Instructions for miRTARGET

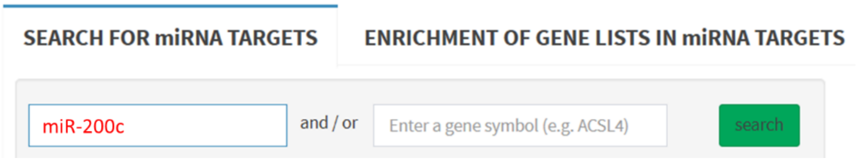
www.mirtarget.com

miRTARGET can perform the following analyzes:

1. Search for target genes of a specific miRNA
2. Search for miRNA regulators of a specific gene
3. Determine whether a specific gene is a target of a specific miRNA
4. Enrichment of gene lists in miRNA targets

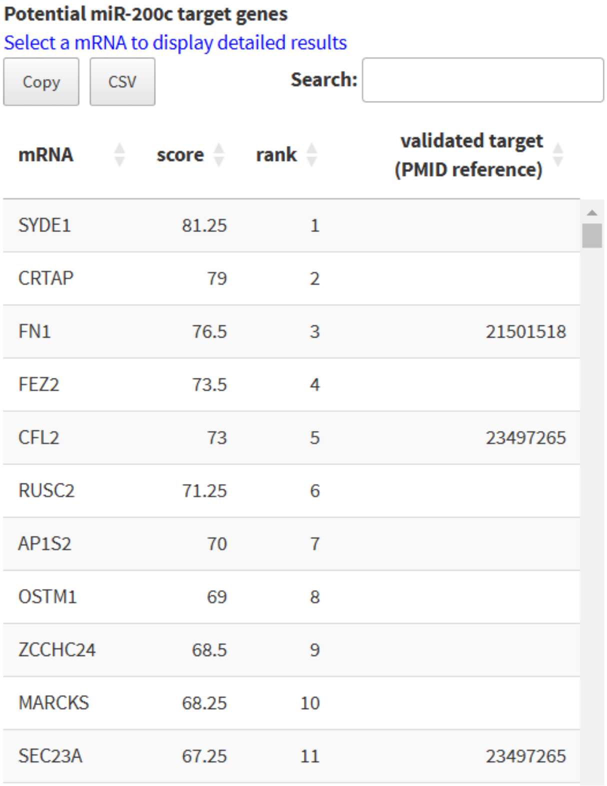
1. Search for target genes of a specific miRNA

Enter a miRNA (e.g. miR-200c) and click search.



miR-200c

The table on the right shows all genes, sorted according to the miRNA target score/rank. Genes with the highest scores represent the top ranked potential miRNA targets. The miRNA target prediction score/rank is based on four criteria: Change in mRNA expression by ectopic miRNA, Change in mRNA expression in miRNA KO/KD mice/cells, Computational miRNA target prediction, and Correlation between miRNA and mRNA expression. The right column indicates validated miRNA targets according to the miRTarBase 9.0.



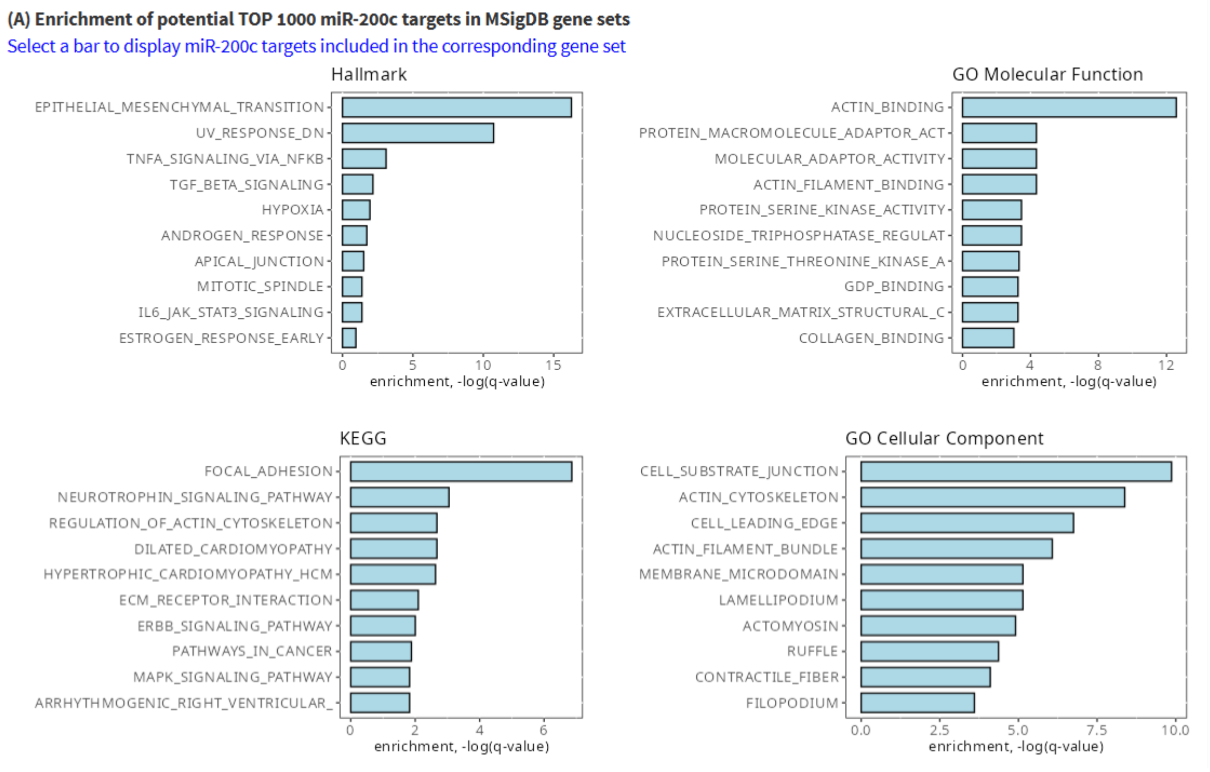
Potential miR-200c target genes

Select a mRNA to display detailed results

mRNA	score	rank	validated target (PMID reference)
SYDE1	81.25	1	
CRTAP	79	2	
FN1	76.5	3	21501518
FEZ2	73.5	4	
CFL2	73	5	23497265
RUSC2	71.25	6	
AP1S2	70	7	
OSTM1	69	8	
ZCCHC24	68.5	9	
MARCKS	68.25	10	
SEC23A	67.25	11	23497265

Enrichment of predicted miRNA targets and their associations with cancer

Plots in (A) show the enrichment of the 1000 highest ranked predicted miRNA target genes in indicated MSigDB gene sets (Hallmark, KEGG, GO Molecular Function, and GO Cellular Component). These graphs provide information about the molecular functions of the potential miRNA targets of and processes / pathways in which they are involved.


(A) Enrichment of potential TOP 1000 miR-200c targets in MSigDB gene sets

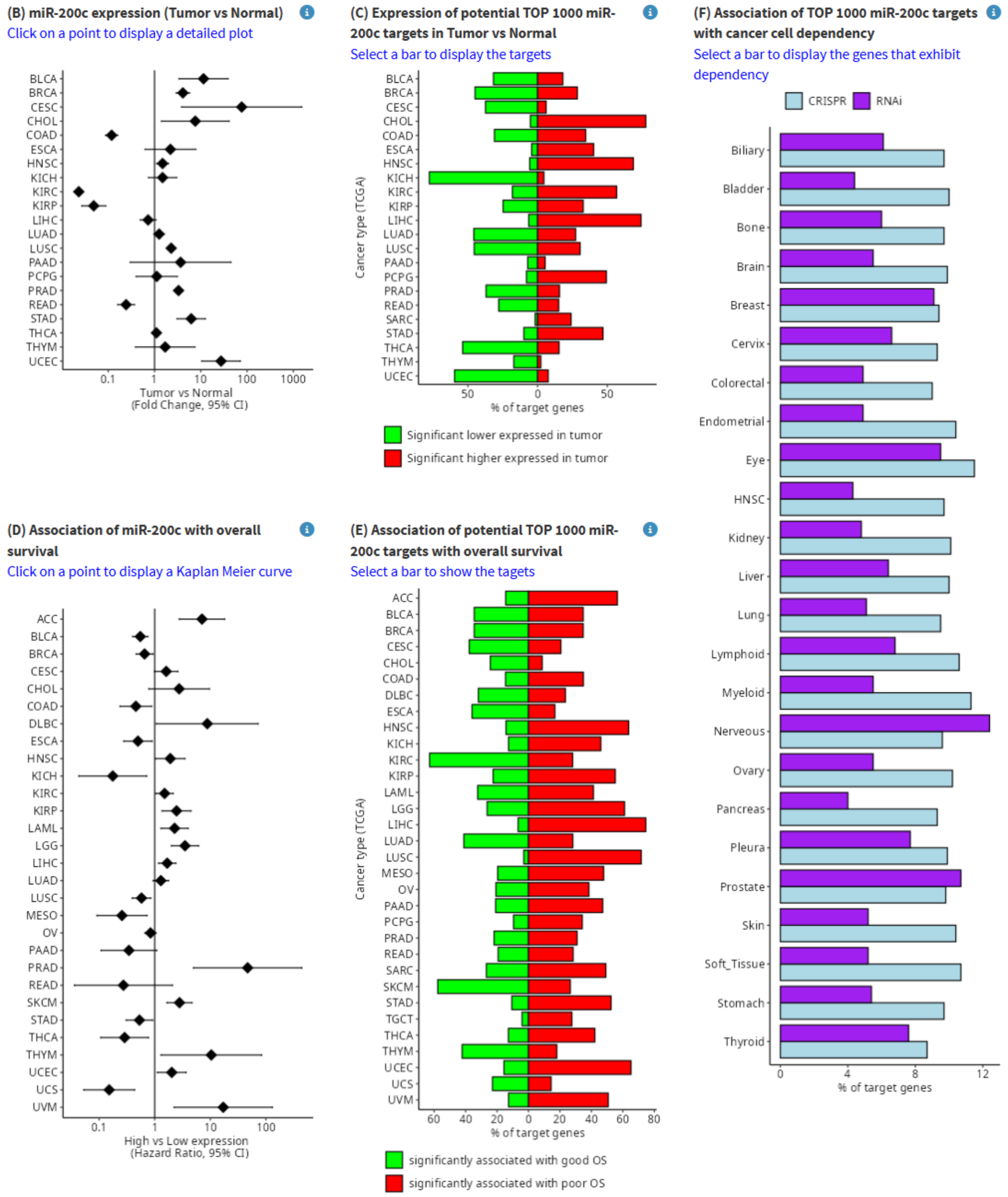
Select a bar to display miR-200c targets included in the corresponding gene set

Plots B and D show the expression of the specified miRNA in tumor vs normal tissue (B) and the association with overall survival (D) in indicated cancer types. These plots are based on the data is from the TCGA database.

