Construction and analysis of the lncRNA-miRNA-mRNA network based on competitive endogenous RNA reveal functional lncRNAs in oral cancer

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Abstract

Background

There is a growing body of evidence suggesting that long non-coding RNAs(lncRNAs) can function as a microRNA(miRNA) sponge in various diseases including oral cancer. However, we are still not very clear about the pathophysiological function of lncRNAs.

Methods

We constructed a lncRNA-miRNA-mRNA network in oral cancer based on the competitive endogenous RNA(ceRNA) theory with the human expression profiles GSE74530 from the Gene Expression Omnibus (GEO) database and used topological analysis to determine the hub lncRNAs in the regulatory ceRNA network. Then, function enrichment analysis was performed by R package Cluster Profiler.

Results

A total of 238 potential co-dysregulated competing triples were obtained in the lncRNA-assoicated ceRNA network of oral cancer, which consisted of 10 lncRNA nodes, 41 miRNA nodes, and 122 mRNA nodes. Additionally, we found three lncRNAs(HCP5 , AGAP11 , HCG22) exhibiting superior potential as diagnostic and prognostic markers of oral cancer.

Conclusions

Our findings will provide novel insights for understanding the ceRNA regulation in oral cancer and identify three novel lncRNAs as potential molecular biomarkers.

1. Introduction

Oral cancer is a malignant neoplasia with low rates of overall survival(1) and Oral squamous cell carcinoma(OSCC) is the most common type of oral cancer(2). It most frequently occurs in the buccal mucosa, tongue, and lower lip, and has higher occurrence in people over fifty (3). The detection of oral cancer is difficult, for the tumor is asymptomatic before reaching a certain size. In that case, early diagnosis of oral cancer is important for improving patient survival. Identification of novel specific biomarkers will facilitate detection and confirmation of the oral cancer diagnosis at the molecular level.

Initially, it is thought that coding RNAs play the most essential roles in cancer, while non-coding RNAs(ncRNAs) are no more than transcriptional noise. However, more and more evidence indicates the important regulatory roles of ncRNAs in the occurrence and progression of various cancers(4). The ncRNAs included circular RNAs, microRNAs, intronic RNAs, and long non-coding RNAs(5). MicroRNA(miRNA) is a representative ncRNA of 18–25 nucleotides in length(6), regulating the expression of target genes by inhibiting their translation and accelerating their degradation(7). MiRNAs have been shown to be involved in many different physiological and pathological processes, including epithelial-mesenchymal transition, metabolism, survival and more. First confirmed in 2005, long non-coding RNA(lncRNA) is another type of ncRNA and also lack protein-coding capacity(8). Typically, the nucleotide sequence length of lncRNAs is between 200 and 10,0000nt. In addition, some lncRNAs may be associated with cancer phenotypes(9) and were chosen as new sets of diagnostic and prognostic biomarkers for various cancers, including, for example, nasopharyngeal carcinoma(10), gastric cancer(11), and prostate cancer(12). However, the role of lncRNAs in the development of OSCC remains to be explored.

In 2011, a novel regulatory mechanism that circular RNAs, lncRNAs, and Pseudogenes can regulate the abundance of miRNAs as molecular sponges has received wide attention, which is called competing endogenous RNA(ceRNA) hypothesis(13). The theory may lead to the clues essential to understand gene regulatory networks in many diseases, including OSCC. For example, as one of the initial findings, pseudogene PTENP1 has the miRNA sponge capacity to regulate PTEN levels in cancer(14). ANRIL is a miRNA sponge of miR-125a-3p that regulate FGFR1 abundance to promote the tumorigenesis of head and neck squamous cell carcinoma(15). Through these studies, lncRNAs have shown potential as biomarkers for diagnostic and prognosis of various diseases.

In this study, according to the analysis of RNA expression profiles in oral cancer patients from Gene Expression Omnibus(GEO) database at the National Center for Biotechnology Information(NCBI), we have screened the differentially expressed lncRNAs(DELs) and mRNAs(DEMs) related to oral cancer. Then, we constructed a lncRNA-associated ceRNA network through combining bioinformatics analyses and correlation analyses to find the hub lncRNAs in OSCC.

2. Materials And Methods

2.1. GEO Data Collection

We downloaded human expression profiles(accession GSE74530) of oral cancer from NCBI GEO(16), which was extracted from a study carried out by Oghumu et al.(17). The expression data were derived from tumor tissue and adjacent non-tumor tissue of six different donors. The microarray platform used to analysis these data was the GPL570[HG-U133_Plus_2] Affymetrix Human Genome U133 Plus 2.0 Array.

2.2. Data Quality Assessment

AffyPLM package(18) in the statistical language R is applied to analyze the data quality at the probe level. The boxplot representations of residual and weigh from probe level fits were obtained, with which we can test the trend consistency of the expression data. The degradation of RNA was assessed with the aid of AffyRNAdeg function of AffyPLM, to assure the consistent trend and RNA integrity of the microarray dataset before further processing.

2.3. Data Pre-processing and Screening of Differentially Expressed LncRNAs and MRNAs

For the identification of each probe's biological significance, the comprehensive gene annotation file in GTF format were obtained from GENCODE. The GENCODE annotation was the default gene annotation displayed in the Ensembl Genome Browser. Transcripts having a length of more than 200 nucleotides and a biotype categorized as "non_coding", "processed_transcript","lincRNA","retained_intron","antisense","sense_overlapping","sense_intronic",and"bidirectional_promoter_lncrna" were labeled as "lncRNAs", while transcripts with a biotype categorized as "protein_coding" were labeled as "mRNAs". Finally, there were 1,210 expressed lncRNAs and 16,434 expressed mRNAs annotated.

The Affymetrix probe level data were obtained through reading CEL files with the function ReadAffy of Affy package(19) in R and then the raw data were preprocessed(background correction, normalization, and summary expression computation). We used bioconductor package limma(20) to explore the differentially expressed genes(DEGs) between adjacent normal tissue and tumors groups, including lncRNAs(DELs) and mRNAs(DEMs). The DELs and DEMs were filtered according to the cut-off criteria of adjusted P-values < 0.05 and $|\log_2$ (fold change)|>1.

2.4. Prediction of Target miRNAs of DELs and MiRNA-Target Interactions

The lncRNA targets of miRNAs were collected using the transcriptome-wide mircoRNA target predictions from miRcode database(21) which contained more than 10,000 lncRNAs. The predictions were based on GENCODE transripts. Interactions between miRNA and mRNA were found in 3 databases of miRNAs(miRTarBase 6.0(22), miRDB(23) and TargetScan 7.0(24))with the criteria that each target mRNA appears in at least 2 of them and the mRNA targets of miRNA-mRNA interactions were merged with DEMs for the further analysis.

2.5. Construction of the LncRNA Associated CeRNA Network and Topological Analysis

An lncRNA-miRNA-mRNA interaction was identified as a potential ceRNA triple according to the following thresholds(25):

(1)Pearson's correlation coefficients(PCCs) between each DELs-DEMs pair in oral cancer were calculated. The DEL-DEM pairs were regarded as co-dysregulated DEL-DEM pairs, with thresholds of PCC value ranking in the top 0.05 percentile(PCC > 0.886) and P-value < 0.05.

