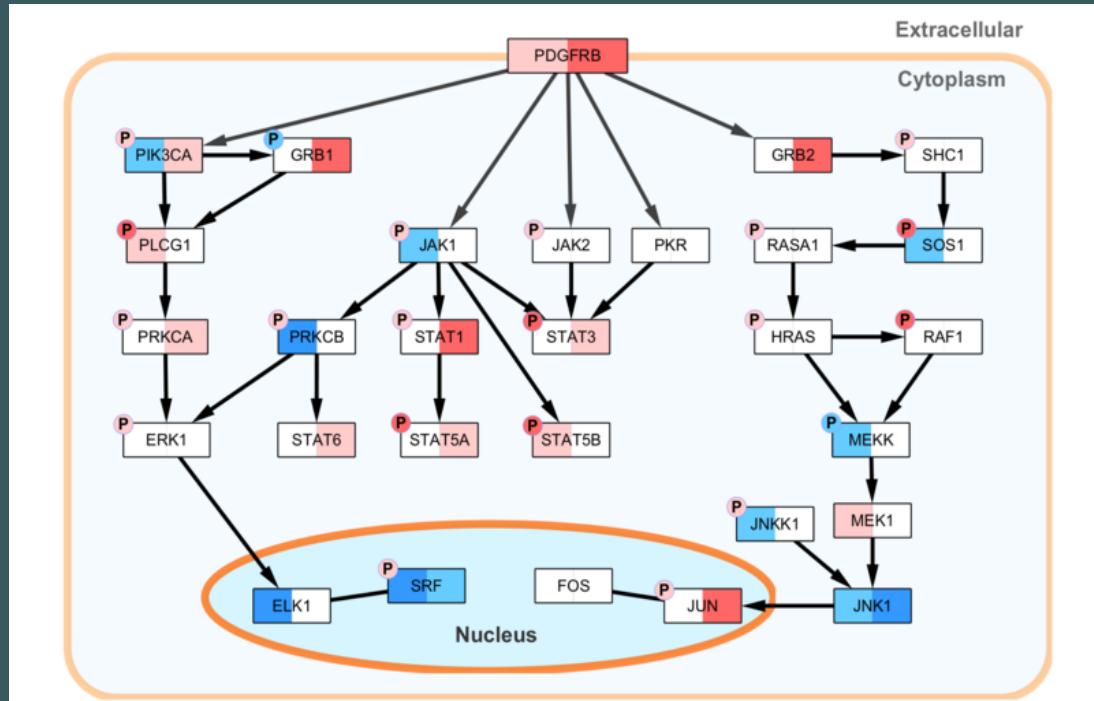
Static image
Zhang et al, Cell 2016



PDGFR-beta pathway with transcriptomic/phosphoproteomic data
Zhang et al, Cell 2016



Machine readability



PDGFR-beta pathway with
transcriptomic/phosphoproteomic data
Zhang et al, *Cell* 2016



Machine readability



www.wikipathways.org

How do we use
them for analysis?





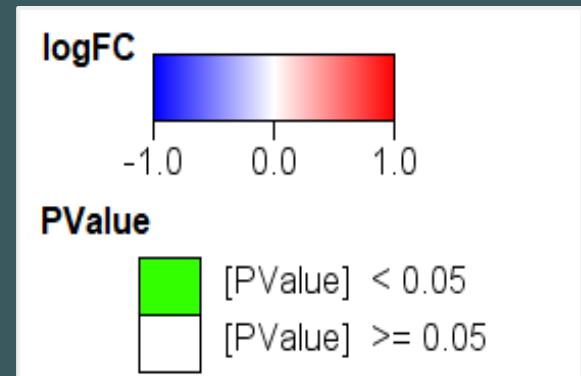
Data Import

- Wizard guiding you through data import
- Multiple identifier systems supported
 - Ensembl, NCBI Gene, Affymetrix, Illumina, HGNC
 - Focus on gene expression for practical
 - PathVisio can handle other types of omics data
- BridgeDb takes care of ID mapping (.bridge file)
- Database (.pgex), log file and default visualization are created for each imported dataset

