

miRNet Tutorial Input Process

Computer and Browser Requirements

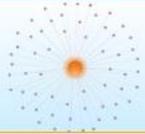
- **A modern web browser with Java Script enabled Chrome, Safari, Firefox, and Internet Explorer 9+**
- **For best performance and visualization, use:**
 - Latest Google Chrome
 - At least 4GB of physical RAM
 - A 15-inch screen or bigger

Goal for this tutorial

➤ Upload different data types to miRNet

- 1) A list of miRNA, mRNA and long non-coding RNA (lncRNA);
- 2) Items selected for associated diseases, small molecules and epigenetic modifiers;
- 3) miRNA/mRNA expression data sets.

Upload a list of miRNA or targets



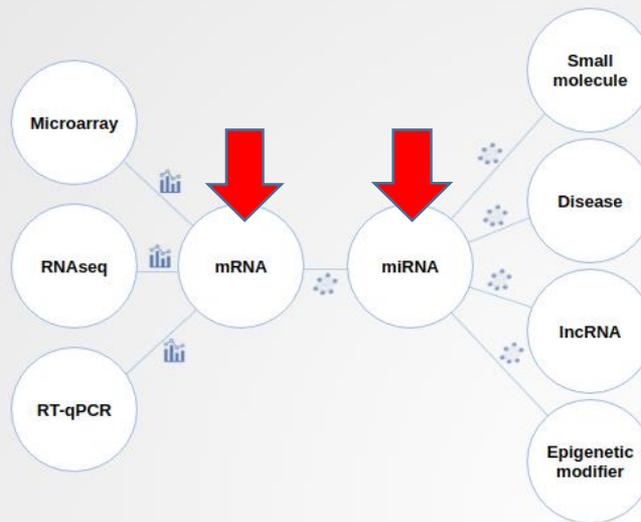
miRNet -- network-based visual analysis of miRNAs, targets and functions

Home ? FAQs Tutorials Resources About

News & Updates

- Added tissue-specific miRNA annotation for human miRNAs (12/26/2017); **NEW**
- miRNet can now automatically recognize and correct pre-miRNAs to their mature forms (12/16/2017); **NEW**
- Updated miRNA gene targets based on new releases of miRTarbase and TarBase (11/16/2017); **NEW**
- Code refactoring for performance improvement (10/25/2017); **NEW**
- Fixed the issue for broken miRBase links (09/25/2017);
- Added support for reducing node overlap for large network layout (09/22/2017);
- Added Force Atlas layout algorithm to

Click on an input area below to start analysis



Selecting one of the data types.

Overview

miRNet is an easy-to-use tool with comprehensive support for **statistical analysis** and **functional interpretation** of data generated in miRNAs studies. Its main features include:

- **Support for various inputs & statistics:** miRNet accepts a list of miRNAs or targets, or a data table from microarray, RNAseq or RT-qPCR experiments. miRNet supports differential analysis using *limma*, *edgeR* and HTqPCR methods; enrichment analysis using standard hypergeometric tests and unbiased random sampling.
- **Comprehensive functional annotation:** miRNet integrates data from eleven different miRNA databases - [TarBase](#), [miRTarBase](#), [miRecords](#), [miRanda](#) (*S. mansoni* only), [miR2Disease](#), [HMDD](#), [PhenomiR](#), [SM2miR](#), [PharmacomiR](#), [EpimiR](#), and [starBase](#). It currently supports nine organisms - Human, Mouse, Rat, Cattle, Chicken, Zebra fish, Fruit fly, *C. elegans* and *S. mansoni*.
- **Creation of miRNA-target interaction networks:** miRNet provides a wide array of options to allow researchers to build miRNA-target interaction networks at different confidence levels. The resulting network can be further optimized using different algorithms to improve visualization and understanding.
- **High-performance network visual analytics:** miRNet offers five types of networks on miRNA-gene, miRNA-disease, miRNA-small molecule, miRNA-IncRNA, and miRNA-epigenetic modifier. The system supports zooming, highlighting, point-and-click, drag-and-drop, enrichment analysis, etc. to enable users to intuitively explore miRNAs, targets and functions.