(2)Once it was confirmed that both mRNA and lncRNA in a co-dysregulated DEL-DEM pair were targeted by the same miRNA, this lncRNA-miRNA-mRNA interaction would be identified as a potential co-dysregulated competing triple.

To give an insight into the roles of lncRNAs in ceRNA network, we assembled all the potential co-dysregulated competing triples to build the lncRNA-miRNA-mRNA network and visualized the regulation network by importing the above interactions into the Cytoscape 3.7.1(26).

Topological analysis is important in discovering information in complex data sets. To study the geometric relationships between data nodes, we computed the node degree and betweeness centrality(BC) of each node, which are both network topological features. The nodes with a high node degree(> 5 connections) and a greater BC value were considered to be the hub nodes in the regulation network, which were more likely to play an important role in oral cancer(27).

2.6. Functional Enrichment Analysis

We performed Gene ontology(GO) gene function enrichment analysis and Kyoto Encyclopedia of Genes and Genomes(KEGG) pathways analysis of mRNAs in the lncRNA-associated regulation network using Cluster Profiler in R(28) to explore functions of the lncRNAs we obtained. Simultaneously, the GO interaction network was built with the Biological Networks Gene Ontology tool(BinGO) in Cytoscape(26). We set p-value < 0.05 and Benjamini corrected P-value < 0.05 as the thresholds of functional categories.

2.7. Construction Of The Key LncRNA Associated Sub-network

The lncRNAs with high node degree as well as BC value were chosen as hub lncRNAs, and the sub-networks were constructed with these lncRNAs, their related miRNAs and mRNAs in the regulation network by Cytoscape. Thereafter, gene functional enrichment analyses were performed for each sub-network.

3. Results

3.1. Data Quality Assessment and Preprocess

Regression analysis of raw data was performed by using affyPLM package in R. The relative log expression(RLE) plot revealed that gene expressions levels in GSE74530 were consistent with the median approaching 0(Fig. 1A), indicating that the quality of expression data was reliable. As for the RNA degradation plot, it showed that the RNA integrity is in good quality(Fig. 1B). Therefore, all 12 samples can be used for further analysis. There were a total of 16434 mRNAs and 1210 lncRNAs that were identified in microarray data using Human Comprehensive gene annotation from GENCODE. As shown in Fig. 1C, there is no obvious outliers in spread and location in boxplot and small discrepancies can be sufficiently removed by normalization. The median of 12 samples were almost at the same level after normalization (Fig. 1D), which corrected the systematic differences between chips effectively.

3.2. The Screening Results Of DELs And DEMs

We identified the 34 DELs and 1641 DEMs by comparing the tumor groups with the adjacent normal tissue group through limma package. The expression of 34 DELs and all DEGs were shown in a heatmap (Fig. 2) and a volcano plot(Fig. 3) respectively. We showed in. The miRNA targets of lncRNAs were predicted using the miRcode database in R. The target mRNAs of these miRNAs were obtained by three highly reliable microRNA target prediction databases (miRTarBase, TargetScan, and miRDB) and the result was intersected with the DEMs mentioned above.

After integrating the two methods above, we got a total of 2137 reliable miRNA-mRNA pairs and 162 predicted lncRNA- miRNA pairs (including 11 lncRNAs, 51 miRNAs and 851 mRNAs) for further analysis.

3.3. Construction Of CeRNA Network In Oral Cancer

To reduce false positives, we calculated the PCC between 34 DELs and 1,641 DEMs and co-expressed pairs with the top five percent PCC value(PCC > 0.886) were defined as the significant co-dysregulated competing pairs. Then the results were merged with 11 lncRNAs, 51 miRNAs and 851 mRNAs. By taking the intersection, a total of 238 potential co-dysregulated competing triples were selected and the full list was shown in Additional file 1.

To go deep into the functional study of lncRNAs acting as miRNA sponge in oral cancer, we built a ceRNA network and applied Cytoscape to perform visualization(Fig. 4A). In the network, there were 10 lncRNA nodes, 41miRNA nodes, 122 mRNA nodes and 238 edges.

3.4. Functional Enrichment Analysis

We speculated on the possible function of each lncRNAs through functional enrichment analysis of their linked mRNAs. All of the mRNA nodes were performed to analyze their function via the GO analysis and the GO interaction network was constructed by BinGO(Additional file 2). Eighteen GO terms were significantly enriched (Fig. 4B and Additional file 3). Out of these, the top three enriched terms were collagen binding, extracellular matrix binding and cell adhesion molecule binding all of which belonged to molecular function. Interestingly, extracellular matrix(ECM) binding was related to the proliferation of OSCC cells(29) and ECM played an important role in the growth and survival of oral cancer cells(30). It was demonstrated that cell adhesion molecules, together with tumor-associated matrix molecules, function in the progression of oral cancer(31). What's more, discoidin domain receptor-1 (DDR1) could be activated by the specific binding with collagens (II,III)(32) and activation of DDR1 have been reported in oral cancer(33). Ten KEGG pathway terms were shown in Fig. 4C, including ECM-receptor interaction, small cell lung cancer, PI3K-Akt signaling pathway, Focal adhesion, TGF-β signaling pathway and more (Additional file 4). Among these ten pathways, ECM-receptor interaction(34), PI3K-Akt signaling pathway(35), TNF signaling pathway(36) and TGF-β signaling pathway(37) were OSCC-related pathways.

3.5. Topological Analysis Of The CeRNA Network

To identify the hub genes in the oral cancer related lncRNA-miRNA-mRNA network, we computed the node degrees. In the study of Han et al.(27), they defined nodes with degree greater than 5 as a hub. Basing on this research, a total of 42 nodes could be chosen as hubs, including 10 lncRNAs, 28 miRNAs, and 4 mRNAs.(Table 1 and Additional file 5) In addition, BC was also calculated as a measure to select the hubs(38)(Table 2). The higher BC value a node had, the more important it would be in the regulatory network(39).

Table 1
The list of differentially expressed genes(node degree > 5).

