Construction of a Myc-associated ceRNA network reveals a prognostic signature in hepatocellular carcinoma

Dan-Dan Zhang,1,2,3,7 Yi Shi,2,7 Ji-Bin Liu,2,7 Xiao-Li Yang,1,7 Rui Xin,1,7 Hui-Min Wang,1,2 Pei-Yao Wang,1 Cheng-You Jia,3 Wen-Jie Zhang,3,5 Yu-Shui Ma,6 and Da Fu1

1Central Laboratory for Medical Research, Shanghai Tenth People’s Hospital, Tongji University School of Medicine, Shanghai 200072, China; 2Cancer Institute, Nantong Tumor Hospital, Nantong 226631, China; 3Department of Pathology, Shihzi University School of Medicine, Shihzei, Xinjiang 832002, China; 4Department of Nuclear Medicine, Shanghai Tenth People’s Hospital, Tongji University School of Medicine, Shanghai 200072, China; 5The Key Laboratories for Xinjiang Endemic and Ethnic Diseases, Shihzi University School of Medicine, Shihzei, Xinjiang 832002, China; 6International Cooperation Laboratory on Signal Transduction, Eastern Hepatobiliary Surgery Hospital/Institute, National Center for Liver Cancer, the Second Medical University, Shanghai 204543, China

Hepatocellular carcinoma (HCC) remains an extremely lethal disease worldwide. High-throughput methods have revealed global transcriptome dysregulation; however, a comprehensive investigation of the complexity and behavioral characteristics of the competing endogenous RNA (ceRNA) network in HCC is lacking. In this study, we extracted the transcriptome (RNA) sequencing data of 371 HCC patients from The Cancer Genome Atlas platform. With the comparison of the high Myc expression (Myc-high) tumor and low Myc expression (Myc-low) tumor groups in HCC, we identified 1,125 differentially expressed (DE) mRNAs, 589 long non-coding RNAs (lncRNAs), and 93 micro-RNAs (miRNAs). DE RNAs predicted the interactions necessary to construct an associated Myc ceRNA network, including 19 DE lncRNAs, 5 miRNAs, and 72 mRNAs. We identified a significant signature (long intergenic non-protein-coding [LINC] RNA 2691 [LINC02691] and LINC02499) that effectively predicted overall survival and had protective effects. The target genes of microRNA (miR)-212-3p predicted to intersect with DE mRNAs included SEC14-like protein 2 (SEC14L2) and solute carrier family 6 member 1 (SLC6A1), which were strongly correlated with survival and prognosis. With the use of the lncRNA-miRNA-mRNA axis, we constructed a ceRNA network containing four lncRNAs (LINC02691, LINC02499, LINC01354, and NAV2 antisense RNA 4), one miRNA (miR-212-3p), and two mRNAs (SEC14L2 and SLC6A1). Overall, we successfully constructed a mutually regulated ceRNA network and identified potential precision-targeted therapies and prognostic biomarkers. Although there have been great advancements in cancer therapy, the 5-year overall survival (OS) rate for HCC is still only 12.10%.1–5 Surgical resection, liver transplantation, and chemotherapy are common therapies,6–11 but they are appropriate only for patients with early-stage disease.16,17 Therefore, it is essential to understand the mechanisms underlying the pathogenesis of HCC and identify novel biomarkers.

In the past several years, the advancement of high-throughput technologies has provided more opportunities for biomarker identification.18–21 Non-coding RNAs (ncRNAs), which include long ncRNAs (lncRNAs) and microRNAs (miRNAs), play a role in cancer progression and are potential biomarkers and therapeutic targets.22–25 lncRNAs are transcripts with more than 200 nucleotides, which previously were thought to have no biological function. But in fact, lncRNAs are involved in several biological processes, including cell differentiation, cancer proliferation, and metastasis.26–28 Competing endogenous RNA (ceRNA), including IncRNA, can interact with mRNA by competitively combining different miRNAs.29–31 miRNAs are a class of ncRNA molecules of 21–24 nucleotides that mediate negative post-transcriptional regulation.32–35 When the miRNA arm-imbalance mechanism is broken in the cell, dysregulation of...
downstream tumor-suppressor genes or oncogenes controlled by aberrant miRNAs occurs, leading to cancer development.36–40 For example, lncRNA LPP antisense RNA 2 has carcinogenic effects and promotes cell proliferation and metastasis through microRNA (miR)-7-5.41 The well-studied tumorigenic lncRNA, which is highly upregulated in liver cancer (HULC), serves as a ceRNA network through miR-372.42 AGAP2 antisense RNA 1, a competitive lncRNA, functions as an oncogene and upregulates annexin A11 expression through miR-16-5p, promoting proliferative capacity in liver cancer.43 These ceRNA networks provide a novel perspective and offer insight into undetected biomarkers for the early diagnosis and treatment of cancer.