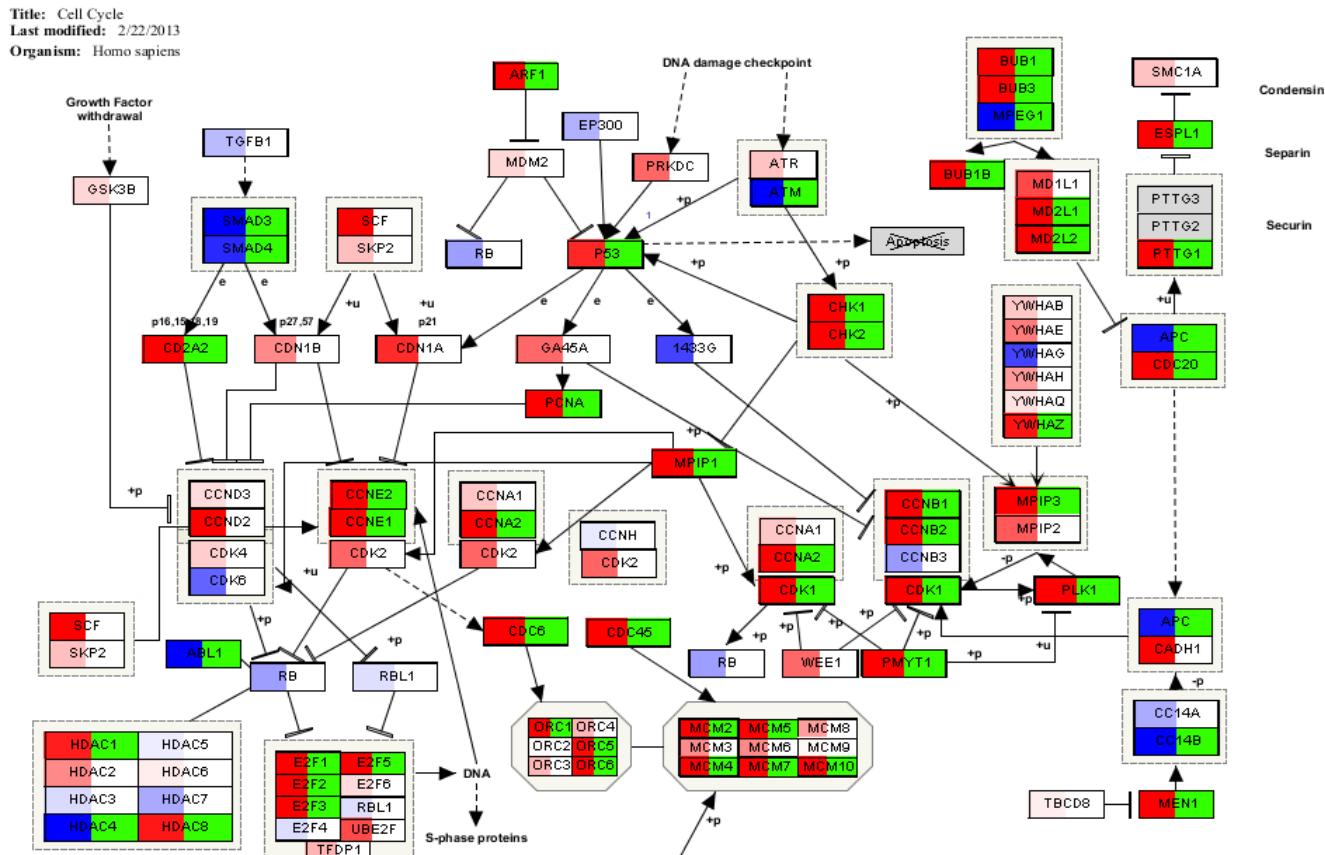
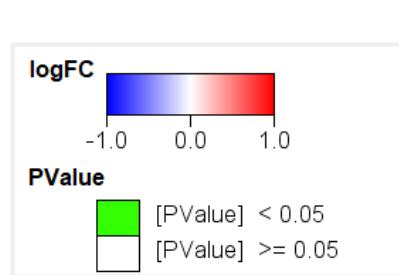


Data Visualizations

- Data visualization on data nodes and interactions
- Visualization styles
 - Color gradient (continuous data like log2FC)
 - Color rules (distinct categories like pval cutoff)
- Multi-omics visualization
- Time-series visualization