Number	Degree	Name	Type
1	27	hsa-miR-17-5p	miRNA
2	24	HCP5	lncRNA
3	21	AGAP11	lncRNA
4	19	hsa-miR-24-3p	miRNA
5	18	hsa-miR-27a-3p	miRNA
6	17	hsa-miR-1297	miRNA
7	17	hsa-miR-23b-3p	miRNA
8	16	hsa-miR-140-5p	miRNA
9	16	hsa-miR-129-5p	miRNA
10	15	CRNDE	lncRNA
11	15	hsa-miR-22-3p	miRNA
12	15	hsa-miR-20b-5p	miRNA
13	14	HCG22	lncRNA
14	13	EPB41L4A-AS1	lncRNA
15	13	hsa-miR-507	miRNA
16	13	hsa-miR-107	miRNA
17	13	hsa-miR-216b-5p	miRNA
18	12	SOX21-AS1	lncRNA
19	12	hsa-miR-3619-5p	miRNA
20	11	UCA1	lncRNA
21	11	hsa-miR-761	miRNA
22	9	hsa-miR-490-3p	miRNA
23	9	hsa-miR-125b-5p	miRNA
24	9	hsa-miR-1244	miRNA
25	9	hsa-miR-139-5p	miRNA
26	7	YOD1	mRNA
27	7	LINC00491	lncRNA
28	7	hsa-miR-125a-5p	miRNA
29	7	hsa-miR-425-5p	miRNA
30	7	hsa-miR-363-3p	miRNA
31	7	hsa-miR-876-3p	miRNA
32	7	hsa-miR-455-5p	miRNA
33	7	hsa-miR-217	miRNA

Number	Degree	Name	Type
34	6	NEK6	mRNA
35	6	MFHAS1	mRNA
36	6	RORA	mRNA
37	6	hsa-miR-33a-3p	miRNA
38	6	LINC00515	lncRNA
39	6	AC073321	lncRNA
40	6	hsa-miR-146b-5p	miRNA
41	6	hsa-miR-508-3p	miRNA
42	6	hsa-miR-135a-5p	miRNA

We found that three lncRNAs(HCP5, AGAP11, and HCG22) not only had higher node degrees, but also had a greater BC, suggesting that they may be potential key regulators controlling the oral cancer related ceRNA network.

Table 2
The list of top 15 high betweenness centrality genes.

Number	Type	name	BetweennessCentrality
1	lncRNA	HCP5	0.26
2	lncRNA	AGAP11	0.16
3	miRNA	hsa-miR-17-5p	0.14
4	miRNA	hsa-miR-24-3p	0.13
5	miRNA	hsa-miR-27a-3p	0.10
6	miRNA	hsa-miR-1297	0.10
7	miRNA	hsa-miR-22-3p	0.09
8	lncRNA	CRNDE	0.09
9	miRNA	hsa-miR-140-5p	0.09
10	miRNA	hsa-miR-129-5p	0.08
11	lncRNA	HCG22	0.08
12	miRNA	hsa-miR-216b-5p	0.07
13	miRNA	hsa-miR-507	0.07
14	miRNA	hsa-miR-23b-3p	0.06
15	miRNA	hsa-miR-107	0.06

3.6. Key LncRNA-miRNA-mRNA Sub-network

Based on the above analysis, we obtained three key lncRNAs(HCP5, AGAP11, and HCG22) that may play a role in oral cancer. Therefore, three more specific functional lncRNA-associated sub-networks with the hub lncRNAs and their linked miRNAs and mRNAs in the regulatory network was constructed. There were 1 lncRNA, 19 miRNAs, 21 mRNAs and 48 edges in AGAP11-associated sub-network(Fig. 5). As shown in Fig. 6, the subnetwork of HCP5 consisted of 1 lncRNA, 23 miRNAs, 53 mRNAs and 110 edges. HCG22 interacted with 14 miRNAs and 34 mRNAs(Fig. 7).

Accordingly, to further understand the biological functions of these three lncRNAs, we performed GO functional enrichment analysis and KEGG pathways analysis for each hub-associated subnetwork. The results of functional enrichment analysis revealed 14 GO terms and 11 pathways terms in the AGAP11-associated sub-network(Fig. 8A). As for HCP5, 15 GO terms and 6 KEGG terms were identified(Fig. 8B). There were 10 differentially enrichment GO terms and 10 pathways related to HCG22(Fig. 8C).

4. Discussion

In our study, we used the expression profiles from NCBI GEO to construct a lncRNA-associated network of oral cancer based on the ceRNA theory. The network consisted of 10 lncRNA nodes, 41 miRNA nodes, and 122 mRNA nodes, according to the results of bioinformatics prediction and correlation analysis. Functional enrichment analysis has been used to reveal biological functions of mRNAs, ceRNA counterparts of lncRNA(39). As a result, three differentially enrichment GO terms(collagen binding(40), extracellular matrix binding(29) and cell adhesion molecule binding(31)) were associated with oral cancer. Moreover, there were four KEGG pathways(TGF-β signaling pathway(37), ECM-receptor interaction(30), TNF-signaling pathway(36) and PI3K-Akt signaling pathway(35)) showing some importance in the tumorigenesis of OSCC. It was pointed out that nodes with a higher degree of connectivity to other nodes are often more important in the network. By applying topology analysis to ceRNA network, we screened out three lncRNAs(HCP5、AGAP11、HCG22) with significantly higher degree and BC in comparison with other nodes, which means that these hubs are essential in network organization and play a critical role in the ceRNA network.

In the results of our research, HCP5 has the greatest number of connection with mRNAs in ceRNA network in oral cancer. However, there is no experimental evidence supporting the contribution of HCP5 to the development of oral cancer. Based on the HCP5 associated sub-network, we proposed that HCP5 might be an essential regulator in oral cancer by sponging miRNAs. Several miRNAs competed by HCP5 and mRNA were associated with oral cancer. For example, miR-139-5p could induce oral cancer cell apoptosis through Akt signaling pathway(41)and could be used as an effective biomarker for detection of tongue squamous cell carcinoma(TSCC)(42). Another study has demonstrated that miR-140-5p targeted ADAM10 and Inhibited the invasion and migration of TSCC cells(43). It has been found that miR-17-5p Inhibits hypoxia-induced apoptosis while regulation of apoptosis is a hallmark of OSCC pathogenesis(44). Zhang et al.(15) found that ANRIL promotes tumorigenesis through sponging miR-125a in oral cancer. The expression changes of miR-125b altered the abundance of its target BAK1 which may control the apoptotic pathway in oral cancer(45). These studies may support our proposal of HCP5's regulatory function in oral cancer. Moreover, Functional annotations of the 53 putative target mRNA in the HCP5-miRNA-mRNA sub-network revealed the biological functions of HCP5. It has been demonstrated that normal cell migration requires the interactions with the extracellular matrix (ECM), which mainly includes collagens, laminins, and fibronectin(30), while changes in composition of ECM may contribute to the development and invasion of oral cancer cells. In detail, a change was observed that more fibrillary collagen type III than thick collagen type I existed in poorly-differentiated squamous cell carcinoma compared with well-differentiated one(46). When oral cancer cells invaded the connective tissue region from the basal membrane, a switch of ECM's composition from laminin-enriched environment to collagen and fibronectin-enriched one would influence metastatic and invasive behavior of tumor cells, since tumor formation was highly sensitive to the microenvironment(47). Another GO term, regulation of transcription by RNA polymerase II also had a connection with oral cancer. Xu et al. have provided evidence that Histone acetylation and RNA polymerase II recruitment at integrin β6 promoter involved in TGF-β1-induced integrin β6 expression in OSCC cells, which would then promote tumorigenesis and metastasis(48). Additionally, six KEGG pathways, including Human papillomavirus infection, ECM-receptor interaction, Small cell lung cancer, TGF-β signaling pathway, PI3K-Akt signaling pathway, and TNF signaling pathway are determined. In accordance with results of a previous study, ECM-receptor interaction pathway was one of the most significantly altered pathways in OSCC samples(49). Tang et al.(36) indicated that TNF-α enhances the invasion and metastasis ability of OSCC cells via the NF-Kb signaling pathway. TGF-β/Smad pathway contributed to oral cancer tumorigenesis(37) and PI3K-Akt signaling pathway was also considered important to the development of OSCC(35). In summary, HCP5 may have the potential to become a novel biomarker for detection and diagnosis of oral cancer.