The Myc gene is located on chromosome 8q24.21, which is dysregulated in most human neoplasia. This region is frequently genetically amplified in various human cancers.44 Moreover, MYC is overexpressed in more than 50% of tumors, including HCC. In this study, we first used the median expression level of Myc to divide the 371 HCC samples into two groups for subsequent analysis: high Myc expression (Myc\textsuperscript{high}) tumor and low Myc expression (Myc\textsuperscript{low}) tumor. We used the transcriptome sequencing data of 371 lncRNAs, 371 mRNAs, and 367 miRNAs of HCC tumor tissue and adjacent normal tissue from The Cancer Genome Atlas (TCGA) platform to identify key differentially expressed (DE) RNAs, and we constructed an associated Myc ceRNA network to reveal the underlying mechanism in HCC carcinogenesis. We identified a prognostic signature (long intergenic non-protein-coding [LINC] RNA 2691 [LINC02691] and LINC02499) that effectively predicts OS and has protective effects.

We constructed a critical ceRNA network that included four lncRNAs (LINC02691, LINC02499, LINC01354, and NAV2 antisense RNA 4 [NAV2-AS4]), one miRNA (miR-212-3p), and two mRNAs (SEC14-like protein 2 [SEC14L2] and solute carrier family 6 member 1 [SLC6A11]). This study provided a better understanding of the pathogenesis of liver cancer from the perspective of polygenic association, thus offering novel insights into targeted combination therapies.

RESULTS

Study process of transcriptome data

The study flow diagram is shown in Figure 1. We divided the transcriptome data of 371 HCC tissues into Myc\textsuperscript{high} tumor and Myc\textsuperscript{low} tumor groups based on the standard median expression of Myc. We selected DE lncRNAs, miRNAs, and mRNAs from the Myc\textsuperscript{high} tumor group and Myc\textsuperscript{low} tumor group. We considered lncRNA-miRNA to be a true interaction target by miR-code. We explored the miRDB database and TargetScan for miRNA-mRNA target prediction to construct a ceRNA network, followed by analyses of expression and survival.

Clinical analysis of Myc overexpression in HCC

To explore whether Myc affects gene expression in liver cancer, we divided liver cancer patients into Myc\textsuperscript{high} tumor and Myc\textsuperscript{low} tumor groups based on the median values of Myc in this study. The mRNA and protein levels of Myc were presented in various normal organs in the Human Protein Atlas (HPA) database (Figures 2A and S1A). The expression of Myc was high in cancer cell lines (Figure S1B). The region of Myc that is genomically altered in liver cancer was expressed through amplification and was found to be altered in 66 (18%) of 366 patients in HCC (Figures 2B and 2C). Myc expression was higher in HCC tissues (n = 369) compared with in normal liver tissues (n = 160, p < 0.01; Figure 2D). The expression of Myc increased as tumor invasion worsened (Figure 2E). Kaplan-Meier survival analysis showed the prognostic potential of Myc expression. These data suggested that Myc was upregulated in liver cancer samples.

DE RNAs in HCC

To better understand the relationship between DE RNAs and tumorigenesis-associated HCC, we downloaded gene-expression
microarrays from TCGA database to search for DE RNAs. The gene-expression microarrays contained 371 samples (lncRNAs, mRNAs); 367 samples (miRNAs) were defined as Myc\textsuperscript{high} and Myc\textsuperscript{low} based on the median expression levels of Myc.\textsuperscript{45} We analyzed the DE genes between the Myc\textsuperscript{high} and Myc\textsuperscript{low} tumor groups with the standard of p < 0.05 and fold change (FC) \( \geq 1.5 \). In total, we found 589 lncRNAs, 93 miRNAs, and 1,125 mRNAs of DE RNAs among the groups. The upregulated DE RNAs included 222 (38\%) lncRNAs, 65 (70\%) miRNAs, and 696 (62\%) mRNAs. The downregulated DE RNAs included 367 (62\%) lncRNAs, 38 (30\%) miRNAs, and 429 (38\%) mRNAs. In Figure 3, the volcano plots show the distribution of DE RNAs (Figure 3A), and the heatmap describes 15 significant DE RNAs (Figure 3B).

### Functional enrichment analysis of DE mRNAs
To better comprehend the mechanisms involved in HCC, we explored the function of 1,125 DE mRNAs from Gene Ontology
(GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analyses using the Metascape database (Figures 4A and 4B). The most enriched GO terms in cellular component (CC), biological process (BP), and molecular function (MF) were “side of membrane,” “response to toxic substance,” and “organic anion transmembrane transporter activity,” respectively. We found the oxidation-reduction process, peroxisome proliferator-activated receptor (PPAR) signaling, and peroxisome pathway to participate in HCC, according to KEGG pathway enrichment analysis.