Data Visualizations

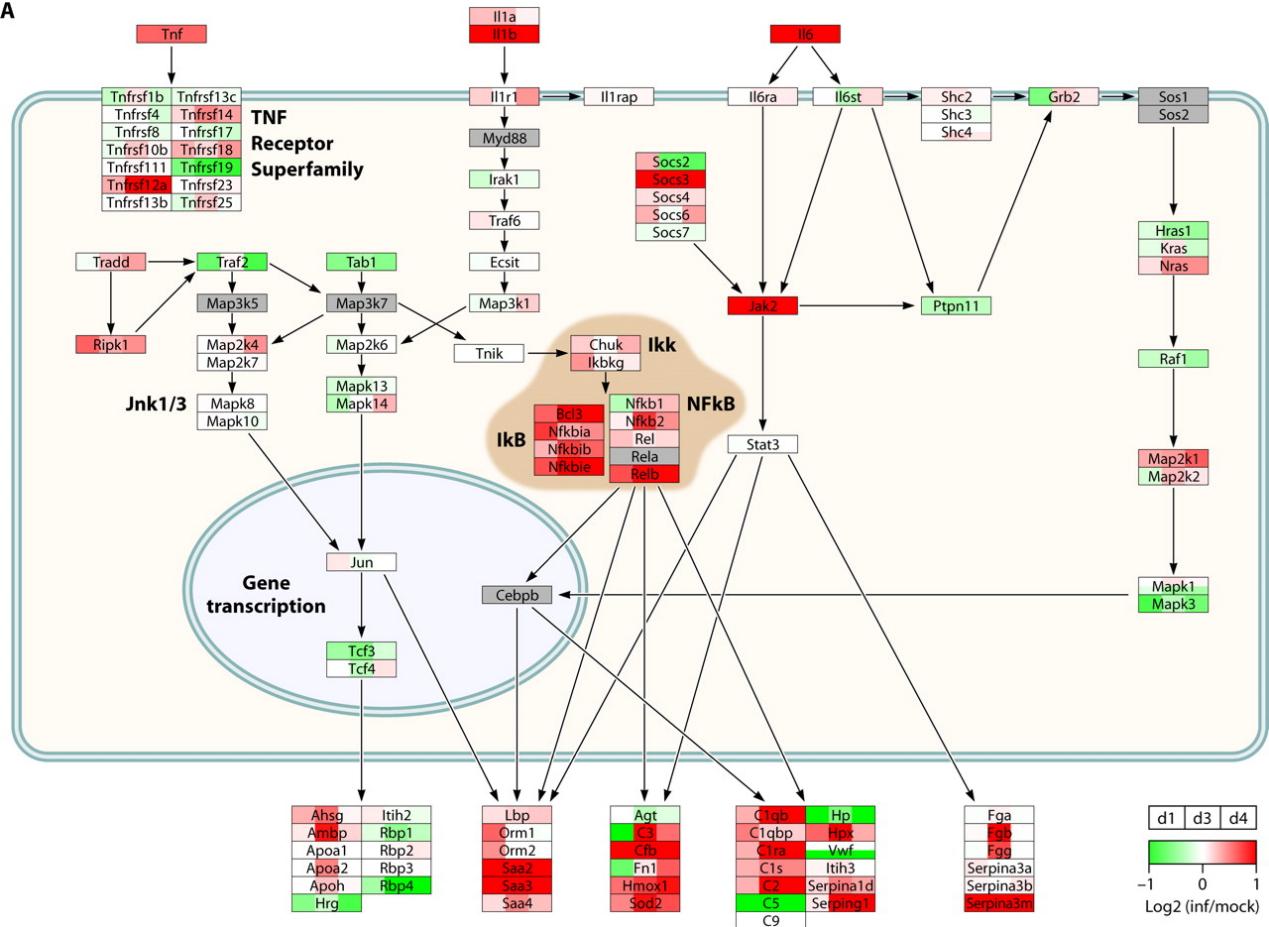


Transcriptomics cancer dataset visualized on the human Cell Cycle Pathway: <https://www.wikipathways.org/instance/WP179>