Importantly, these results provided us with important information regarding the diagnostic and prognostic role of lncRNAs in oral cancer and pointed out three lncRNAs (HCP5、AGAP11、HCG22) as candidate prognosis biomarkers or potential therapeutic targets. However, since there were no verification experiments in our study, the functional role of three lncRNAs were still in need of further research.

5. Conclusion

Overall, we constructed a lncRNA–miRNA–mRNA network based on the ceRNA theory, enabling us to screen and analyze the lncRNAs that play functional roles in the progression of OSCC as miRNA sponges. We further identified three hub lncRNAs (HCP5、AGAP11、HCG22) in the complex ceRNA network. This study not only offered a unique insight into the ceRNA regulation network in OSCC, but also laid the foundation for further experimental and clinical research.

6. Abbreviations

GEO, Gene Expression Omnibus database; ceRNA, competing endogenous RNA; lncRNA, long non-coding RNA; mRNA, messenger RNAs; miRNA, microRNA; OSCC, Oral squamous cell carcinoma; ncRNA, non-coding RNA; NCBI, National Center for Biotechnology Information; DEG, differentially expressed gene; DEL, differentially expressed lncRNA; DEM, differentially expressed mRNA; PCC, Pearson's correlation coefficient; GO, Gene ontology; KEGG, Kyoto Encyclopedia of Genes and Genomes; BC, betweeness centrality; BinGO, Biological Networks Gene Ontology tool; RLE, relative log expression; ECM, extracellular matrix;

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Availability of data and materials

The datasets used and/or analyzed during the current study are available in the GEO repository, [https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE74530].

Competing interests

The authors declare no potential conflicts of interest.

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Additional Figure Captions

Additional file 2. Gene ontology (GO) terms interaction network. Yellow nodes mean nodes with P-value <0.05 and Benjamini corrected P-value <0.05.

Additional file 5. All node degree analysis reveals the distribution of the points with different node degrees in ceRNA network.

Figures

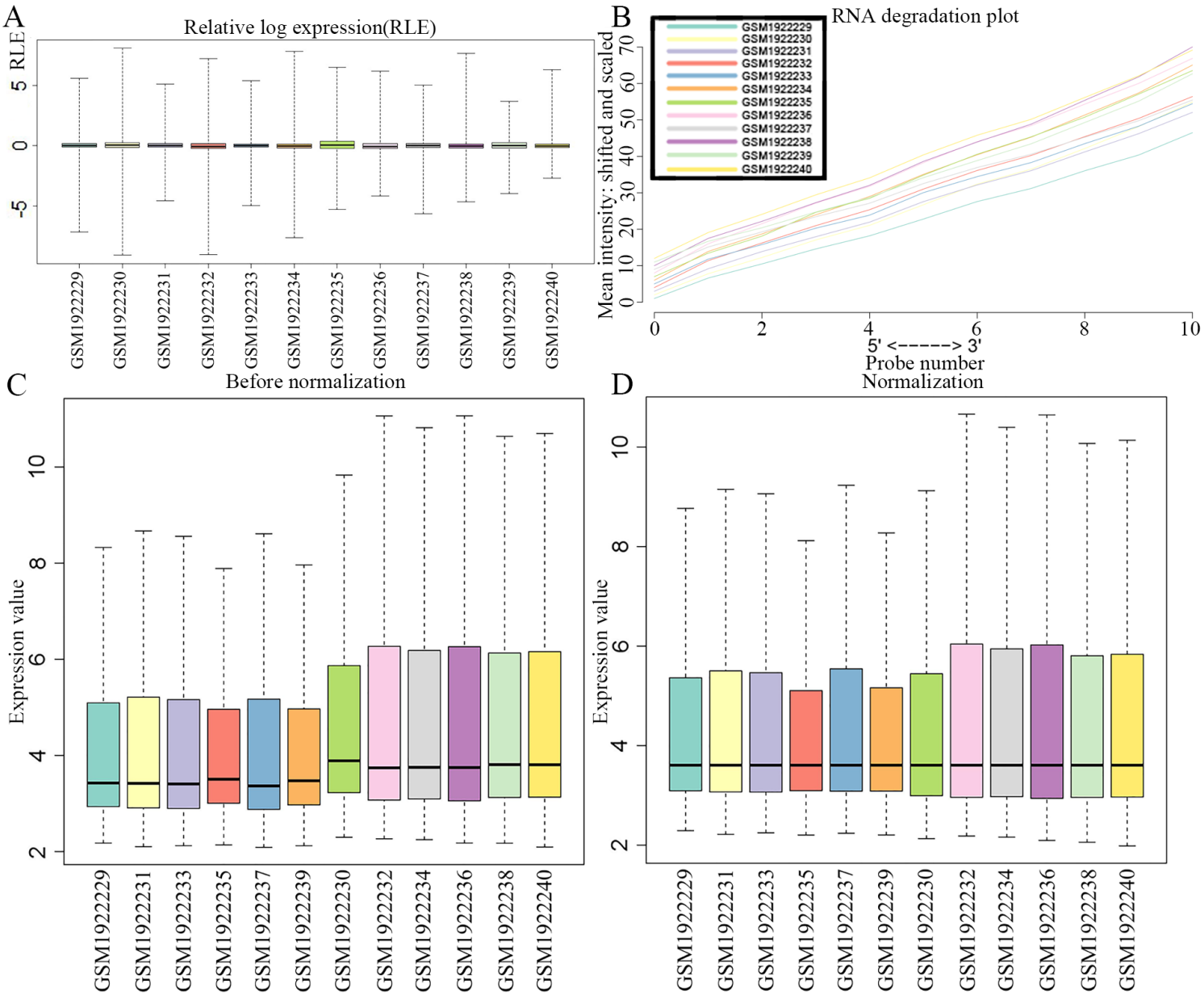

Figure 1

Data quality assessment. (A) Boxplot representation of the relative log expression(RLE). (B) RNA degradation plot, twelve curves represent twelve different samples, respectively. (C) Boxplot of intensity distributions in the raw data. (D) Boxplot of intensity distributions in the normalized data. The relative expression values are comparable among all twelve samples after normalization.