**Construction of the ceRNA network and identification of hub RNAs**

We used miRNA target informatic tools to identify DE RNAs in the ceRNA. The ceRNA network was constructed using Cytoscape 3.7 (Figure 5A) and included 19 IncRNAs, 5 miRNAs, and 72 mRNAs. The ceRNA network included 19 IncRNAs (11 downregulated and 8 upregulated) and 5 miRNAs (4 downregulated and 1 upregulated). In addition, 72 mRNAs (58 upregulated and 14 downregulated) and 5 miRNAs (4 downregulated and 1 upregulated) were used to construct

---

**Figure 3. Identification of DE genes**

(A) Volcano maps of DE IncRNAs, miRNAs, and mRNAs between two groups: Mychigh and Myclin in HCC. (B) Heatmap of DE IncRNAs (top), miRNAs (middle), and mRNAs (bottom). Red and blue spots represent significantly upregulated and downregulated RNAs, respectively.
We employed a large ceRNA network based on node-weighting arithmetic to recognize highly interacted hub clustering (Figure 5B). The hub RNAs contained 14 lncRNAs (6 upregulated and 8 downregulated), 5 miRNAs (2 upregulated and 3 downregulated), and 11 mRNAs (9 upregulated and 2 downregulated).

Comprehensive analysis of DE lncRNAs in the ceRNA network

LncRNAs have an impact on the responses of miRNAs and mRNAs, commanding the upstream portion of the ceRNA network. The expression of lncRNAs is associated with OS in cancer patients. Therefore, we analyzed 19 DE lncRNAs of ceRNA in this study and
found differential expression in the Myc\textsuperscript{high} tumor group (n = 185) and Myc\textsuperscript{low} tumor group (n = 186) using the heatmap (Figure 6A).

Among the 19 DE lncRNAs, we screened potential prognosis-associated lncRNAs by univariate and multivariate Cox regression analyses. The final risk score method established that an lncRNA signal was a significant prognostic factor for HCC, as follows: prognostic index

Figure 6. Comprehensive analysis of DE lncRNAs in the ceRNA network
(A) Differential expression of 19 lncRNAs in the ceRNA network by a heatmap. (B) Validation of the two lncRNA signatures by Kaplan-Meier survival and ROC curves based on risk scores. (C) Prediction of lncRNA subcellular localization; a score ranging from 0 and 1 was given. (D) Expression of LINC02691, LINC02499, LINC01354, and NAV2-AS4 was analyzed in the Myc\textsuperscript{high} tumor group (n = 185) and Myc\textsuperscript{low} tumor group (n = 186) in HCC. (E) Survival analysis.

Figure 5. Myc ceRNA network in HCC
(A) Blue rhombus represents downregulated lncRNAs, red ellipses represent upregulated miRNAs, and blue triangles represent downregulated mRNAs; red rhombus represents upregulated lncRNAs, blue ellipses represent downregulated miRNAs, and red triangles represent upregulated mRNAs. (B) The hub 30 genes in the ceRNA network.
compared with the Myclow tumor group (n = 18; Figure 7B). miR-212-3p was highly expressed in the Mychigh tumor group (n = 23), showing that miR-212-3p was a factor that promoted tumor growth. Patients with high miR-212-3p expression had a poor prognosis (Figure 7A). Moreover, the Kaplan-Meier survival curve for four miRNAs (Figure 7A). The expression of SEC14L2 and SLC6A1 was lower in most cancers, except for renal cell clear carcinoma and HCC. The expression of SEC14L2 was significantly lower in cancer tissues compared with normal liver tissues (n = 10) taken from the Gene Expression Omnibus (GEO) database (Figures 8A and 8B). No genetic alterations were found in SEC14L2 and SLC6A1 in 371 liver cancers; however, SEC14L2 was altered in 1 (0.3%) of 366 patients, and SLC6A1 was altered in 5 (1.4%) of 366 patients with liver cancer in the cBioPortal database. In addition, SEC14L2 and SLC6A1 had a lower expression in liver cancer (n = 10) compared with paired-normal tissue samples (n = 10) taken from the Gene Expression Omnibus (GEO) profiles (Figures 8E and 8F).

Expression of SEC14L2 and SLC6A1 in various cancers
We analyzed the transcription and protein levels of SEC14L2 and SLC6A1 in various organ tissues using the HPA database (Figures S4 and S5). To further evaluate SEC14L2 and SLC6A1 expression in human cancer, we used RNA sequencing data to examine SEC14L2 and SLC6A1 expression between tumor and normal tissues. The expression of SEC14L2 was significantly lower in some cancers, including bladder urothelial carcinoma, cholangiocarcinoma, kidney chromophobe, lung adenocarcinoma, and HCC. SLC6A1 expression was lower in most cancers, except renal cell clear carcinoma and HCC (Figure S6). The expression of SEC14L2 and SLC6A1 was low in various cancer cell lines (Figure S7).

Association of immune infiltration with SEC14L2 and SLC6A1 expression in HCC
To explore the role of SEC14L2 and SLC6A1 in immune infiltration in HCC, we evaluated the relationship between differential expression gene and immune cell infiltration by the TIMER platform. A positive correlation between SEC14L2 expression and immune cell infiltration (Cor = 0.172, p = 1.34e-03). The positive correlation of SEC14L2 and SLC6A1 expression was with B cells (Cor = −0.271, p = 3.33e-07), CD8⁺ T cells (Cor = −0.224, p = 3.00e-05), macrophages (Cor = −0.325, p = 7.55e-10), neutrophils (Cor = −0.224, p = 2.64e-05), and dendritic cells (Cor = −0.237, p = 1.02e-05) (Figure 8G). We
researched the correlation between SEC14L2 and SLC6A1 expression and gene biomarkers of immune cells. The results were most strongly correlated with T cells and T cell exhaustion (Table S1). The correlation between SEC14L2 and gene biomarkers of immune cells in tumor and normal samples is shown in Table S2.