Data Visualizations

A

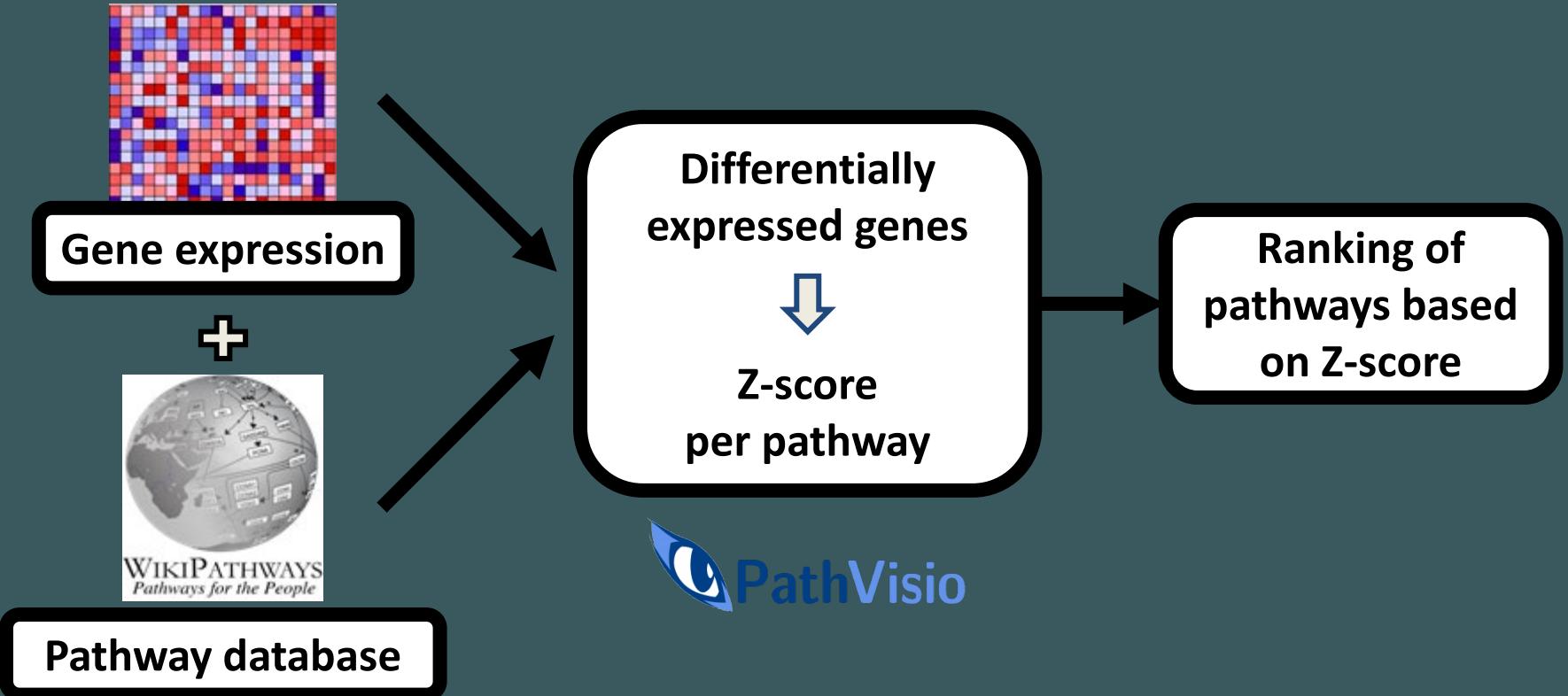


Time-series data visualization
(virus infection over multiple days)
Into the eye of the cytokine storm.

<https://doi.org/10.1128/MMBR.05015-11>



Pathway Statistics





Pathway Analysis Methods

- **Overrepresentation analysis:**
 - Input list → e.g. significantly up- or down-regulated genes
 - Background list → e.g. all measured genes
 - Statistical test → e.g. Fisher's exact test (hypergeometric test)

$$Z\text{-score} = \frac{(r - n \frac{R}{N})}{\sqrt{n \frac{R}{N} \left(1 - \frac{R}{N}\right) \left(1 - \frac{n-1}{N-1}\right)}}$$

- Z-Score is calculated for each pathway
 - Results in ranked list of pathways
- Four variables in the formula: N, R, n, r

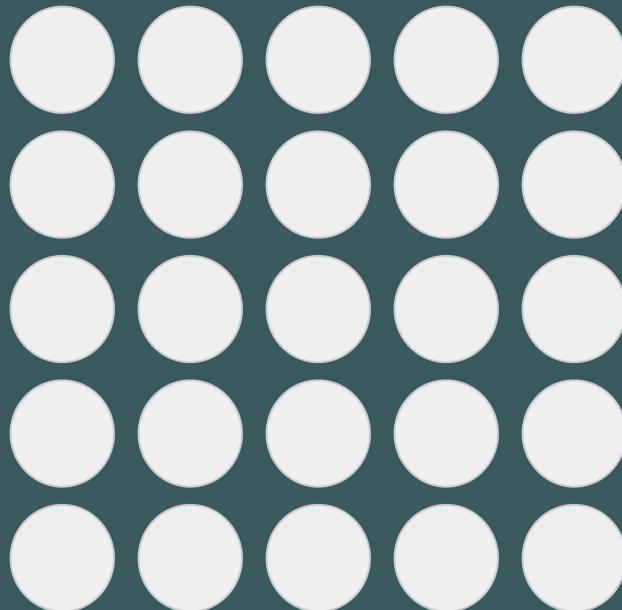


Z-Score ORA

$$Z\text{-score} = \frac{(r - n \frac{R}{N})}{\sqrt{n \frac{R}{N} (1 - \frac{R}{N})(1 - \frac{n-1}{N-1})}}$$