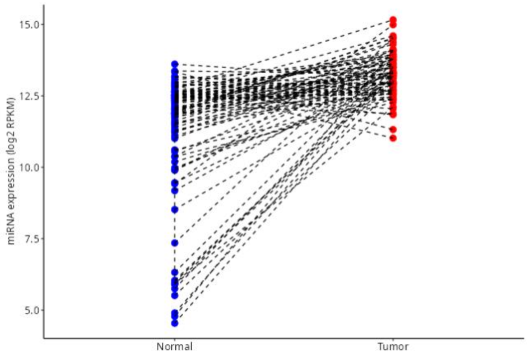
Plots C and E show the association of predicted miRNA target genes with tumor vs normal tissue (C) and overall survival (E) in indicated cancer types. The plot (C) shows the percentage of the top 1000 highest ranked predicted miRNA target genes that are expressed significantly higher (red) or lower (green) in tumor vs normal tissue. The plot (E) shows the percentage of the top 1000 highest ranked predicted miRNA target genes that are significantly associated with poor (red) or good (green) survival. For example ca. 50% of the 1000 highest ranked predicted miR-200c targets are significantly associated with poor overall survival in Adrenocortical carcinoma (ACC).

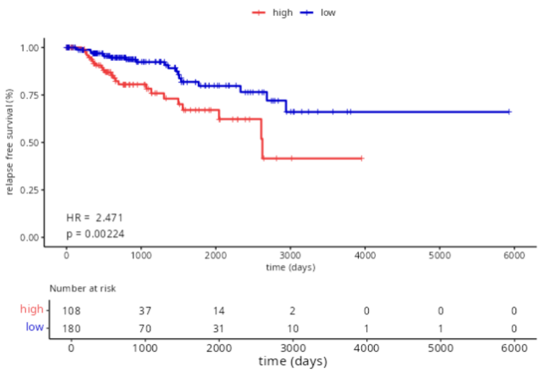
The plot (F) shows the association of predicted miRNA target genes with cancer cell dependency. The percentage of the top 1000 highest ranked predicted miRNA target genes that show a dependency in cancer cell lines from indicated cancer types are shown. A gene is considered dependent if at least 10% of cancer cell lines from a specific cancer entity display a dependency score of less than -0.5. Data is from the Dependency Map Portal (www.depmap.org). The CRISPR and RNAi indicates whether the data is based on CRISPR (Chronos) or shRNA (DEMETER2) screening.

These plots provide information whether the predicted targets of a specific miRNA have predominantly oncogenic or tumor suppressive properties in indicated cancer entities.



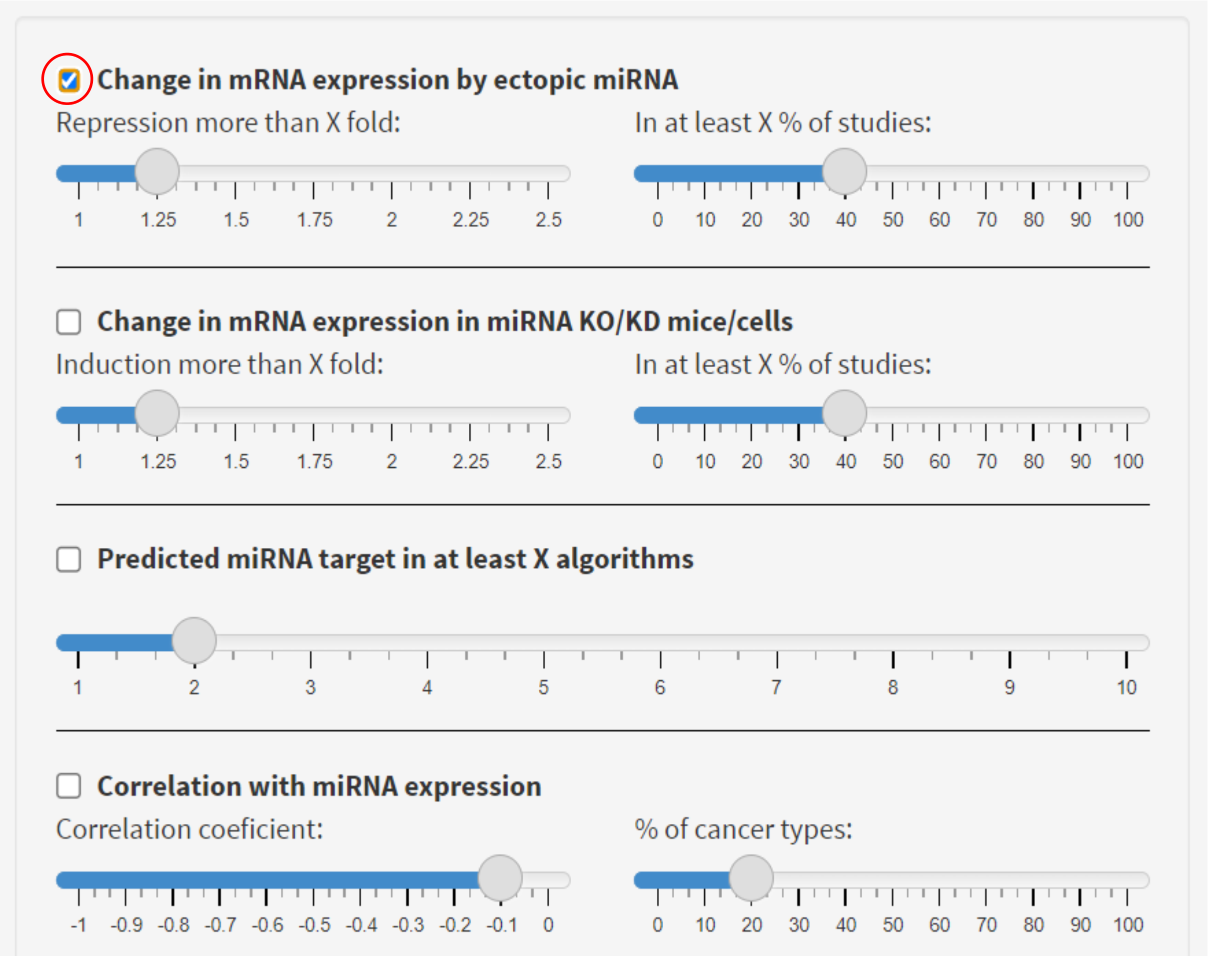
A click on plots B and D shows the underlying tumor/normal plot or the Kaplan Meier plot for the clicked cancer type. A click on plots C, E, and F shows the mRNAs that fulfill the indicated criteria.





Adjustment of selection criteria for miRNA target prediction

The miRNA target prediction score/rank is based on four criteria: Change in mRNA expression by ectopic miRNA, Change in mRNA expression in miRNA KO/KD mice/cells, Computational miRNA target prediction, and Correlation between miRNA and mRNA expression. Each criterion can be modified by selecting the corresponding checkbox and adjusting the parameters with sliders.



☑ **Change in mRNA expression by ectopic miRNA**

Repression more than X fold:

In at least X % of studies:

☐ **Change in mRNA expression in miRNA KO/KD mice/cells**

Induction more than X fold:

In at least X % of studies:

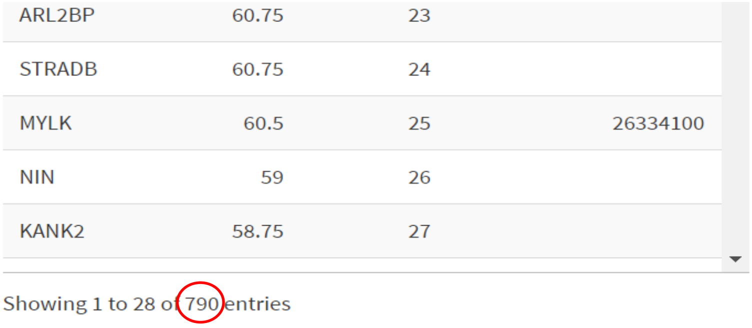
☐ **Predicted miRNA target in at least X algorithms**

☐ **Correlation with miRNA expression**

Correlation coeficient:

% of cancer types:

The modification of criteria will adjust the list/table of predicted miRNA target genes to show only targets that fulfil these criteria.