**Correlation between methylation and SEC14L2 expression in HCC**

The UALCAN database showed that SEC14L2 is highly methylated in the HCC tissue database (Figure 9A). We analyzed the relationship between SEC14L2 methylation and clinical information using the MEXPRESS database. We found significant methylation of SEC14L2 in various clinical factors, including new tumor events after initial treatment, histological type, gender, tumor stage, and OS. The methylation of SEC14L2 occurred on multiple sites, including cg03673688, cg22352499, and cg23665603 (r = 0.441, 0.447, and 0.389, respectively) (Figure 9B). We described the relationship between the methylation sites (cg03673688, cg22352499, and cg23665603) of SEC14L2 and clinical factors of patients using the MethSurv database (Figure 9C).

Figure 7. Expression and survival analysis of DE miRNAs

(A) Kaplan-Meier survival analysis of miR-212-3p, miR-217, miR-216b-5p, miR-375, and miR-146b-5p in 371 HCC tissues. (B) The miR-212-3p expressed in the Myc\textsuperscript{high} tumor group (n = 184) and Myc\textsuperscript{low} tumor group (n = 183). (C) The miR-212-3p expression in the TNM stage in 371 liver cancer samples. (D) Expression level of miR-212-3p in 371 HCC tissues compared with 50 adjacent normal tissues. (E) In 50 pairs of HCC tissues, miR-212-3p was highly expressed in liver cancer. (F) A schematic diagram of the miR-212-3p of 3' UTR in four lncRNA-binding sites. (G) The high-risk group had high miRNA expression and low lncRNA expression. The low-risk group had low miRNA expression and high lncRNA expression.
Figure 8. Analysis of SEC14L2 and SLC6A1
(A and B) Comparison of SEC14L2 and SLC6A1 expression in neoplasm development and association with prolonged prognosis. (C and D) Verification of SEC14L2 and SLC6A1 expression in normal tissues and HCC tissues in liver. (E and F) Verification of SEC14L2 and SLC6A1 in HCC relative to paired normal tissue samples from GEO profiles. (G) Immune cell infiltration of SEC14L2 and SLC6A1 in HCC using the TIMER database.
Construction of the lncRNA-miRNA-mRNA network

To verify the ceRNA mechanism in HCC, we constructed a lncRNA-miRNA-mRNA network after analysis, which included four lncRNAs, one miRNA, and two mRNAs (Figure 10A). We analyzed the correlation between Myc and these key RNAs (Figure S8A). We found that miR-212-3p is a connecting linker gene that participates in ceRNA pathways, whereas lncRNAs indirectly regulated mRNA expression by preferentially encompassing the miRNA response region. We analyzed LINC02691, LINC02499, LINC01354, and NAV2-AS4 and found that lncRNAs were positively correlated with mRNAs (SEC14L2 and SLC6A1) through the same miRNA, miR-212-3p (Figure 10B). Spearman correlation analysis showed that miR-212-3p was negatively regulated in lncRNAs and mRNAs of HCC (Figure 10C). The correlation among lncRNAs, miRNA, and mRNAs is shown (Figure S8B).

Relationship between key RNAs and clinical features

We analyzed the relationship between key RNAs (lncRNAs, miRNAs, and mRNAs) and clinical information, including age, gender, body mass index (BMI), race, TNM stage, metastatic lymph node metastasis, diameter, and prior malignancy (Table S3). SEC14L2 and SLC6A1 had significant differences regarding BMI, tumor stage, and diameter (p < 0.05). TNM, diameter, lymph node metastasis, distant metastasis, and prior malignancy were significantly related to HCC prognosis (p < 0.05). LINC02499 was the lncRNA most significantly correlated with the clinical factors (Table S4).

DISCUSSION

HCC is the fifth–most common cancer, with the second–highest mortality rate worldwide.50–52 The incidences of liver cancer and mortality have been increasing in the past few years, particularly in Asia. Traditional surgery is no longer the preferred treatment for HCC, so new therapeutic directions need to be explored.53–55 Molecular targeted therapy is an important topic for the treatment of HCC; however, the precise molecular mechanisms remain unknown. The development of high-throughput transcriptome sequencing technologies and the ceRNA hypothesis have been proposed, in which lncRNAs and mRNAs interact with each other through shared miRNAs.56–59 Recently, studies have been conducted to construct a ceRNA network between HCC tissues and adjacent nontumor liver tissues.60 The molecular mechanisms of ceRNA associated with Myc remain unclear in HCC.