N = 25

background list (total number of measured genes in experiment)





Z-Score ORA

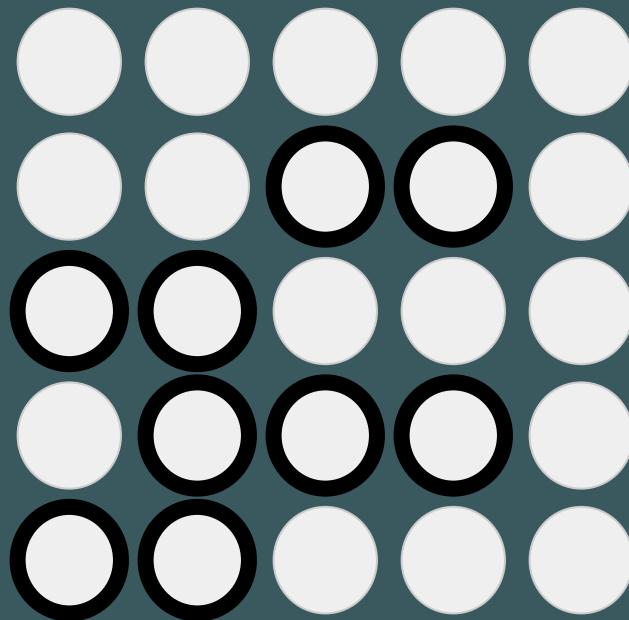
$$Z\text{-score} = \frac{(r - n \frac{R}{N})}{\sqrt{n \frac{R}{N} (1 - \frac{R}{N}) (1 - \frac{n-1}{N-1})}}$$

N = 25

background list (total number of measured genes in experiment)

R = 9

input list (number of changed genes in experiment)





Z-Score ORA

$$Z\text{-score} = \frac{(r - n \frac{R}{N})}{\sqrt{n \frac{R}{N} (1 - \frac{R}{N}) (1 - \frac{n-1}{N-1})}}$$

N = 25

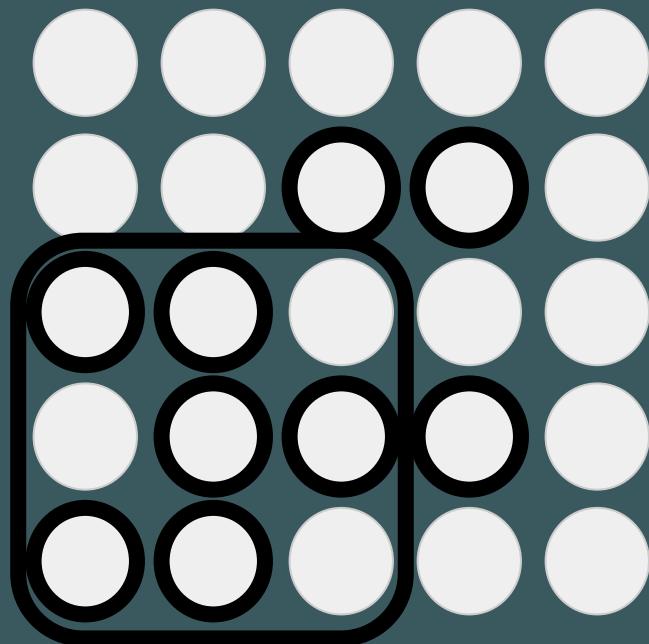
background list (total number of measured genes in experiment)

R = 9

input list (number of changed genes in experiment)

n = 9

total number of genes in pathway



Pathway X



Z-Score ORA

$$Z\text{-score} = \frac{(r - n \frac{R}{N})}{\sqrt{n \frac{R}{N} (1 - \frac{R}{N}) (1 - \frac{n-1}{N-1})}}$$

N = 25

background list (total number of measured genes in experiment)