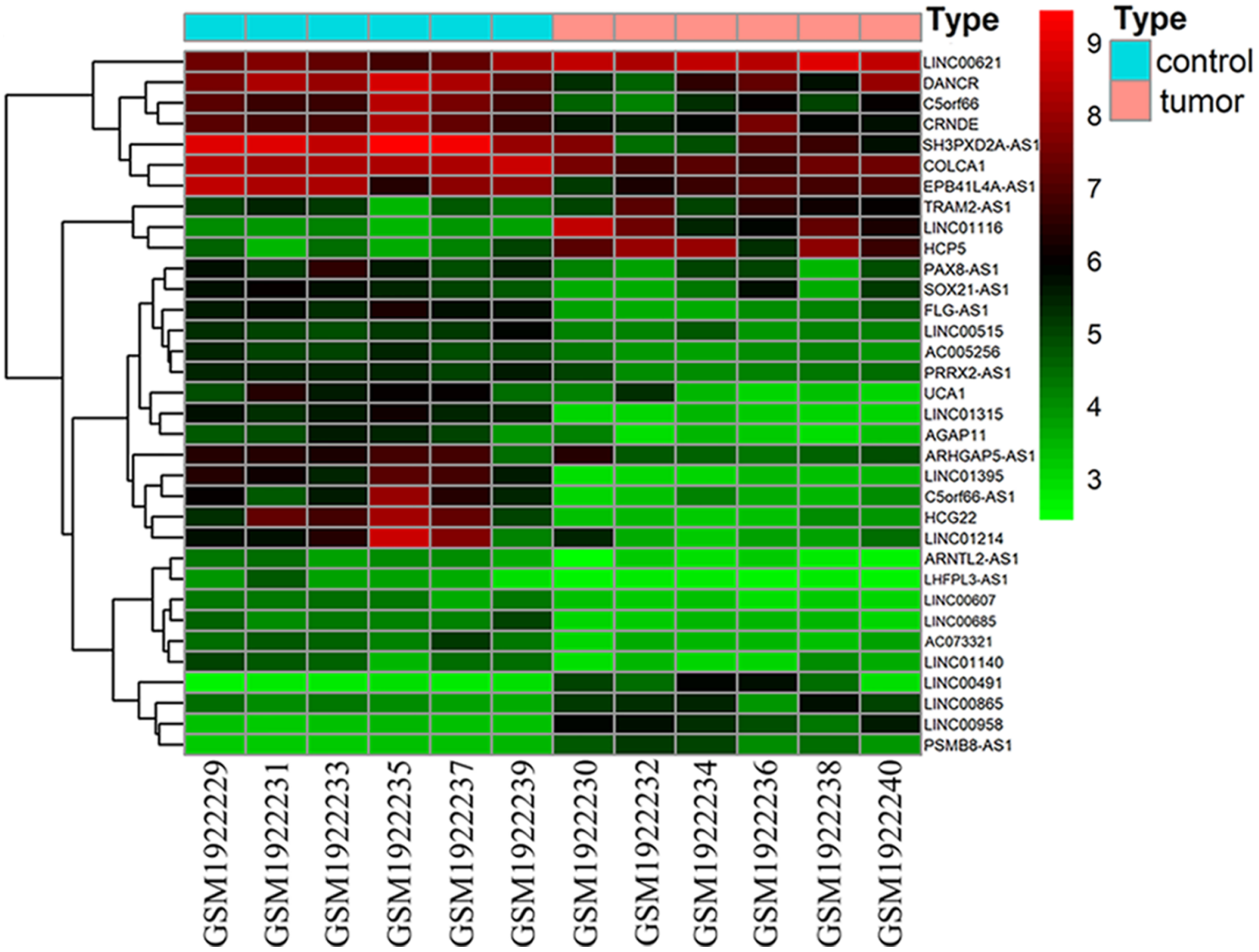

Figure 2

Heatmap of differentially expressed lncRNAs in oral cancer. The horizontal axis shows the names of twelve samples. The vertical axis presents the gene names.

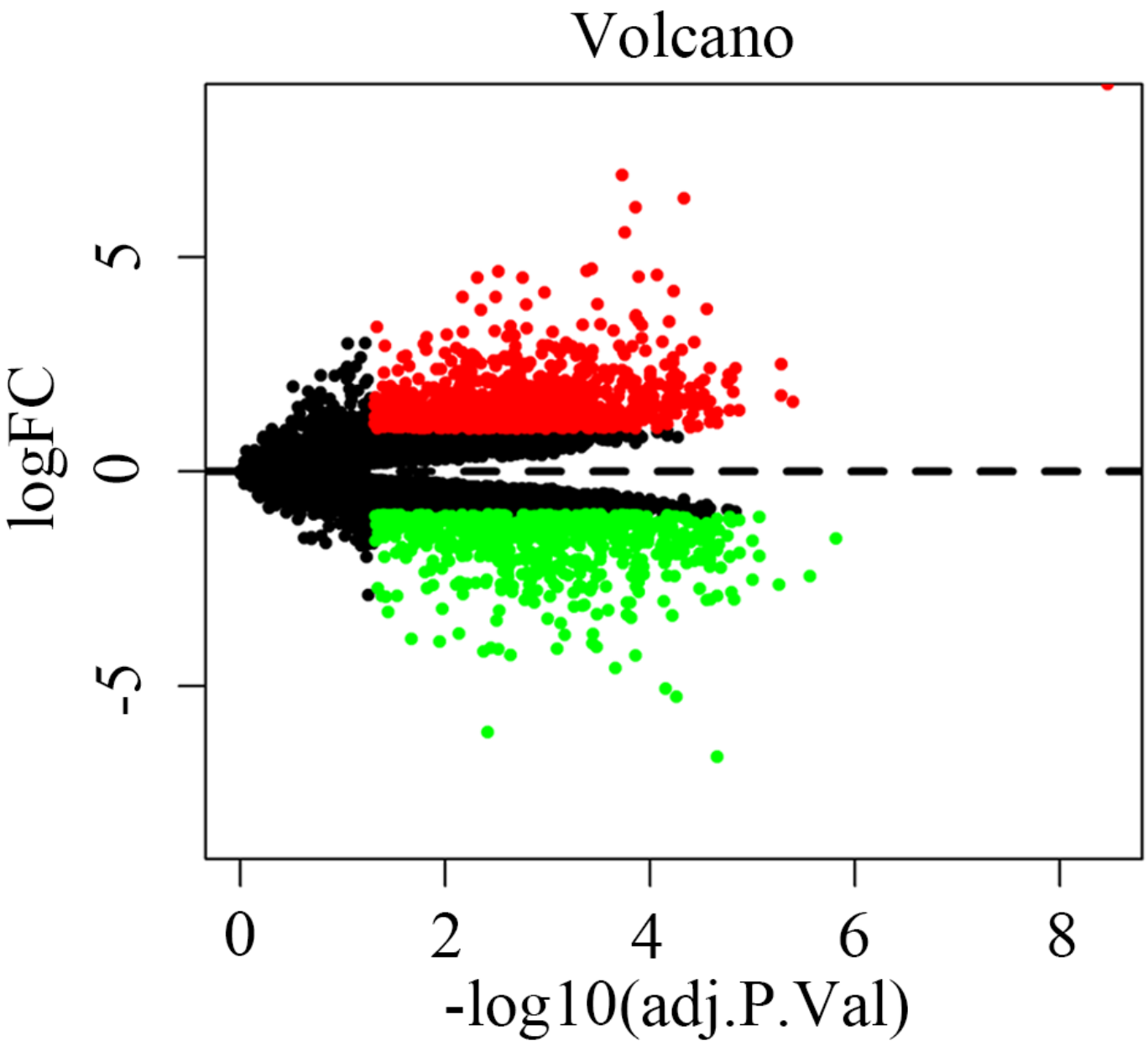

Figure 3

Volcano plot of all differentially expressed genes in oral cancer. FC are fold-change. Downregulated genes are green and upregulated genes are red.