Please Cite

Fan Y, Siklenka, K., Arora, SK., Ribeiro, P., Kimmins, S. and Xia, J. (2016) [miRNet - dissecting miRNA-target interactions and functional associations through network-based visual analysis](#). Nucl. Acids Res. 44 W135-141

1. Upload a list of miRNA

miRNA Upload

Enter a list of miRNAs below: ?

Organism: H. sapiens (human)

ID type: miRBase ID

Tissue (human only): Kidney (551)

Target type: Genes

hsa-mir-101-3p
hsa-mir-133b
hsa-mir-147a
hsa-mir-3140-3p
hsa-mir-361-5p
hsa-mir-510-5p

Submit

Proceed

Try Examples

- 8 organisms
- 2 ID types : miRBase ID and Accession number
- 5 different targets
- 53 tissues for human miRNA

1. Choose the parameters above.
Copy and paste a list of miRNA

You can try our example first

3. Click "Proceed" to the interaction table

2. Click "Submit". A confirm dialog will show on the top right corner.

2. Upload a list of gene

🏠 ▶ Gene Upload

Enter a list of genes below: ?

Organism: H. sapiens (human)

ID type: Official gene symbol

Tissue (human only): Bone marrow (598)

ACAT1
DDX3X
MEGF9
FMR1
HAPLN1
IL12A
OLR1
RNF6
SOD3
ELOVL4
MRAP2

Submit

Proceed

Try Examples

You can try our example first

1. Choose the parameters above. Copy and paste a list of mRNA here.

2. Click "Submit". A confirm dialog will show up on the top right corner.

3. Click "Proceed" to the interaction table

- 8 organisms
- 3 ID types : Entrez ID, Ensembl ID and gene symbol.
- 53 tissues for human miRNA

3. Upload a list of lncRNA

🏠 ▶ List Upload

Enter a list of lncRNAs below: ?

Organism: H. sapiens (human)

ID type: Official gene symbol

miRNA source: Cervix

MEG8
XIST
HCG18
TUG1
NEAT1
H19
HCG18
MALAT1
EMX2OS
SNHG7
SNHG1
KCNQ1OT1

Submit

Proceed

Try Examples

Only the data for human are available

ID type : gene symbol, Entrez ID and Ensembl ID

1. Copy and paste a list of lncRNA

2. Click "Submit"

3. Click "Proceed" to the interaction table

You can try our example first

Upload selected items

miRNet -- network-based visual analysis of miRNAs, targets and functions

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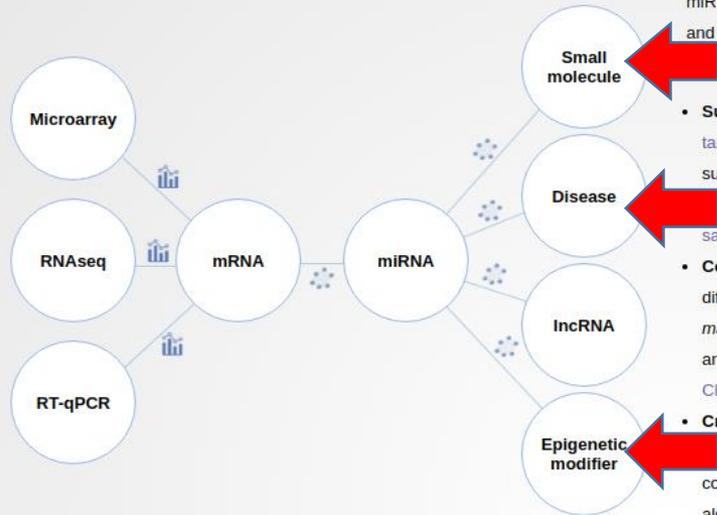
News & Updates

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Click on an input area below to start analysis



Selecting one of the data types.