R = 9

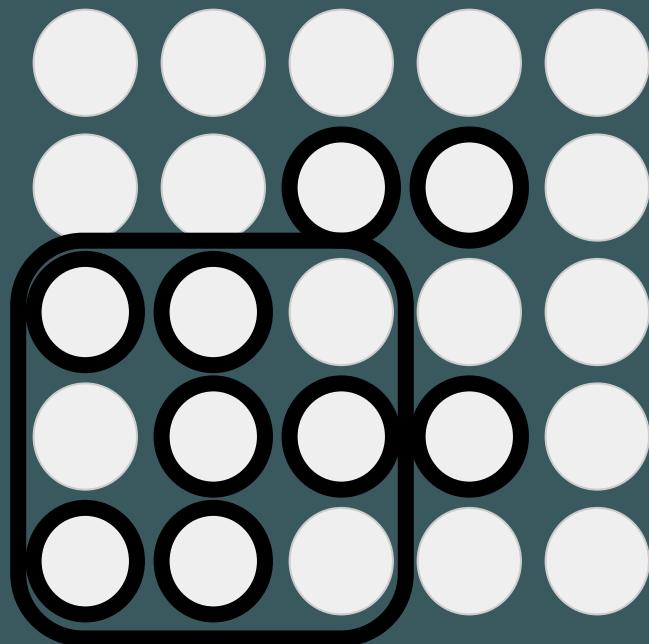
input list (number of changed genes in experiment)

n = 9

total number of genes in pathway

r = 6

number of changed genes in pathway



Pathway X



Z-Score ORA

$$Z\text{-score} = \frac{(r - n \frac{R}{N})}{\sqrt{n \frac{R}{N} (1 - \frac{R}{N}) (1 - \frac{n-1}{N-1})}}$$

N = 25

background list (total number of measured genes in experiment)

R = 9

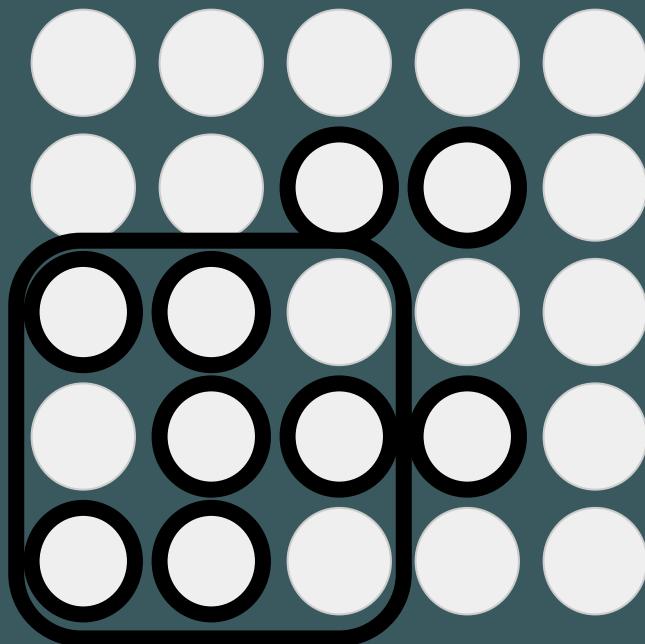
input list (number of changed genes in experiment)

n = 9

total number of genes in pathway

r = 6

number of changed genes in pathway



Pathway X

Z-Score for pathway X = 2.347



Z-Score ORA

- What does the Z-Score tell you?
 - Z-Score > 1.96
 - Significantly more genes than expected are changed in the pathway → altered pathways in the experiment (different between the groups)
 - Z-Score = 0
 - Distribution of changed genes in the pathway is the same as in the complete dataset
 - Z-Score = < -1.96
 - Significantly less genes than expected are changed in the pathway → very stable pathway (not affected in experiment)



Pathway Statistics

- Overrepresentation analysis to identify altered pathways in a dataset of interest
- Hypergeometric test (Fisher's exact test)

Up-regulated pathways (log2FC > 2, p-value < 0.05)	Z-score	Perm. p-value
Cell Cycle	6.12	0.001
G1 to S cell cycle control	4.26	0.002
Synaptic Vesicle Pathway	3.89	0.001
DNA Damage Response	3.88	0.002
ATM Signaling Pathway	3.80	0.001

Down-regulated pathways (log2FC < -2, p-value < 0.05)	Z-score	Perm. p-value
Complement and Coagulation Cascades	5.87	0.001
Complement Activation	5.84	0.001
Adipogenesis	5.49	0.001
Differentiation of white and brown adipocyte	5.44	0.001
Triacylglyceride Synthesis	4.53	0.001



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INSTITUTES**



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and many contributors and curators
around the world



On to the practical!