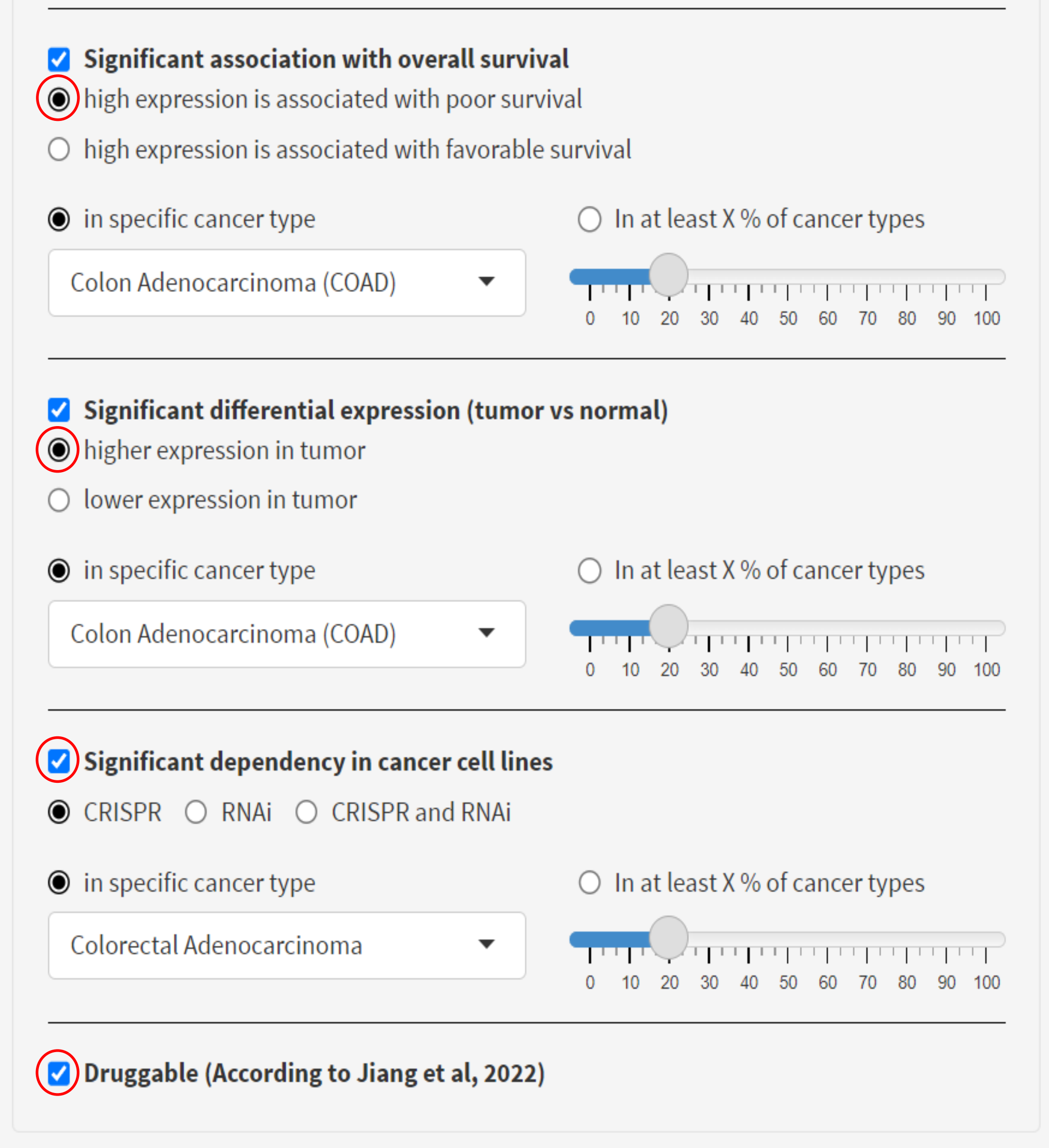
ARL2BP	60.75	23	
STRADB	60.75	24	
MYLK	60.5	25	26334100
NIN	59	26	
KANK2	58.75	27	

Showing 1 to 28 of 790 entries

Identification of predicted miRNA target genes that are associated with cancer

For users interested in cancer research, we added additional criteria, which allow further selection of predicted miRNA target genes according to their association with cancer.

For example to identify predicted miRNA target genes that display oncogenic properties users can select the checkboxes “Significant association with overall survival / high expression is associated with poor survival”, “Significant differential expression (tumor vs normal) / higher expression in tumor”, “Significant dependency I cancer cell lines”, and/or “Druggable”. Then select either “in specific cancer type” or “in at least X % of cancer types”.



☑ **Significant association with overall survival**

● high expression is associated with poor survival

○ high expression is associated with favorable survival

● in specific cancer type ○ In at least X % of cancer types

Colon Adenocarcinoma (COAD)

☑ **Significant differential expression (tumor vs normal)**

● higher expression in tumor

○ lower expression in tumor

● in specific cancer type ○ In at least X % of cancer types

Colon Adenocarcinoma (COAD)

☑ **Significant dependency in cancer cell lines**

● CRISPR ○ RNAi ○ CRISPR and RNAi

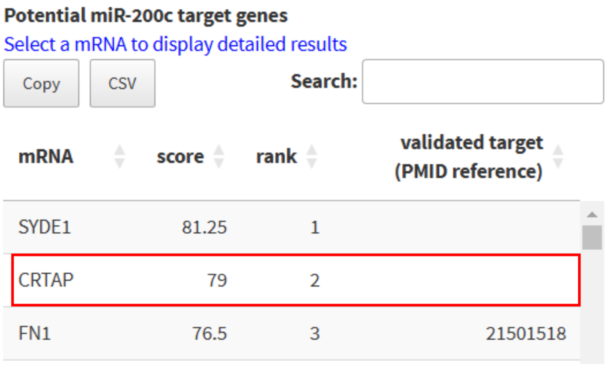
● in specific cancer type ○ In at least X % of cancer types

Colorectal Adenocarcinoma

☑ **Druggable (According to Jiang et al, 2022)**

Information about a specific predicted miRNA target gene

Users can click on a gene in the table with predicted miRNA targets to display the data that was used to calculate the miRNA target prediction score / rank.



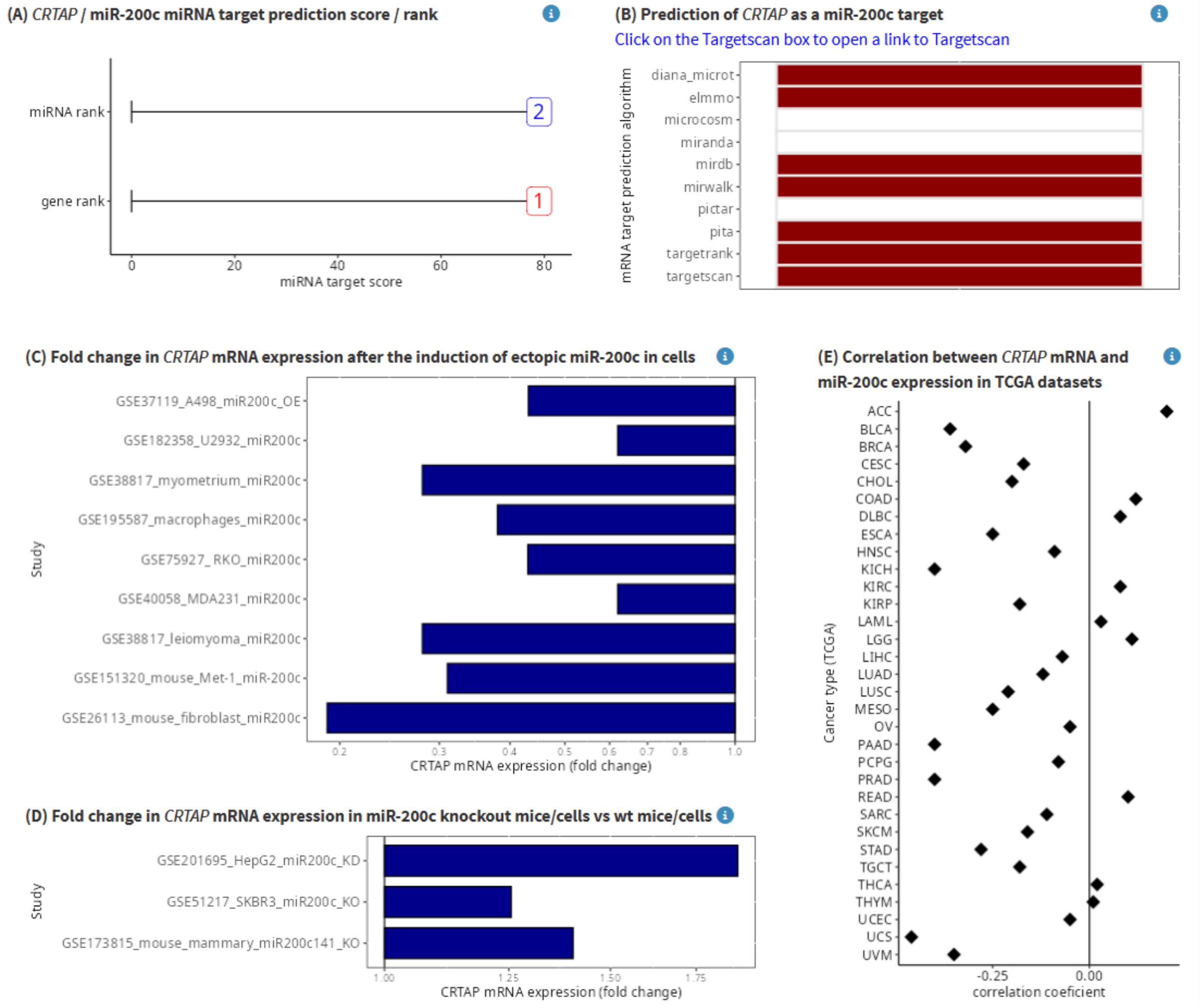
Potential miR-200c target genes

Select a mRNA to display detailed results

mRNA	score	rank	validated target (PMID reference)
SYDE1	81.25	1	
CRTAP	79	2	
FN1	76.5	3	21501518

The miRNA target prediction score and rank are displayed in plot (A). The miRNA rank represents the rank of the selected gene as a potential target of the specified miRNA among all genes (1 to 24397). The gene rank represents the rank of the specified miRNA as a potential regulator of the selected gene among all miRNAs (1 to 1745).

The miRNA target prediction score/rank is based on four criteria, which are showed in plots B - E: (B) Computational miRNA target prediction by indicated algorithms, (C) Fold change in target mRNA expression by ectopic miRNA, (D) Fold change in mRNA expression in miRNA KO/KD mice/cells, and (E) Correlation between miRNA and mRNA expression in tumor tissue from indicated cancer entities.



(A) *CRTAP* / miR-200c miRNA target prediction score / rank

(B) Prediction of *CRTAP* as a miR-200c target

Click on the Targetscan box to open a link to Targetscan

(C) Fold change in *CRTAP* mRNA expression after the induction of ectopic miR-200c in cells

(D) Fold change in *CRTAP* mRNA expression in miR-200c knockout mice/cells vs wt mice/cells

(E) Correlation between *CRTAP* mRNA and miR-200c expression in TCGA datasets

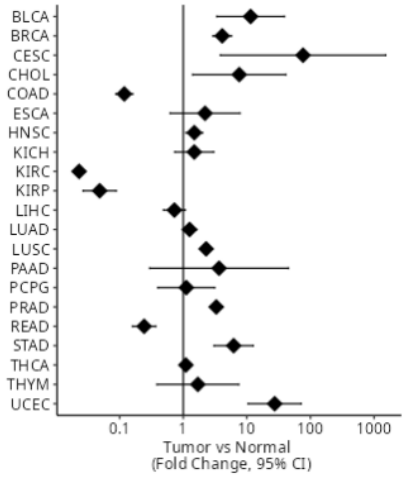
Associations of a specific miRNA and its predicted target mRNA with cancer

The plots (F) and (G) show the associations of the selected miRNA (F) and mRNA (G) with overall survival in indicated cancer types. The plots (H) and (I) show the difference in expression of the selected miRNA (H) and mRNA (I) between tumor and normal tissue in indicated cancer types. These plots are based on the data from the TCGA database.