Overview

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- **Support for various inputs & statistics:** miRNet accepts a list of miRNAs or targets, or a data table from microarray, RNAseq or RT-qPCR experiments. miRNet supports differential analysis using *limma*, *edgeR* and HTqPCR methods; enrichment analysis using standard hypergeometric tests and unbiased random sampling.
- **Comprehensive functional annotation:** miRNet integrates data from eleven different miRNA databases - [TarBase](#), [miRTarBase](#), [miRecords](#), [miRanda](#) (*S. mansoni* only), [miR2Disease](#), [HMDD](#), [PhenomiR](#), [SM2miR](#), [PharmacomiR](#), [EpiMiR](#), and [starBase](#). It currently supports **nine** organisms - Human, Mouse, Rat, Cattle, Chicken, Zebra fish, Fruit fly, *C. elegans* and *S. mansoni*.
- **Creation of miRNA-target interaction networks:** miRNet provides a wide array of options to allow researchers to build miRNA-target interaction networks at different confidence levels. The resulting network can be further optimized using different algorithms to improve visualization and understanding.
- **High-performance network visual analytics:** miRNet offers five types of networks on miRNA-gene, miRNA-disease, miRNA-small molecule, miRNA-IncRNA, and miRNA-epigenetic modifier. The system supports zooming, highlighting, point-and-click, drag-and-drop, enrichment analysis, etc. to enable users to intuitively explore miRNAs, targets and functions.

Upload selected items

1. Choose a tissue type and choose the items by dragging or selecting or searching

The data for human are available on diseases and epigenetic modifiers. The data for human and mouse are available on small molecules

We provide a list of disease, molecule and epigenetic modifier for users to choose, because there is no standard IDs available and use of free text is error-prone.

Select diseases from our database:

H. sapiens (human)

Load Data

Tissue:

Bone marrow (598)

Available		Selected
Creutzfeldt-Jakob Syndrome		Acute promyelocytic leukemia (APL)
Dermatitis, atopic		Bladder cancer
Diabetic Nephropathies		Cerebellar neurodegeneration
Diarrhea predominant irritable bowel syndrome (IBS-D)		Dermatomyositis (DM)
Diffuse large B-cell lymphoma (DLBCL)		Diabetes type 2
Down syndrome (DS)		
Duchenne muscular dystrophy (DMD)		
Endometrial cancer		
Endometriosis		
Epithelial ovarian cancer (EOC)		
Esophageal cancer		
Esophageal squamous cell carcinoma		
Essential thrombocythemia (ET)		
Facioscapulohumeral muscular dystrophy (FSHD)		
Follicular lymphoma (FL)		
Follicular thyroid carcinoma (FTC)		
frontotemporal dementia		

Submit

2. Click "Submit"

3. Click "Proceed"

Proceed

Upload expression data sets

The screenshot displays the miRNet website interface. At the top, the title "miRNet -- network-based visual analysis of miRNAs, targets and functions" is shown. Below the title is a navigation bar with links for Home, FAQs, Tutorials, Resources, and About. The main content area is divided into three sections: News & Updates, a central diagram, and Overview.

News & Updates

- Added tissue-specific miRNA annotation for human miRNAs and updated miRNA-gene interaction from miRTarbase and TarBase (12/26/2017); **NEW**.
- Code refactoring for performance improvement (10/25/2017); **NEW**.
- Fixed the issue for broken miRBase links (09/25/2017); **NEW**.
- Added support for reducing node overlap for large network layout (09/22/2017); **NEW**.
- Added Force Atlas layout algorithm to support large network visualization (09/21/2017); **NEW**.
- Fixed broken links for downloading the mir target data files (08/25/2017);
- Enhanced data IO for improved

Please Cite

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Click on an input area below to start analysis

```
graph LR; Microarray --> mRNA; RNAseq --> mRNA; RT-qPCR --> mRNA; mRNA --> miRNA; miRNA --> Small_molecule[Small molecule]; miRNA --> Disease; miRNA --> lncRNA; miRNA --> Epigenetic_modifier[Epigenetic modifier];
```

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Selecting one of the data types.

Xia Lab @ McGill (last updated 2017-Dec-26)

Data Formatting

- Manipulate data headings in a spreadsheet program like MS Excel
- Save as a **tab delimited** .txt file
- The headings **#NAME** and **#CLASS:** (**all capital letters**) must be used

#NAME is for sample names (first row in your data)

#CLASS is for the clinical metadata.

The screenshot below shows the labels for the experimental condition.