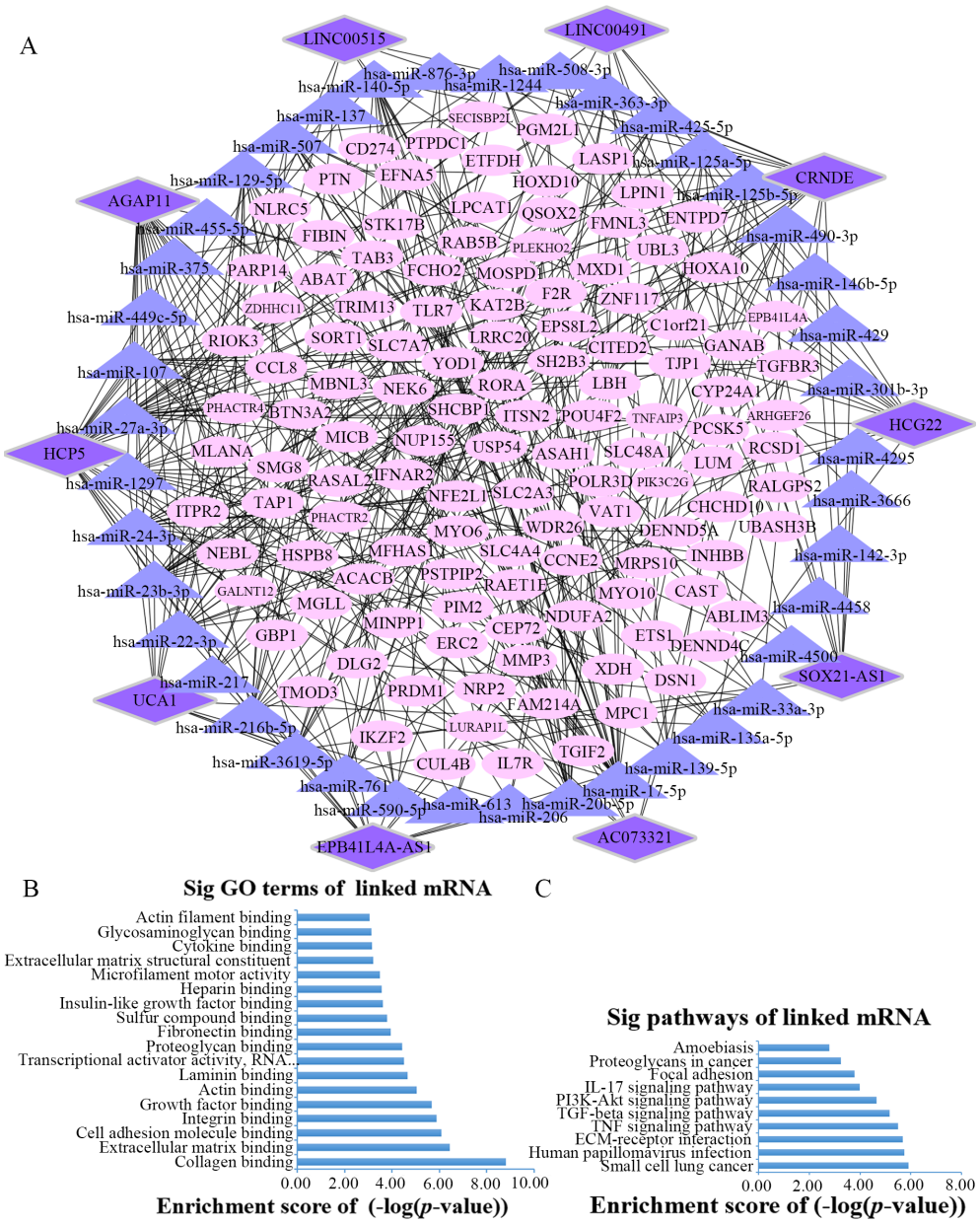

Figure 4

The lncRNA associated ceRNA network and barplots of function enrichment analyses. (A)The lncRNA-miRNA-mRNA ceRNA network. The parallelograms represent lncRNAs, the ellipses represent mRNAs, and the triangles represent miRNAs. (B) The top 18 most significant Gene ontology terms. (C) The top 10 most significant pathway terms.