Plot (J) shows the dependency of cancer cell lines from indicated cancer entities on the selected gene. A lower gene effect/dependency score means that a cell line is more dependent on the selected gene. A score of less than -0.5 indicates a strong dependency and a score of less than -1 suggests essentiality. A score of 0 is equivalent to a gene that is not essential, whereas a score of -1 corresponds to the median of all common essential genes. This plot is based on the data is from the Dependency Map Portal (www.depmap.org). The CRISPR and RNAi indicates whether the data is based on CRISPR (Chronos) or shRNA (DEMETER2) screening.

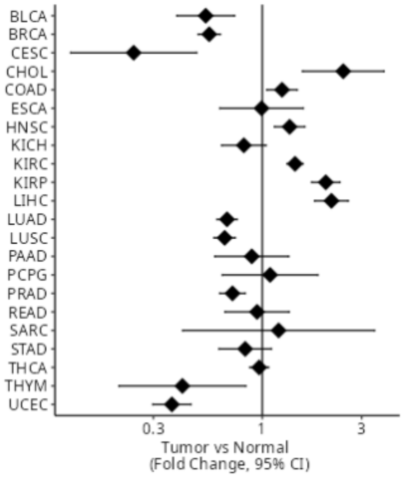
(F) miR-200c expression (Tumor vs Normal)

Click on a point to display a detailed plot

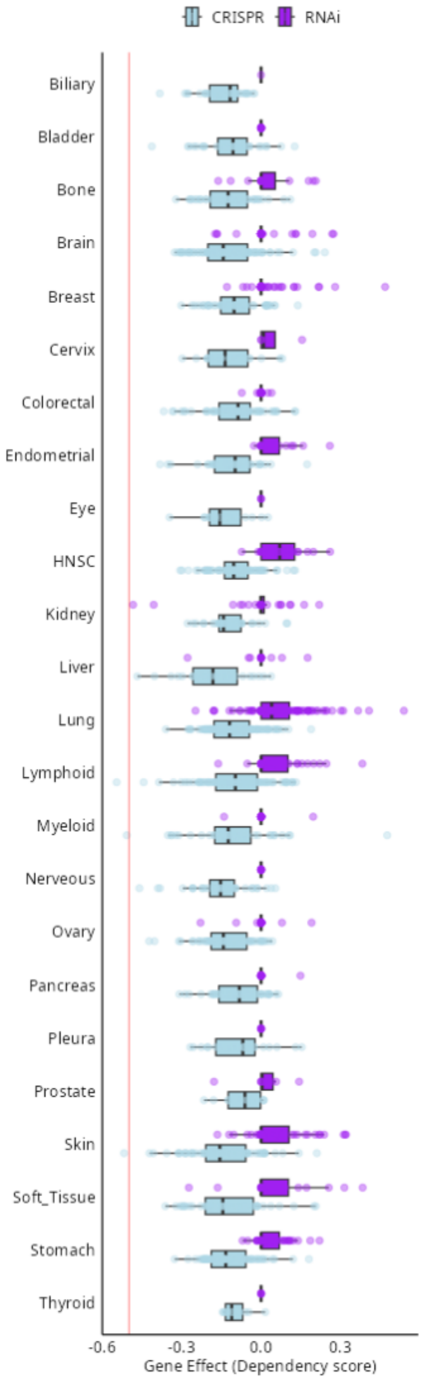


(I) *CRTAP* expression (Tumor vs Normal)

Click on a point to display a detailed plot

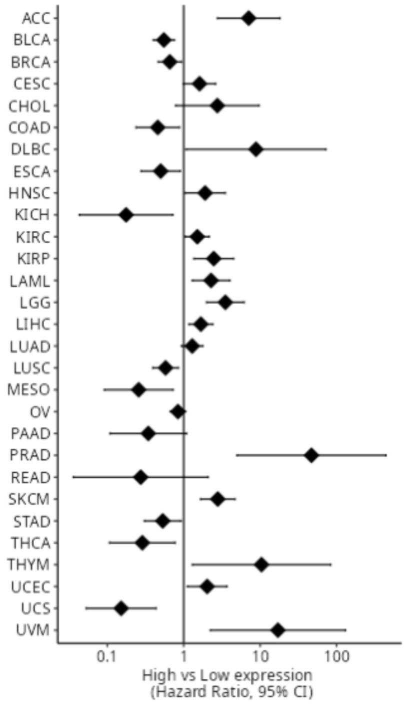


(J) Dependency of cancer cell lines from indicated cancer entities on *CRTAP*



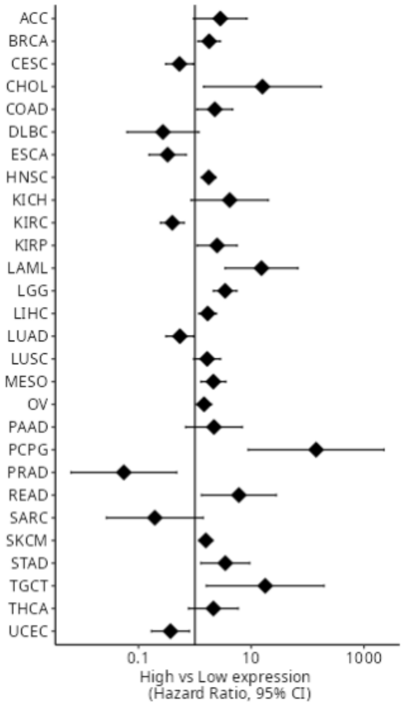
(H) Association of miR-200c with overall survival

Click on a point to display a Kaplan Meier curve



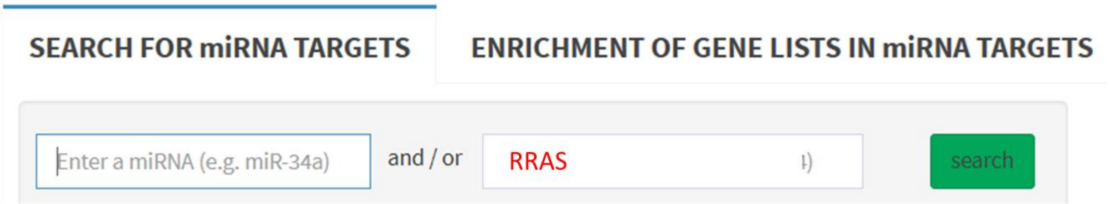
(G) Association of *CRTAP* with overall survival

Click on a point to display a Kaplan Meier curve



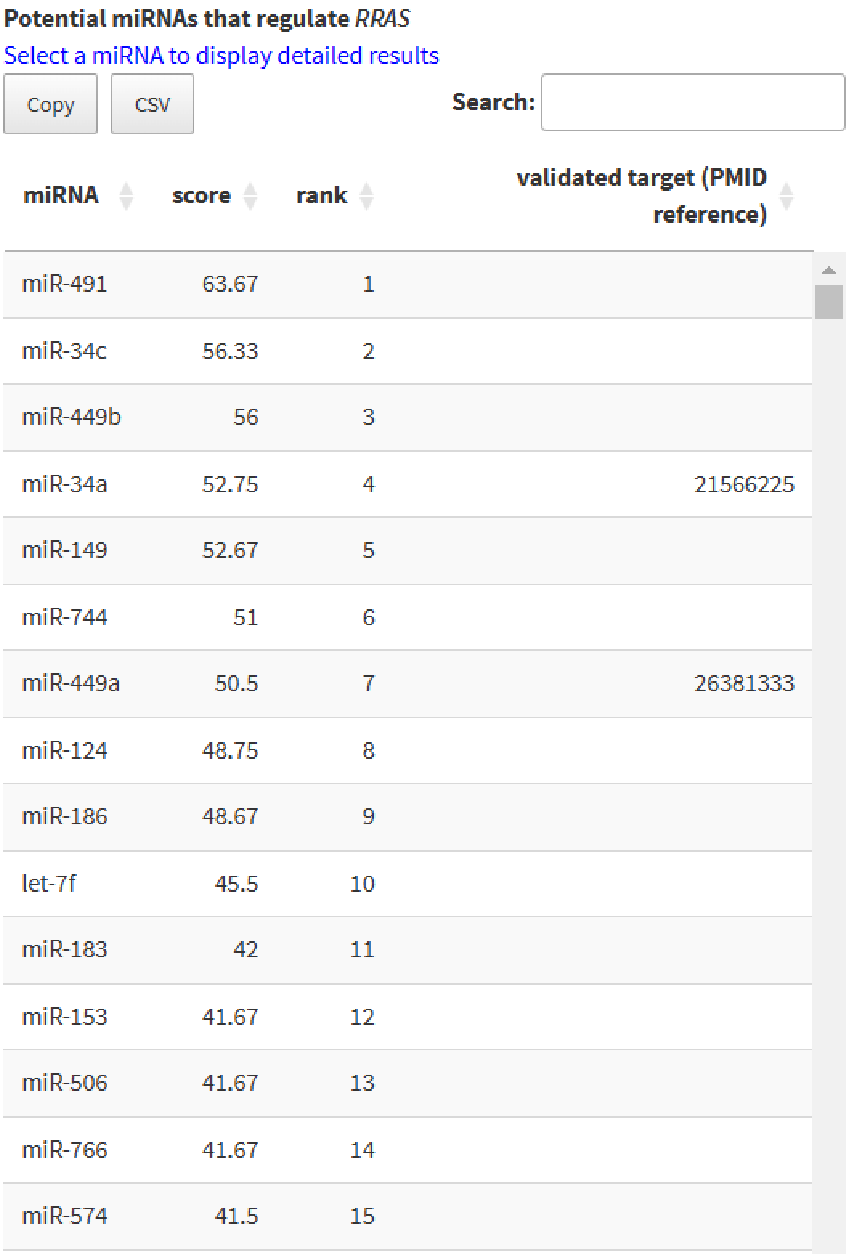
2. Search for miRNA regulators of a specific gene

Enter a gene symbol (e.g. RRAS) and click search.