Myc is an oncogene that is dysregulated in >50% of tumors.51–64 In this study, we first analyzed the large cohort of associated Myc transcriptome profiling with HCC using TCGA database. We selected DE lncRNAs, miRNAs, and mRNAs by comparing the Myc-high tumor tissues with Myc-low tumor tissues based on Myc expression, and we constructed a ceRNA network that identified 1125 DE mRNAs, 589 DE lncRNAs, and 93 DE miRNAs.

Numerous studies have reported that lncRNAs play a critical role in different cancers.65–68 lncRNA MIR503HG inhibits cell proliferation and promotes apoptosis in triple-negative breast cancer (TNBC) cells via the miR-224-5p/HOXA9 axis.69 IncTPF (long non-coding idiopathic pulmonary fibrosis) promotes pulmonary fibrosis by targeting hnRNP-L, depending on its host gene ITGBL1.70 lncRNA LINC00858 functions through the miR-153-3p/Rab13 axis, which promotes cell proliferation and infiltration in HCC.71 Therefore, we analyzed 19 DE lncRNAs in the ceRNA network by multidirectional analysis and calculated risk scores through univariate and multivariate Cox regression analyses. Our results showed that LINC02691, LINC02499, LINC01354, and NAV2-AS4 were significant in predicting OS in HCC, especially LINC02691 and LINC02499. Therefore, the prognostic signature of lncRNAs might act as a prognostic and diagnostic marker for HCC.

Among the prognostic signature lncRNAs, the oncogenic function of LINC01354 has been studied in a variety of cancers—in particular, in colorectal carcinoma, gastric carcinoma, lung carcinoma, and neck squamous cell carcinoma—in which LINC01354 accelerates the proliferation response, migration, and infiltration of cancer cells.72–76 This study found suppression of LINC01354 expression in Myc-high tumor tissues compared with Myc-low tumor tissues. LINC02499 is highly expressed in HCC and inhibits cell-proliferation capacity, migration, and immune cell infiltration in HCC77 and was first analyzed in the associated Myc ceRNA network. Therefore, we successfully analyzed a series of lncRNAs with functions in the associated Myc ceRNA network. lncRNAs, including LINC02691 and LINC01354, were found to be strongly negatively regulated in HCC through the miR-212-3p/SEC14L2 axis.

miRNA connects lncRNA and mRNA in various cancers, including HCC, acting as a bridge for the ceRNA network.78–81 miR-96 reduces HCC migration, functioning as a therapeutic target in this disease.82 miR-221-3p promotes HCC by downregulating the expression of O6-methylguanine-DNA methyltransferase.83 In this study, we analyzed five miRNAs in the ceRNA network, and only miR-212-3p overexpression was correlated with poor OS. The ceRNA network illustrates that has-miR-212-3p may promote cancer cell migration and invasion.

In this comprehensive study, we screened a ceRNA axis, including the downstream coding gene of SEC14L2 and SLC6A1. We analyzed downstream genes through GO enrichment analysis and KEGG pathway analysis. We found that the GO terms of the dysregulated mRNAs (SEC14L2 and SLC6A1) in HCC could be classified into MF, CC, and BP to further explain the pathways involved. In terms of MF, organic anion transmembrane transporter activity and vitamin E binding suggested that HCC may be a multigene-related disease. During this study, the PPAR signaling pathway was the
most important KEGG pathway in the Metascape database, which also has been found in several other cancer types.88–94

SEC14L2-like phosphatidylinositol transfer proteins SEC14L3/SEC14L2 mediated Wnt/Ca²⁺ signaling by acting as GTPase proteins.95 SLC6A1 is overexpressed in prostate cancer and is associated with drug resistance and a poor prognosis96 but is downregulated in HCC.97 SEC14L2 and SLC6A1 mRNAs serve as novel transcriptional targets related to Myc and suppress HCC progression and thus may be a promising target for the treatment of Myc-driven HCC. Our

Figure 10. Correlation of linear regression analysis between DE IncRNAs and mRNAs
(A) The identified IncRNA-miRNA-mRNA axis is integrated into a circuit map. (B) Relationship of IncRNAs and mRNAs. (C) Correlation between hsa-miR-212-3p and IncRNAs and mRNAs.

www.moleculartherapy.org
results show that Kaplan-Meier survival analysis of ceRNA-correlated genes demonstrated that 19 of the 72 mRNAs had a statistically significant impact on prognosis. Many genes significantly influenced the OS of HCC patients; however, only SEC14L2 and SLC6A1 participated in establishing a complete endogenous regulation network.

This study had some limitations. Because of the lack of other HCC-associated samples, we did not perform further in vitro and in vivo experiments on clinical samples. Additionally, some exploratory experiments remain necessary to identify the functions of the unreported RNAs (lncRNAs, miRNAs, and mRNAs) in this study.

In summary, we introduced a Myc-ceRNA network from genome-wide transcriptome data by various bioinformatics analyses to provide a comprehensive analysis. This approach identified some key RNAs that were significantly associated with prognosis and that provide potential prognostic and diagnostic biomarkers for HCC.