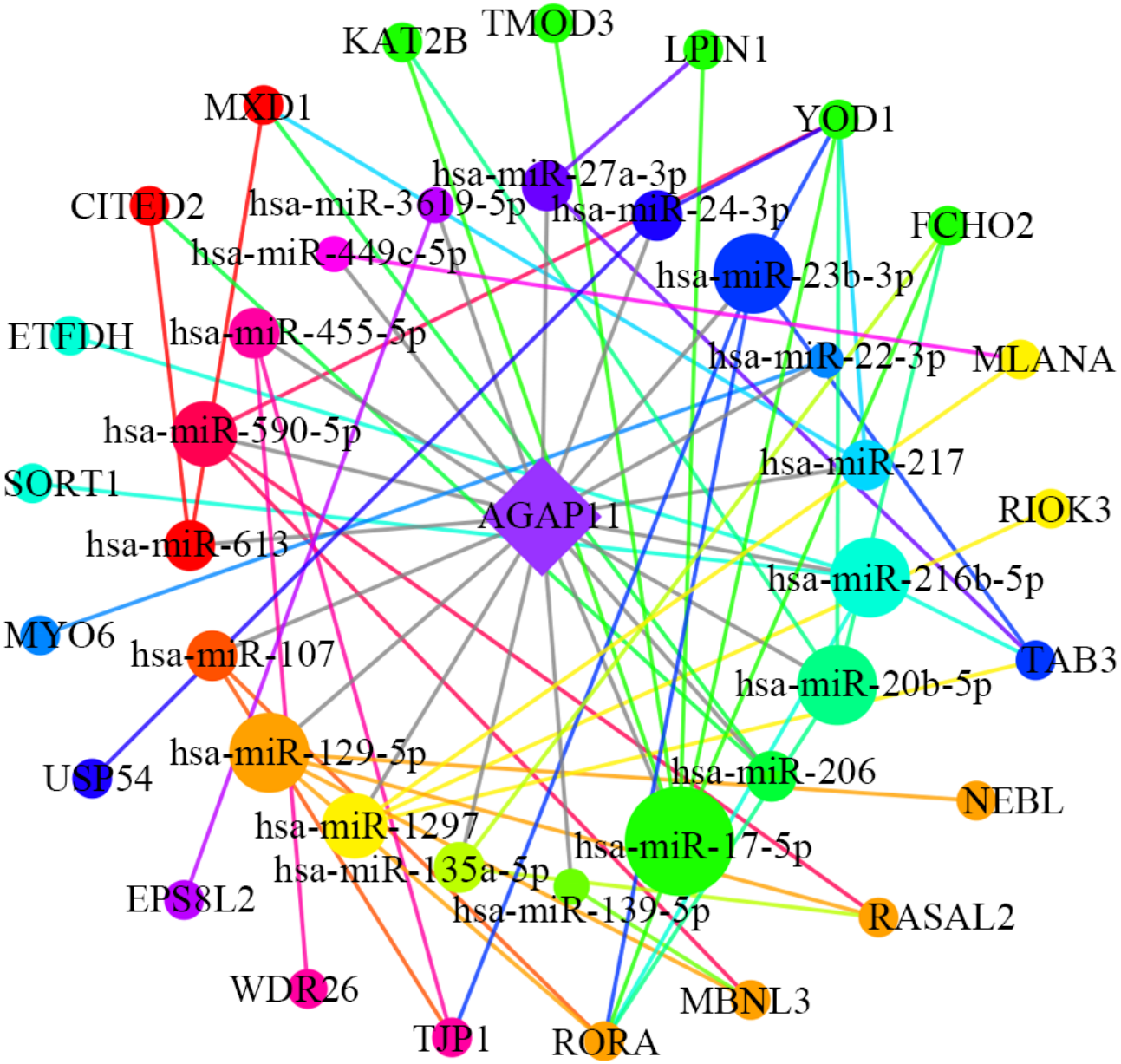

Figure 5

The sub-network of hub lncRNA AGAP11. The rhombuses represent lncRNAs, the circles on the inner loop represent miRNAs, and the circles on the outer loop represent mRNAs. The bigger size circles have, the more nodes they are connected to.

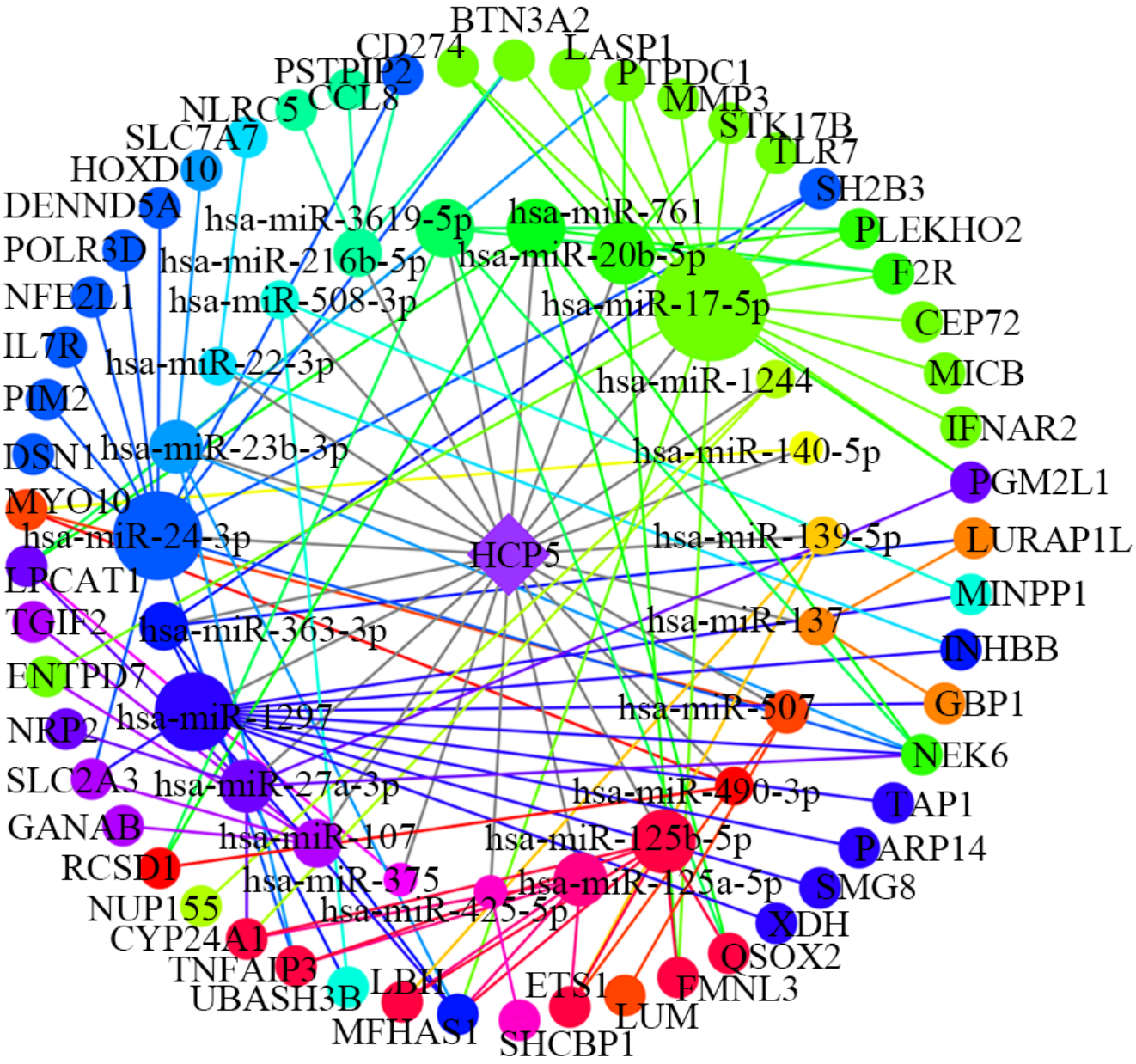

Figure 6

The sub-network of hub lncRNA HCP5. The rhombuses represent lncRNAs, the circles on the inner loop represent miRNAs, and the circles on the outer loop represent mRNAs. The bigger size circles have, the more nodes they are connected to.