RRAS

The table on the right shows all miRNAs, sorted according to the miRNA target score/rank. miRNAs with the highest scores represent the top ranked potential regulators of the specified genes. The miRNA target prediction score/rank is based on four criteria: Change in mRNA expression by ectopic miRNA, Change in mRNA expression in miRNA KO/KD mice/cells, Computational miRNA target prediction, and Correlation between miRNA and mRNA expression. The right column indicates whether the regulation of the specified gene by the indicated miRNA has already been validated.



Potential miRNAs that regulate *RRAS*

Select a miRNA to display detailed results

miRNA	score	rank	validated target (PMID reference)
miR-491	63.67	1	
miR-34c	56.33	2	
miR-449b	56	3	
miR-34a	52.75	4	21566225
miR-149	52.67	5	
miR-744	51	6	
miR-449a	50.5	7	26381333
miR-124	48.75	8	
miR-186	48.67	9	
let-7f	45.5	10	
miR-183	42	11	
miR-153	41.67	12	
miR-506	41.67	13	
miR-766	41.67	14	
miR-574	41.5	15	

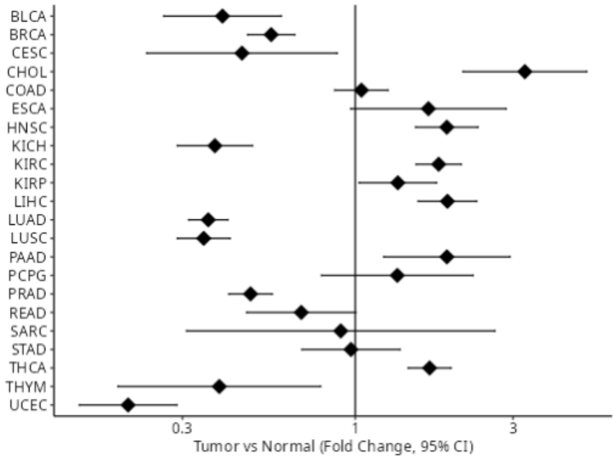
Association of predicted miRNA regulators with cancer

Plots A and C show the expression of the specified gene in tumor vs normal tissue (A) and the association with overall survival (C) in indicated cancer types. These plots are based on the data is from the TCGA database. Plots B and D show the association of miRNAs that potentially regulate the specified gene with tumor vs normal tissue (B) and overall survival (D) in indicated cancer types. The plot (B) shows the percentage of the top 100 highest ranked predicted miRNAs that are expressed significantly higher (red) or lower (green) in tumor vs normal tissue. The plot (D) shows the percentage of the top 100 highest ranked predicted miRNAs that are significantly associated with poor (red) or good (green) survival. For example ca. 75% of the 100 highest ranked miRNAs that potentially regulate RRAS are significantly higher expressed in tumors vs normal tissue in Bladder carcinoma (BLCA).

These plots provide information whether the miRNAs that potentially regulate the specified gene have predominantly oncogenic or tumor suppressive properties in indicated cancer entities.

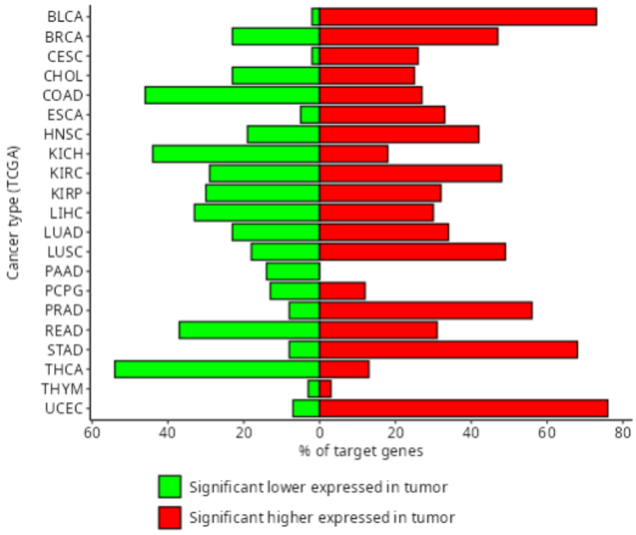
(A) *RRAS* expression (Tumor vs Normal)

Click on a point to display a detailed plot



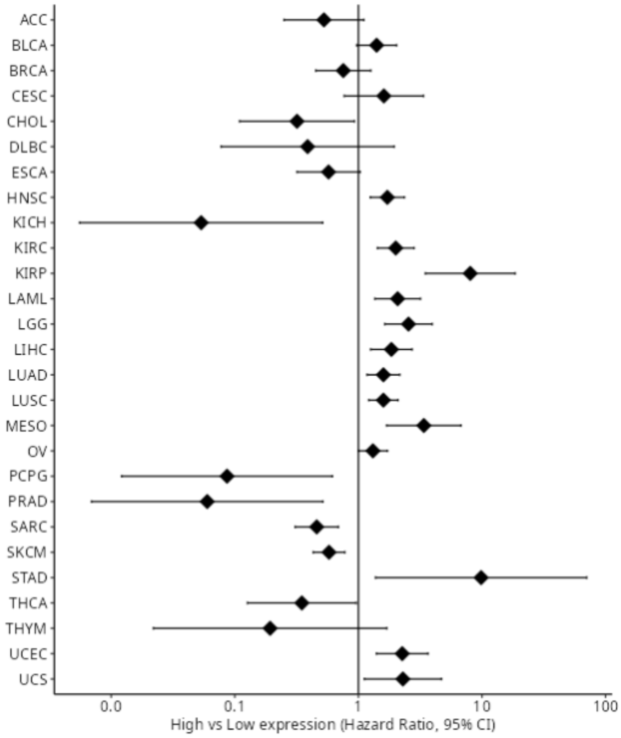
(B) Expression of potential TOP 100 *RRAS* regulating miRNAs in Tumor vs Normal

Select a bar to show the tagets



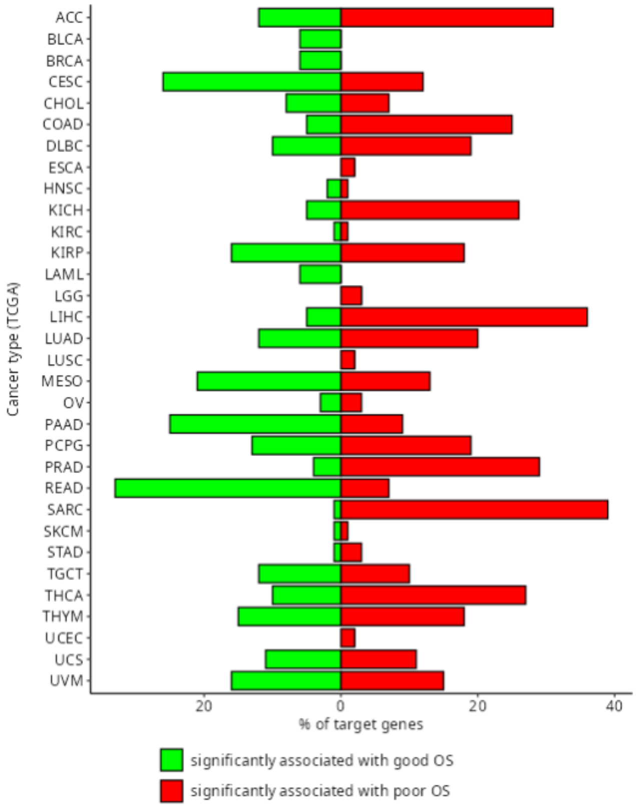
(C) Association of *RRAS* with overall survival

Click on a point to display a Kaplan Meier curve



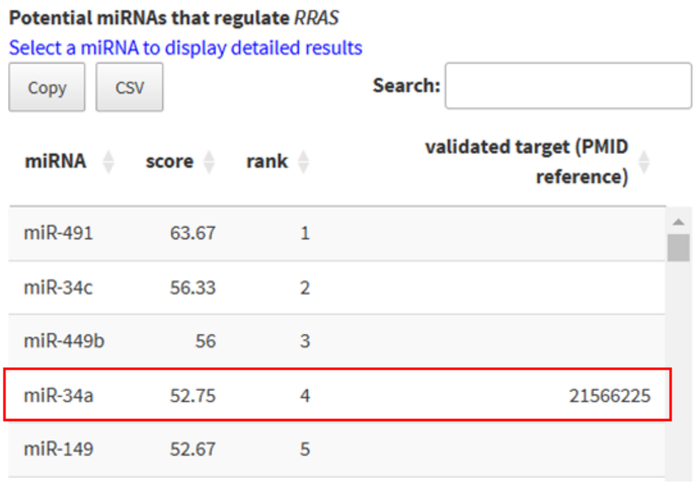
(D) Association of potential TOP 100 *RRAS* regulating miRNAs with overall survival

Select a bar to show the miRNAs



Information about a specific miRNA and the specified target gene

Users can click on a miRNA in the table with predicted miRNA regulator to display the data that was used to calculate the miRNA target prediction score / rank.



Potential miRNAs that regulate *RRAS*

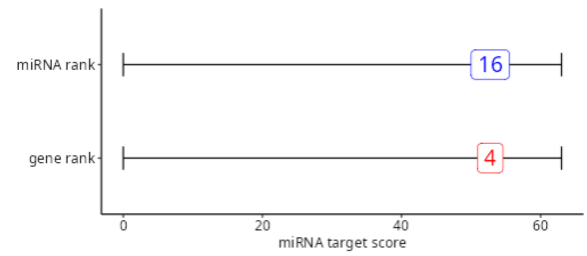
Select a miRNA to display detailed results

miRNA	score	rank	validated target (PMID reference)
miR-491	63.67	1	
miR-34c	56.33	2	
miR-449b	56	3	
miR-34a	52.75	4	21566225
miR-149	52.67	5	

The miRNA target prediction score and rank are displayed in plot (A). The miRNA rank represents the rank of the selected gene as a potential target of the specified miRNA among all genes (1 to 24397). The gene rank represents the rank of the specified miRNA as a potential regulator of the selected gene among all miRNAs (1 to 1745).