MATERIALS AND METHODS
Data processing and analysis of DE proteins in HCC
We downloaded the transcriptome sequencing data of 421 lncRNAs and mRNAs and 417 miRNAs from 50 patients with HCC through TCGA data portal (https://portal.gdc.cancer.gov/). The 421 RNAs (417 miRNAs) were from 371 tumor tissues and 50 normal tissues. For external validation, we used 10 liver cancer samples and paracancer samples as validation sets from GEO profiles.

Functional enrichment analysis of DE mRNA
Metascape (http://metascape.org) is an informatics functional annotation tool that integrates many dominating databases to comprehensively analyze genes. We analyzed the enriched function of DE mRNAs using the GO and KEGG pathways in the Metascape website, with the restrictions of p < 0.01, a minimum count of 3, and an enrichment factor of >1.5. We obtained DE mRNAs, lncRNAs, and miRNAs by comparing the Myc\textsuperscript{high} and Myc\textsuperscript{low} tumor groups, with thresholds of p < 0.05 and a FC of ≥1.5.\tiny{18}

Construction of the ceRNA network and identification of hub RNAs
Cytoscape is a visualized software for network data, which provides users with a more picturesque biological process network.\tiny{100} We used lncRNA as a true interaction miRNA target explored by miRcode (http://www.mircode.org/) and used miRDB (http://mirdb.org/) and TargetScan (http://www.targetscan.org/) for miRNA-mRNA target gene prediction. We employed these predicted relationship pairs to construct an endogenous competitive network based on the predicted miRNA expression relationship, which was visualized by Cytoscape version 3.7.0 software. We calculated the density connected degree of gene nodes in a ceRNA network, and we employed the top 30 genes with the highest confidence scores as hub genes.

Prediction of the subcellular location of IncRNAs
The subcellular localization of IncRNA has an impact on its function. Therefore, we predicted the sequences of the significant IncRNA biomarkers obtained from the LNCipedia (https://lncipedia.org/). Then we used the sequences of IncRNAs to predict the subcellular localization in IncLocator (http://www.csbio.sjtu.edu.cn/biocinf/IncLocator/).

A score for each potential subcellular localization of IncRNA included the cytoplasm, nucleus, ribosome, cytosol, and exosome. We analyzed the final results using GraphPad Prism 8.3.

cBioPortal database analysis
The cBioPortal (http://www.cbioportal.org/) is an online source application, which provides information on somatic mutations, altered copy number, and mRNA expression, used to visualize cancer genomics data.\tiny{101–104} In this study, we used cBioPortal to show SEC14L2 and SLC6A1 genetic changes in HCC.

Immune infiltrate analysis of SEC14L2 and SLC6A1
The TIMER database (http://cistrome.org/TIMER/) is a compositive resource that provides systematic analysis of the abundance of immune cells in the infiltrate of various cancers.\tiny{105} We analyzed the expression of SEC14L2 and SLC6A1 in a variety of cancers, as well the correlation between their expression levels and the abundance of the immune cells. In addition, we evaluated the correlation of SEC14L2 and SLC6A1 expression with the biomarkers of immune cells. Gene Expression Profiling Interactive Analysis (http://gepia.cancer-pku.cn/) is an open database that contains tumor and normal samples from TCGA and Genotype-Tissue Expression databases. We used this database to confirm the correlation between DE mRNA and markers of immune cells in tumor and normal samples.

Methylation analysis of SEC14L2
The UALCAN database (http://ualcan.path.uab.edu/) is a data-mining platform, in which the methylation of DE mRNA in tumors can be queried.\tiny{106–108} In this study, we used the UALCAN database to analyze SEC14L2 methylation in liver cancer tissues and paracancerous tissues. The MethSurv database (https://biit.cs.ut.ee/methsurv/) is an open website used to obtain CpG methylation data,\tiny{109} and it contains significant information about a single CpG. We screened DE mRNA using the MethSurv database and then verified the most important methylated site associated with HCC patient outcomes. MEXPRESS (https://mexpress.be/) is a data-visualization tool used to visualize TCGA expression and the relationship between methylation expression and clinical information.\tiny{110–112}

The HPA database
The HPA database (https://www.proteinatlas.org/) was initiated in 2003 to map all human proteins in cells, tissues, and organs. In this study, we assembled the protein and RNA expression levels of SEC14L2 and SLC6A1 in various cancers tissues using this database.\tiny{113–115}
Cancer Cell Line Encyclopedia (CCLE) database analysis

CCLE (https://portals.broadinstitute.org/ccle) is an open-source website, which consists of a large amount of human cancer cell lines, including genomic data, gene expression, copy number, and abundant sequencing data. We analyzed the expression of SEC14L2 and SLG6A1 in HCC cell lines using the CCLE database.

Relationship between key RNAs and clinical features

We selected key RNAs according to the previously described analyses, which are presented as the median ± standard deviation. We conducted nonparametric tests to determine whether the expression of RNAs was correlated with the following clinical features: age (≥60 years versus <60 years), gender (female versus male), tumor stage (I + II versus III + IV); BMI (≤18.5, 18.6–23.9, 24–27.9, and ≥28), race (Asian versus White), diameter (≥5 cm versus <5 cm), lymph node and distant metastases (positive versus negative), and prior malignancy (yes versus no). p < 0.05 was used as the cutoff value.