#NAME	Sample1	Sample2	Sample3	Sample4	Sample5	Sample6	Sample7	Sample8	Sample9
#CLASS	Y	N	N	Y	N	Y	Y	N	N
100_g_at	-3.06	-2.25	-1.15	-6.64	0.4	1.08	1.22	1.02	1.15
1000_at	-1.36	-0.67	-0.17	-0.97	-2.32	-5.06	0.28	1.32	0.73
1002_f_at	1.61	-0.27	0.71	-0.62	0.14		0.11	0.98	0.54
1008_f_at	0.93	1.29	-0.23	-0.74	-2	-1.25	1.07	1.27	1.02

Data Upload

A green check will appear to indicate each step is successfully completed

1. Upload a well formatted file (you can try the example first).

2. Click "Submit" to confirm each step. A confirm dialog will show up on the top right corner.

① Data Upload ✓	Upload file	<input type="button" value="Browse..."/> No file selected.	<input type="button" value="Submit"/>
② Annotation	Specify organism	<input type="text" value="H. sapiens (human)"/>	<input type="button" value="Submit"/>
	ID type	<input type="text" value="Affymetrix Human Genome U95 (chip hgu95av2)"/>	

Hover over the question marks for more information about each step.

Data Annotation

Annotation ✓	Specify organism	H. sapiens (human)	Submit
	ID type	Affymetrix Human Genome U95 (chip hgu95av2)	
	Tissue (human only)	Bone marrow (598)	

- Specifying the organism type and ID type allows miRNet to annotate your data;
- Eight organisms are supported;
- Entrez ID, Ensembl ID, gene symbol, miRNA ID and miRNA accession are supported for RNA-seq and RT-qPCR analysis;
- Affymetrix, Illumina and Agilent Microarray probe ID are supported for Microarray analysis.
- Supporting 53 tissues for human miRNA annotation.

Data Normalization

Microarray Profiling

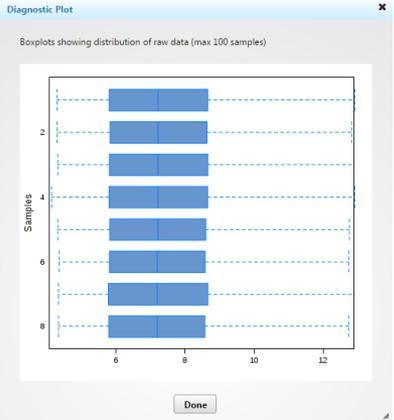
② Annotation ✓

ID type: Affymetrix Human Genome U95 (chip hgu95av2)

③ Normalization ✓

Normalization procedure: -- No normalization --

[View Data](#) [Submit](#)



Diagnostic Plot

Boxplots showing distribution of raw data (max 100 samples)

Samples: 2, 4, 6, 8

Done

The diagnostic plot shows eight horizontal boxplots, one for each sample. The y-axis is labeled 'Samples' and has tick marks at 2, 4, 6, and 8. The x-axis has tick marks at 0, 8, 16, and 22. Each boxplot shows the distribution of raw data for that sample, with a median line, a box representing the interquartile range, and whiskers extending to the minimum and maximum values. The distributions appear relatively consistent across samples, with medians around 8-10 and boxes spanning from approximately 4 to 16.

- This example dataset has been normalized and needed no normalization
- If raw data is uploaded, normalization can be applied
 - Different normalization methods are provided for different data types

Data Analysis

The screenshot shows a web interface for data analysis, divided into three main sections: "Comparisons of Interest", "Feature Selection", and "Specify Network".

- Comparisons of Interest:** Includes a "Specify comparison" section with dropdowns for "absent" and "present" separated by "versus". A callout box explains: "If there is an error with matching your data to our library, please download your result and manually modify your data label. More details in FAQ #5".
- Feature Selection:** Includes input fields for "Adjusted p-value" (0.01) and "Log2 fold change" (1.0). A "Directions" dropdown is set to "Both directions", with a callout box listing options: "Both direction", "Upregulate only", and "Downregulate only". A "Download Result" button is also present.
- Specify Network:** Includes a "Choose target" dropdown set to "microRNA".

At the bottom of the interface, there is a "Try Examples" button and a "Proceed" button. A callout box explains: "If upload data is gene, only miRNA network can be presented. If the data is miRNA, you can choose gene, disease, molecule, lncRNA and epigenetic modifier network". Another callout box points to the "Proceed" button, stating: "Click 'Proceed' to the interaction table".

==END==