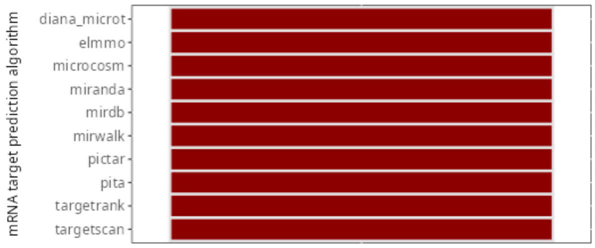
The miRNA target prediction score/rank is based on four criteria, which are showed in plots B - E: (B) Computational miRNA target prediction by indicated algorithms, (C) Fold change in target mRNA expression by ectopic miRNA, (D) Fold change in mRNA expression in miRNA KO/KD mice/cells, and (E) Correlation between miRNA and mRNA expression in tumor tissue from indicated cancer entities.

(A) *RRAS* / miR-34a miRNA target prediction score / rank

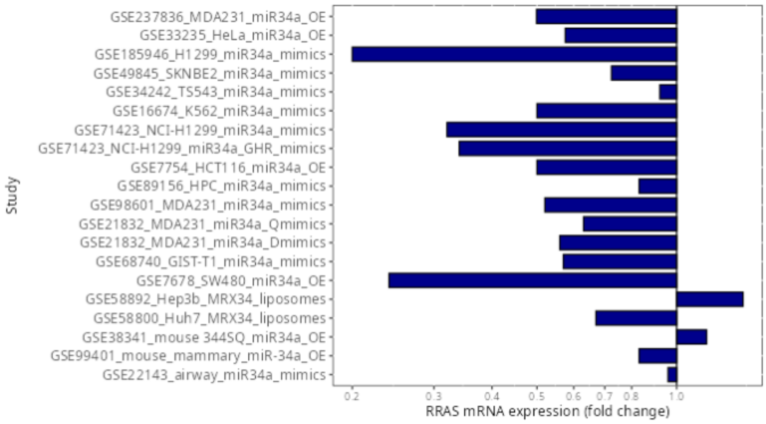


(B) Prediction of *RRAS* as a miR-34a target

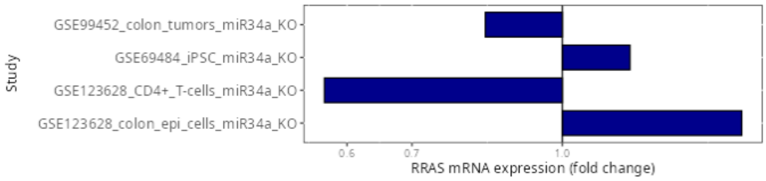
Click on the Targetscan box to open a link to Targetscan



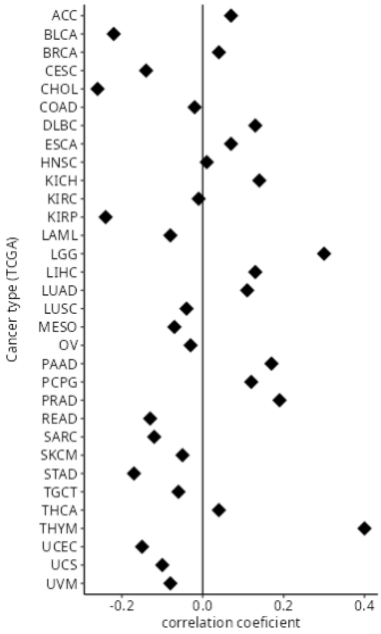
(C) Fold change in *RRAS* mRNA expression after the induction of ectopic miR-34a in cells



(D) Fold change in *RRAS* mRNA expression in miR-34a knockout mice/cells vs wt mice/cells



(E) Correlation between *RRAS* mRNA and miR-34a expression in TCGA datasets



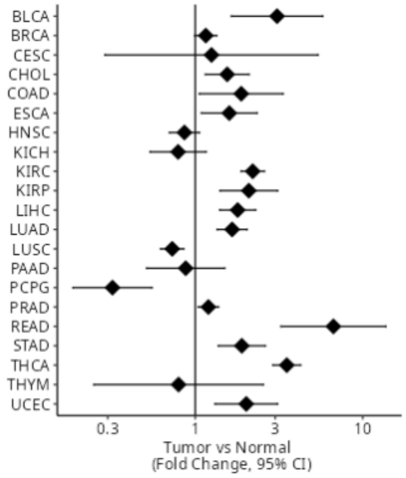
Associations of a specific miRNA and its predicted target mRNA with cancer

The plots (F) and (G) show the associations of the selected miRNA (F) and mRNA (G) with overall survival in indicated cancer types. The plots (H) and (I) show the difference in expression of the selected miRNA (H) and mRNA (I) between tumor and normal tissue in indicated cancer types. These plots are based on the data from the TCGA database.

Plot (J) shows the dependency of cancer cell lines from indicated cancer entities on the selected gene. A lower gene effect/dependency score means that a cell line is more dependent on the selected gene. A score of less than -0.5 indicates a strong dependency and a score of less than -1 suggests essentiality. A score of 0 is equivalent to a gene that is not essential, whereas a score of -1 corresponds to the median of all common essential genes. This plot is based on the data is from the Dependency Map Portal (www.depmap.org). The CRISPR and RNAi indicates whether the data is based on CRISPR (Chronos) or shRNA (DEMETER2) screening.

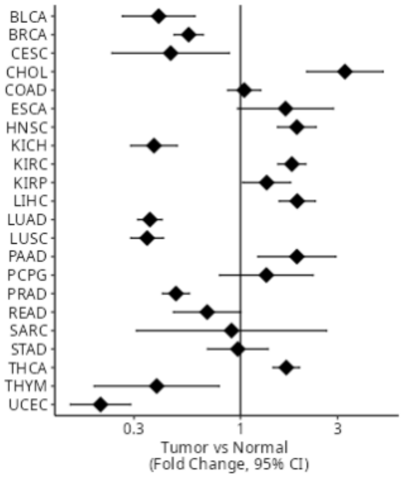
(F) miR-34a expression (Tumor vs Normal)

Click on a point to display a detailed plot

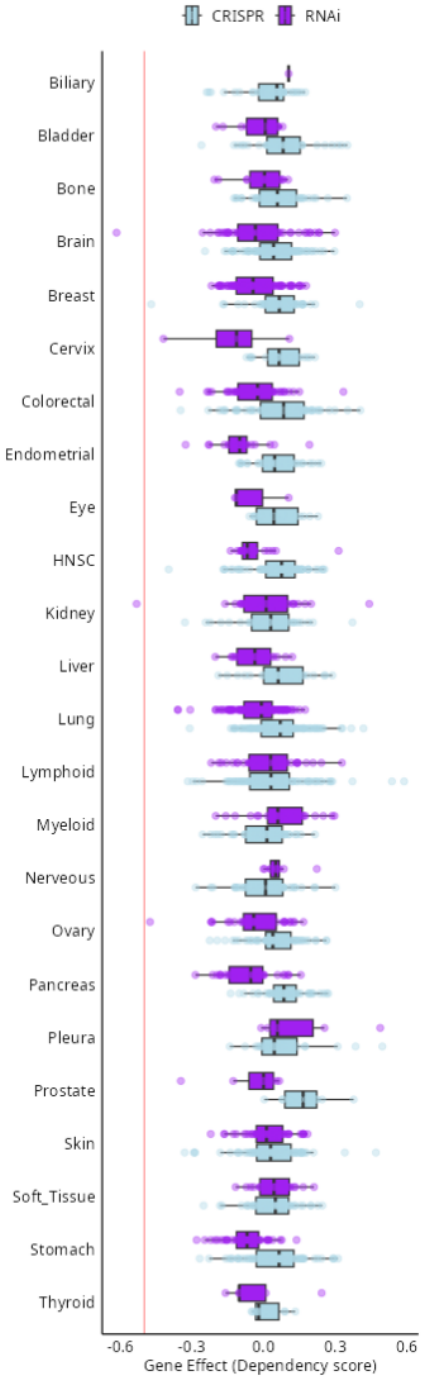


(I) *RRAS* expression (Tumor vs Normal)

Click on a point to display a detailed plot

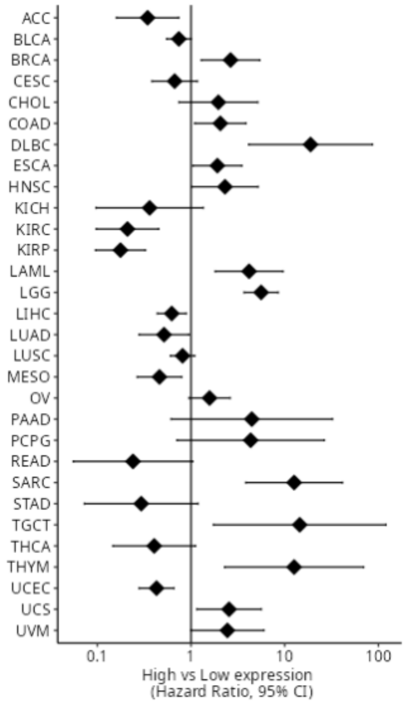


(J) Dependency of cancer cell lines from indicated cancer entities on *RRAS*



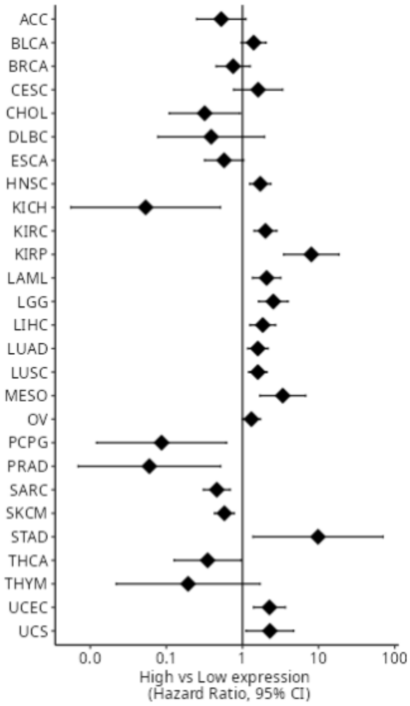
(H) Association of miR-34a with overall survival

Click on a point to display a Kaplan Meier curve



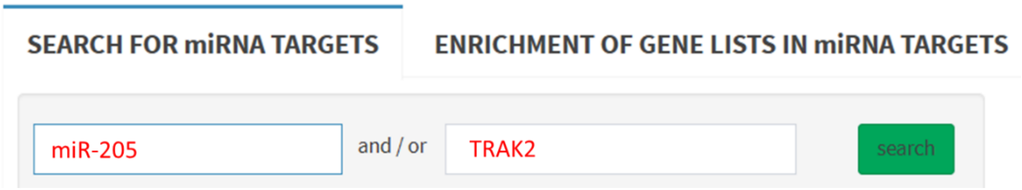
(G) Association of *RRAS* with overall survival

Click on a point to display a Kaplan Meier curve



3. Determine whether a specific gene is a target of a specific miRNA

Enter a miRNA (e.g. miR-133a) and a gene symbol (e.g. TRAK2) and click search.