Statistical analysis

We compared differential expression between the Myc<sup>hi</sup> and Myc<sup>lo</sup> tumor groups using the nonparametric test. The correlation and survival analysis of the relative expression of the important lncRNA-miRNA-mRNA network were processed using GraphPad Prism 8.3. We explored potential lncRNAs using Cox regression analyses in the R package version 4.0. We completed all differential expression analyses using nonparametric tests. p < 0.05 was considered statistically significant.

SUPPLEMENTAL INFORMATION

Supplemental information can be found online at https://doi.org/10.1016/j.omtn.2021.04.019.

ACKNOWLEDGMENTS

We would like to thank Prof. Li-Ping Gu for data analysis and critical discussion of the manuscript. This study was supported partly by grants from the National Natural Science Foundation of China (81972214, 81772932, 81472202, and 81302065); Shanghai Natural Science Foundation (20ZR1472400); Natural Science Foundation of Hunan Province of China (2020WK2020); Construction of Clinical Medical Centre for Tumor Biological Samples in Nantong (HS2016004); Jiangsu 333 Program (BRA2017205); and Wu Jieping Medical Foundation (320.6750.14326).

AUTHOR CONTRIBUTIONS

Y.-S.M., D.F., and W.-J.Z. designed the study and contributed to study materials and consumables. D.-D.Z., Y.S., J.-B.L., X.-L.Y., R.X., H.-M.W., P.-Y.W., C.-Y.J., Y.-S.M., and D.F. collected data. D.-D.Z., Y.S., and Y.-S.M. performed the statistical analyses and interpreted the data. D.-D.Z., Y.-S.M., and D.F. wrote the manuscript. All authors contributed to the final version of the manuscript and approved the final manuscript.

DECLARATION OF INTERESTS

The authors declare no competing interests.

REFERENCES

1. Liu, Z., Lin, Y., Zhang, J., Zhang, Y., Li, Y., Liu, Z., Li, Q., Luo, M., Liang, R., and Ye, J. (2019). Molecular targeted and immune checkpoint therapy for advanced hepatocellular carcinoma. J. Exp. Clin. Cancer Res. 38, 447.
2. Ma, Y.S., Huang, T., Zhong, X.M., Zhang, H.W., Cong, X.L., Xu, H., Lu, G.X., Yu, F., Xue, S.B., Ju, Z.W., and Fu, D. (2018). Proteogenomic characterization and comprehensive integrative genomic analysis of human colorectal cancer liver metastasis. Mol. Cancer 17, 139.
3. Zhen, L., Zhao, Q., Li, J., Deng, S., Xu, Z., Zhang, L., Zhang, Y., Fan, H., Chen, X., Liu, Z., et al. (2020). miR-301a-PTEN-AKT Signaling Induces Cardiomyocyte Proliferation and Promotes Cardiac Repair Post-MI. Mol. Ther. Nucleic Acids 22, 251–262.
4. Zhang, X., Wang, D., Liu, B., Jin, X., Wang, X., Pan, J., Tu, W., and Shao, Y. (2020). IMP3 accelerates the progression of prostate cancer through inhibiting PTEN expression in a SMURF1-dependent way. J. Exp. Clin. Cancer Res. 39, 190.
5. Sun, L.L., Xiao, L., Du, X.L., Hong, L., Li, C.L., Jiao, J., Li, W.D., and Li, X.Q. (2019). MiR-205 promotes endothelial progenitor cell angiogenesis and deep vein thrombosis recanalization and resolution by targeting PTEN to regulate Akt/autophagy pathway and MMP2 expression. J. Cell. Mol. Med. 23, 8493–4504.
6. Ho, D.W., Tsui, Y.M., Sze, K.M., Chan, L.K., Cheung, T.T., Lee, E., Sham, P.C., Tsui, S.K., Lee, T.K., and Ng, I.O. (2019). Single-cell transcriptomics reveals the landscape of intra-tumoral heterogeneity and stenosis-related subpopulations in liver cancer. Cancer Lett. 459, 176–185.
7. Bray, F., Ferlay, J., Soerjomataram, I., Siegel, R.L., Torre, L.A., and Jemal, A. (2018). Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J. Clin. 68, 39–424.
8. Yu, F., Chen, B., Dong, P., and Zheng, J. (2020). HOTAIR Epigenetically Modulates PTEN Expression via MicroRNA-29b: A Novel Mechanism in Regulation of Liver Fibrosis. Mol. Ther. 28, 2703.
9. Hutchings, C., Phillips, J.A., and Djamgoz, M.B.A. (2020). Nerve input to tumours: Pathophysiological consequences of a dynamic relationship. Biochim. Biophys. Acta. Rev. Cancer 1874, 188411.
10. Siemers, N.O., Holloway, J.L., Chang, H., Chasalow, S.D., Ross-MacDonald, P.B., Voliva, C.F., and Szustakowski, J.D. (2017). Genome-wide association analysis identifies genetic correlates of immune infiltrates in solid tumors. PLoS ONE 12, e0179726.
11. Hu, J., Dong, Y., Ding, L., Dong, Y., Wu, Z., Wang, W., Shen, M., and Duan, Y. (2019). Local delivery of arsenic trioxide nanoparticles for hepatocellular carcinoma treatment. Signal Transduct. Target. Ther. 4, 28.
12. Zhao, Y., Zhang, Y.N., Wang, K.T., and Chen, L. (2020). Lenvatinib for hepatocellular carcinoma: From preclinical mechanisms to anti-cancer therapy. Biochim. Biophys. Acta Rev. Cancer 1874, 188391.
13. Jasirwan, C.O.M., Hasan, I., Sulaiman, A.S., Lesmana, C.R.A., Kurniawan, J., Kalista, K.F., Nababan, S.H., and Gani, R.A. (2020). Risk factors of mortality in the patients with hepatocellular carcinoma: A multicenter study in Indonesia. Curr. Probl. Cancer 44, 100480.
14. Wang, Z., Yu, W., Qiang, Y., Xu, L., Ma, F., Ding, P., Shi, L., Chang, W., Mei, Y., and Ma, X. (2020). Luki3-PV Inhibits Hepatocellular Carcinoma Progression by Downregulating HDAC2 Expression. Mol. Ther. Oncolytics 17, 547–561.
15. Wei, L., Wang, X., Lv, L., Liu, J., Xing, H., Song, Y., Xie, M., Lei, T., Zhang, N., and Yang, M. (2019). The emerging role of microRNAs and long noncoding RNAs in drug resistance of hepatocellular carcinoma. Mol. Cancer 18, 147.
16. Li, Y., Li, G., Tao, T., Kang, X., Liu, C., Zhang, X., Wang, C., Li, C., and Guo, X. (2019). The α-opioid receptor (MOR) promotes tumor initiation in hepatocellular carcinoma. Cancer Lett. 453, 1–9.
17. Kim, Y., Jo, M., Schmidt, J., Luo, P., Prakash, T.P., Zhou, T., Klein, S., Xiao, X., Post, N., Yin, Z., and MacLeod, A.R. (2019). Enhanced Potency of GalNAc-Conjugated Antisense Oligonucleotides in Hepatocellular Cancer Models. Mol. Ther. 27, 1547–1557.
by b-AP1 as a potential therapy for tumors with p53 deficiency. Signal Transduct. Target. Ther. 5, 30.