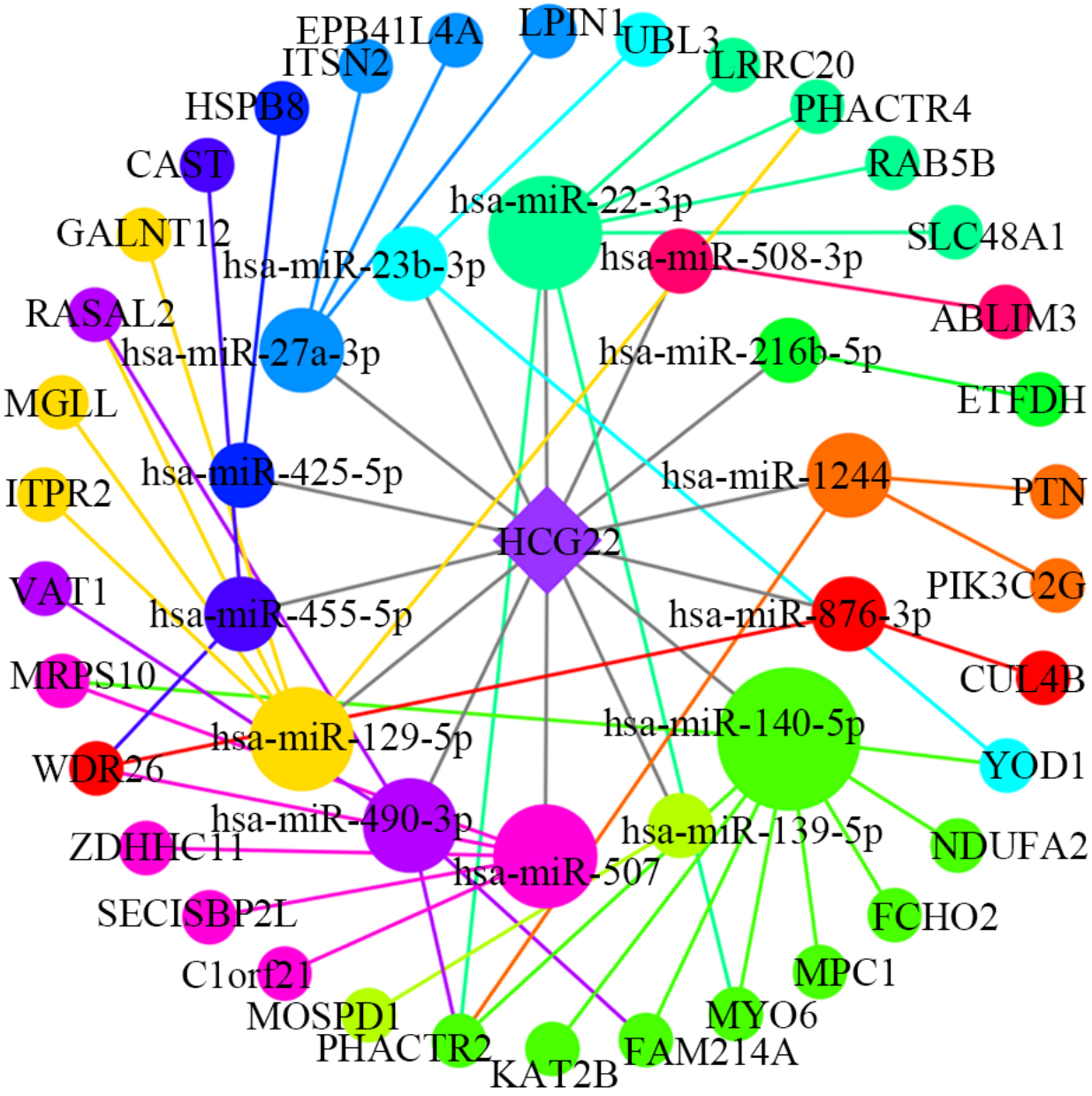

Figure 7

The sub-network of hub lncRNA HCG22. The rhombuses represent lncRNAs, the circles on the inner loop represent miRNAs, and the circles on the outer loop represent mRNAs. The bigger size circles have, the more nodes they are connected to.

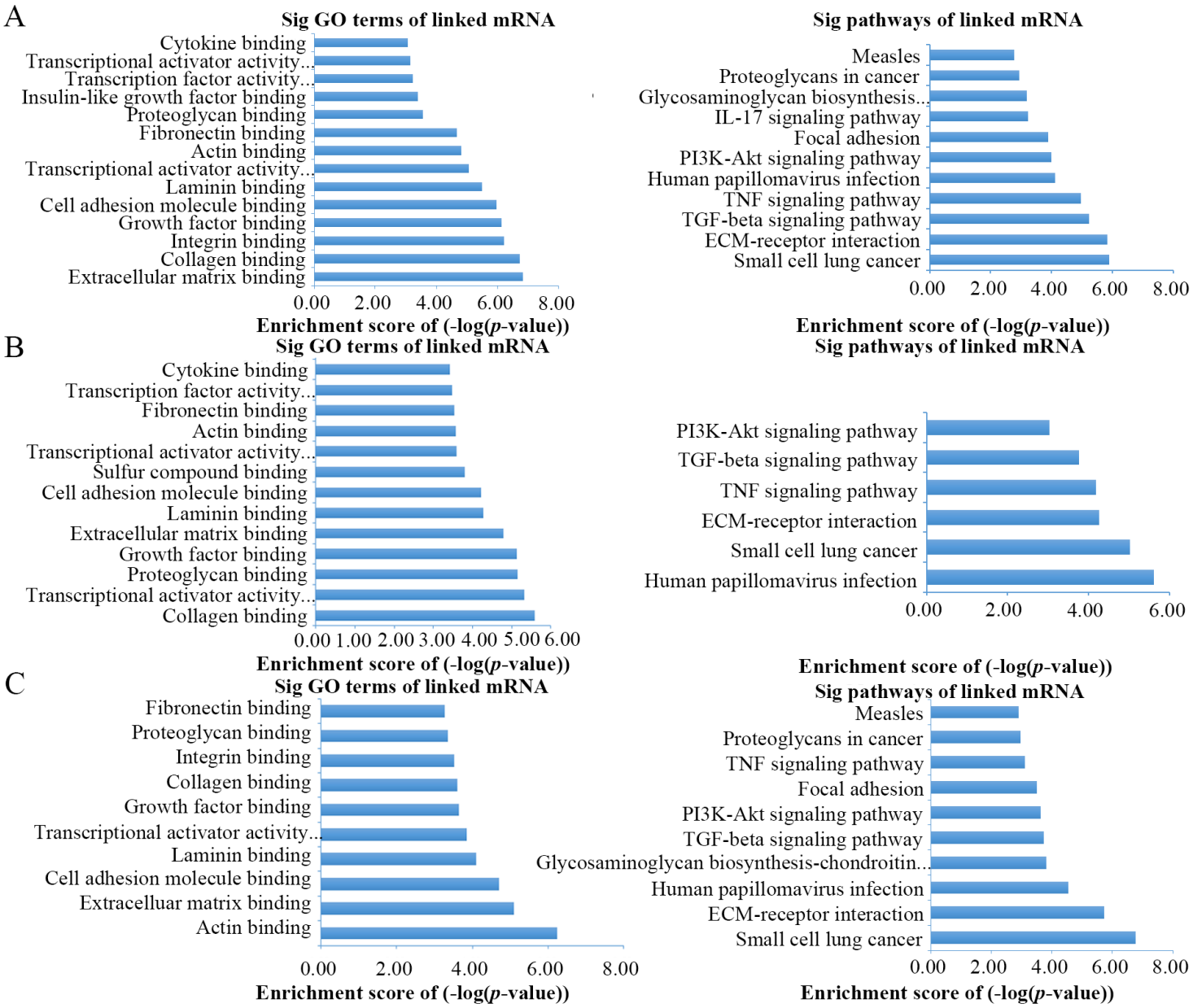

Figure 8

The barplots of function enrichment analyses. (A) Functional enrichment analyses for AGAP11-related mRNAs. (B) Functional enrichment analyses for HCP5-related mRNAs. (C) Functional enrichment analyses for HCG22-related mRNAs.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Additionalfile4.docx
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