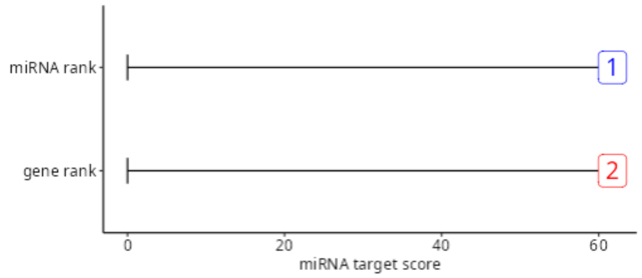
miR-205

TRAK2

The miRNA target prediction score and rank are displayed in plot (A). The miRNA rank represents the rank of the selected gene as a potential target of the specified miRNA among all genes (1 to 24397). The gene rank represents the rank of the specified miRNA as a potential regulator of the selected gene among all miRNAs (1 to 1745).

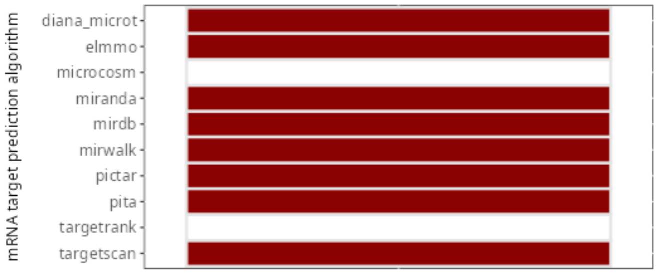
The miRNA target prediction score/rank is based on four criteria, which are showed in plots B - E: (B) Computational miRNA target prediction by indicated algorithms, (C) Fold change in target mRNA expression by ectopic miRNA, (D) Fold change in mRNA expression in miRNA KO/KD mice/cells, and (E) Correlation between miRNA and mRNA expression in tumor tissue from indicated cancer entities.

(A) ***TRAK2*** **/ miR-205 miRNA target prediction score / rank**

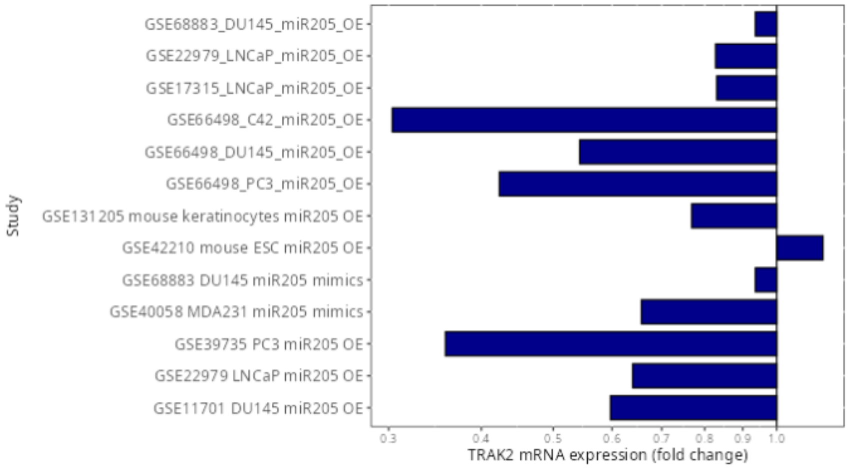


(B) Prediction of ***TRAK2*** **as a miR-205 target**

Click on the Targetscan box to open a link to Targetscan

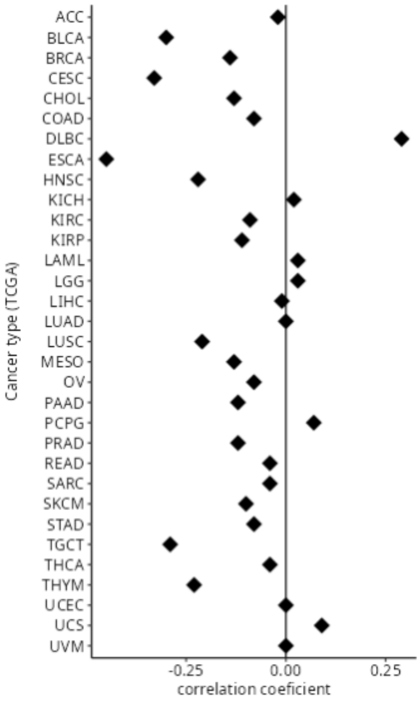


(C) Fold change in ***TRAK2*** **mRNA expression after the induction of ectopic miR-205 in cells**



(D) Fold change in ***TRAK2*** **mRNA expression in miR-205 knockout mice/cells vs wt mice/cells**

(E) Correlation between ***TRAK2*** **mRNA and miR-205 expression in TCGA datasets**



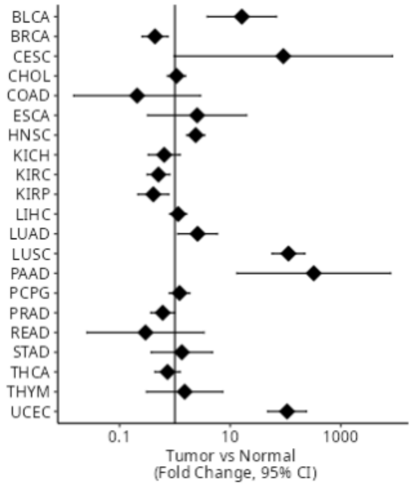
Associations of a specific miRNA and its predicted target mRNA with cancer

The plots (F) and (G) show the associations of the selected miRNA (F) and mRNA (G) with overall survival in indicated cancer types. The plots (H) and (I) show the difference in expression of the selected miRNA (H) and mRNA (I) between tumor and normal tissue in indicated cancer types. These plots are based on the data from the TCGA database.

Plot (J) shows the dependency of cancer cell lines from indicated cancer entities on the selected gene. A lower gene effect/dependency score means that a cell line is more dependent on the selected gene. A score of less than -0.5 indicates a strong dependency and a score of less than -1 suggests essentiality. A score of 0 is equivalent to a gene that is not essential, whereas a score of -1 corresponds to the median of all common essential genes. This plot is based on the data is from the Dependency Map Portal (www.depmap.org). The CRISPR and RNAi indicates whether the data is based on CRISPR (Chronos) or shRNA (DEMETER2) screening.

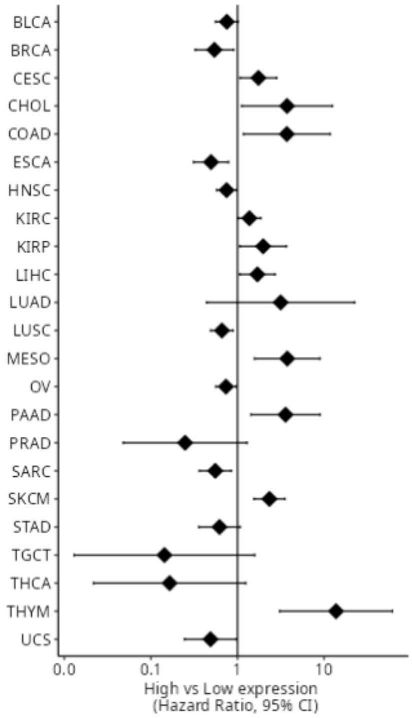
(F) miR-205 expression (Tumor vs Normal)

Click on a point to display a detailed plot



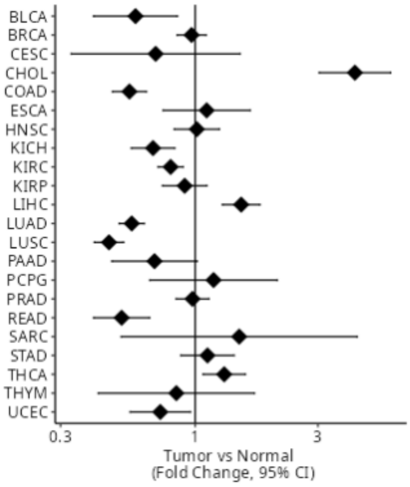
(H) Association of miR-205 with overall survival

Click on a point to display a Kaplan Meier curve



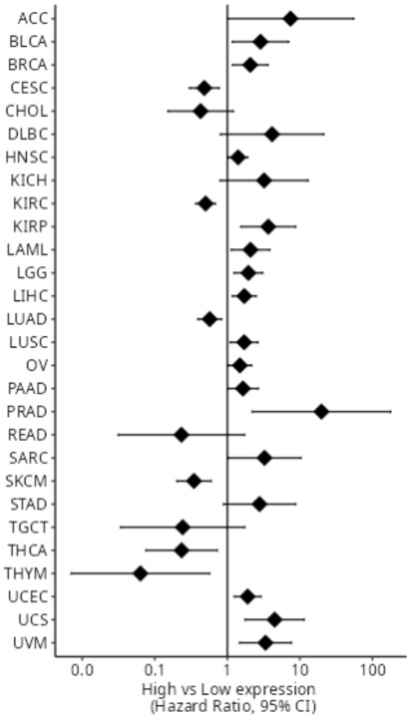
(I) *TRAK2* expression (Tumor vs Normal)

Click on a point to display a detailed plot

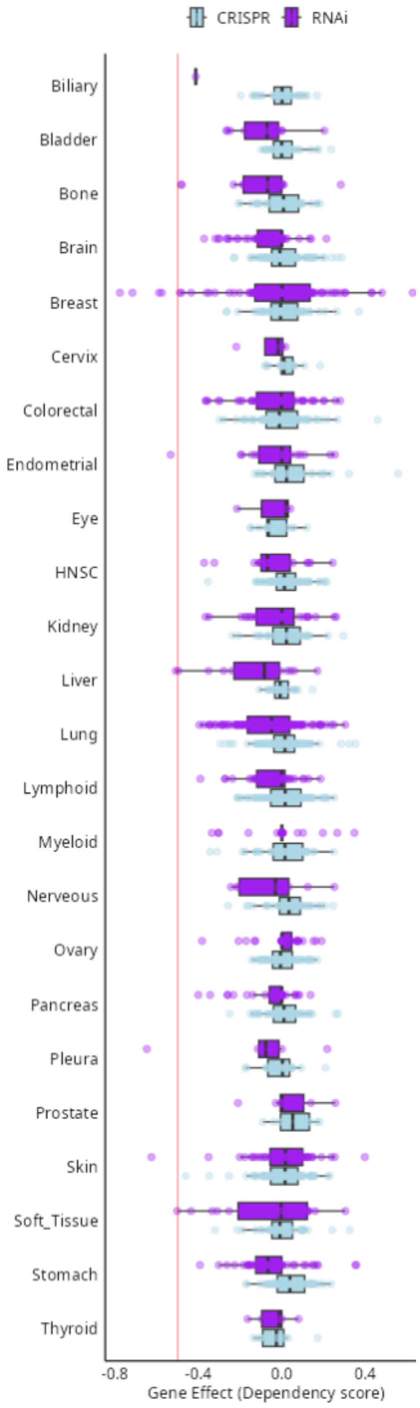


(G) Association of *TRAK2* with overall survival

Click on a point to display a Kaplan Meier curve

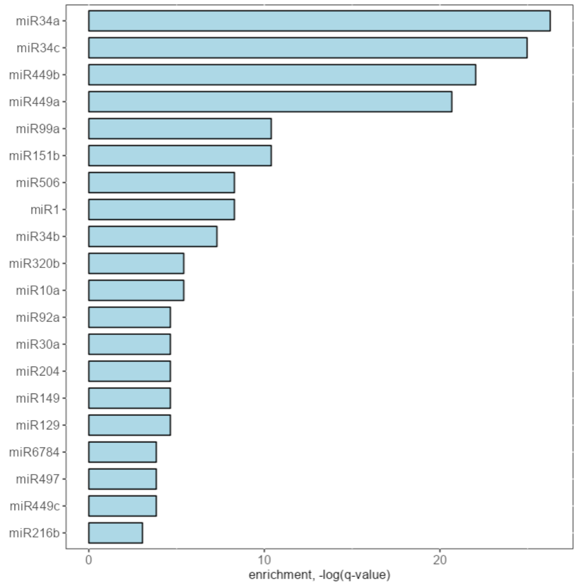


(J) Dependency of cancer cell lines from indicated cancer entities on *TRAK2*



4. THE ENRICHMENT OF GENE LISTS IN miRNA TARGETS TAB

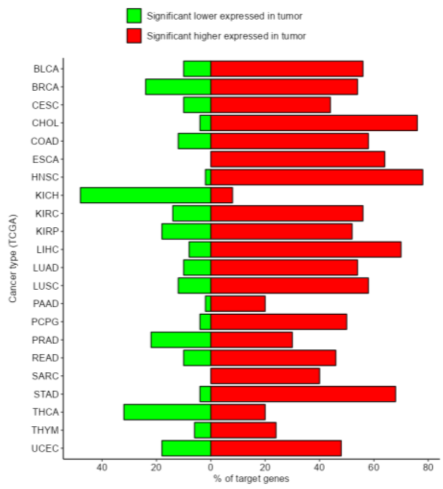
In this tab users can paste a list of genes and miRTARGET will calculate whether potential targets of any miRNA are enriched in the provided list of genes.



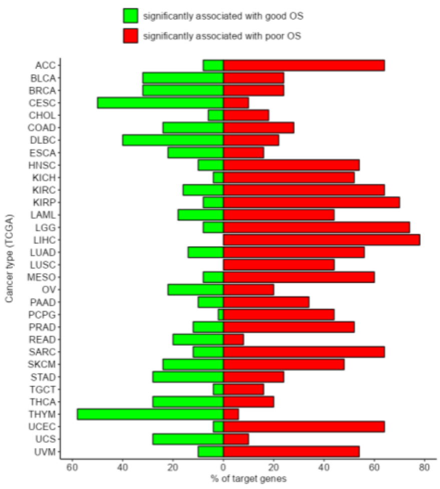
(A) Enrichment of genes from the gene list in targets of indicated miRNA

Select a miRNA bar to display its potential targets that are present in the gene list

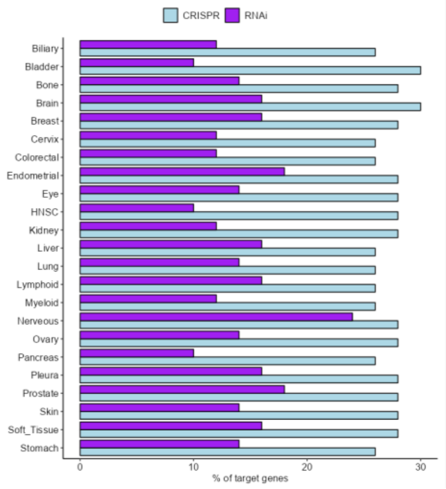
In addition, Plots B and C show the association of genes from the provided gene list with tumor vs normal tissue (B) and overall survival (C) in indicated cancer types. The plot (B) shows the percentage of genes that are expressed significantly higher (red) or lower (green) in tumor vs normal tissue. The plot (D) shows the association of genes from the provided gene list with cancer cell dependency. The percentage of genes that show a dependency in cancer cell lines from indicated cancer types are shown. A gene is considered dependent if at least 10% of cancer cell lines from a specific cancer entity display a dependency score of less than -0.5. Data is from the Dependency Map Portal (www.depmap.org). The CRISPR and RNAi indicates whether the data is based on CRISPR (Chronos) or shRNA (DEMETER2) screening. These plots provide information whether the genes from the provided gene list have predominantly oncogenic or tumor suppressive properties in indicated cancer entities.



(B) Expression of genes from the gene list in Tumor vs Normal

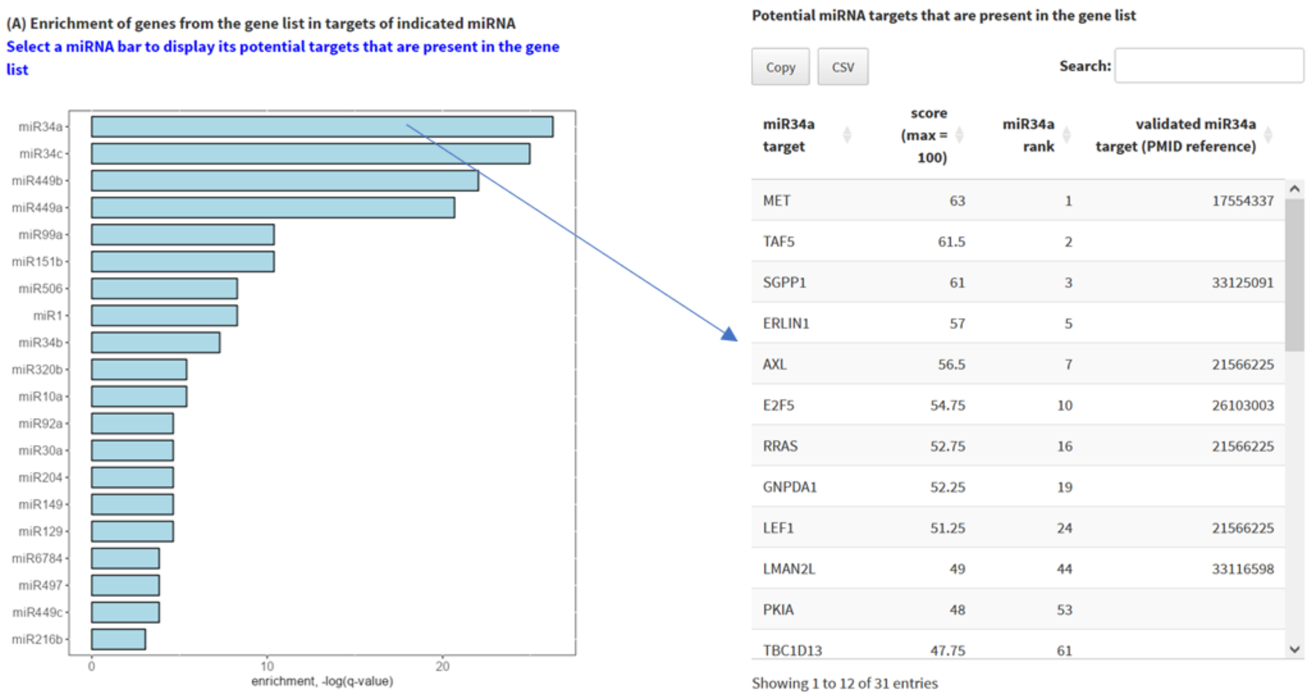


(C) Association of genes from the gene list with overall survival



(D) Association of genes from the gene list with cancer cell lines dependency

Users can click on a miRNA bar to display only the genes from the provided gene list that are predicted targets of that miRNA



(A) Enrichment of genes from the gene list in targets of indicated miRNA

Select a miRNA bar to display its potential targets that are present in the gene list

Potential miRNA targets that are present in the gene list

miR34a target	score (max = 100)	miR34a rank	validated miR34a target (PMID reference)
MET	63	1	17554337
TAF5	61.5	2	
SGPP1	61	3	33125091
ERLIN1	57	5	
AXL	56.5	7	21566225
E2F5	54.75	10	26103003
RRAS	52.75	16	21566225
GNPDA1	52.25	19	
LEF1	51.25	24	21566225
LMAN2L	49	44	33116598
PKIA	48	53	
TBC1D13	47.75	61	

Showing 1 to 12 of 31 entries