55. Xu, H., Chen, G.F., Ma, Y.S., Zhang, H.W., Zhou, Y., Liu, G.H., Chen, D.Y., Ping, J., Liu, Y.H., Mou, X., and Fu, D. (2020). Hepatic Proteome Changes and Sirt1/AMPK Signaling Activation by Oxymatrine Treatment in Rats With Non-alcoholic Steatosis. Front. Pharmacol. 11, 216.

56. Zhao, B., Ke, K., Wang, Y., Wang, F., Shi, Y., Zheng, X., Yang, X., Liu, X., and Liu, J. (2020). HIF-1z and HDAC1 mediated regulation of FAM19A2-miR22a signaling contributes to hypoxia induced HCC metastasis. Signal Transduct. Target. Ther. 5, 118.

57. Xiao, Y., Najeeb, R.M., Ma, D., Yang, K., Zhong, Q., and Liu, Q. (2019). Upregulation of CENPM promotes hepatocarcinogenesis through multiple mechanisms. J. Exp. Clin. Cancer Res. 38, 458.

58. Tam, B.Y., Chiu, K., Chung, H., Bossard, C., Nguyen, J.D., Creger, E., Eastman, B.W., Mak, C.C., Ibanez, M., Ghiats, A., et al. (2020). The CLK inhibitor SM08502 induces anti-tumor activity and reduces Wnt pathway gene expression in gastrointestinal cancer models. Cancer Lett. 473, 186–197.

59. Li, M., Shao, J., Guo, Z., Jin, C., Wang, L., Wang, F., Jia, Y., Zhu, Z., Zhang, Z., Zhang, F., et al. (2020). Novel mitochondrial-targeting copper(II) complex induces HK2 malfunction and inhibits glycosylation via Drp1-mediating mitophagy in HCC. J. Cell. Mol. Med. 24, 3091–3107.

60. Chen, Y., Zhao, H., Li, H., Feng, X., Tang, H., Qiu, C., Zhang, J., and Fu, B. (2020). LINC01234/MicroRNA-31-5p/AEG3 Axis Mediates the Proliferation and Chemoresistance of Hepatocellular Carcinoma Cells. Mol. Ther. Nucleic Acids 19, 168–178.

61. Shao, S., Song, X., Jiang, W., Chen, Y., Niu, Z., Wu, G., and Jiang, J. (2019). MicroRNA-621 acts as a tumor radiosensitizer by directly targeting SETDB1 in hepatocellular carcinoma. Mol. Ther. 27, 355–364.

62. Zeng, C., Liu, S., Liu, S., Yu, X., Lai, J., Wu, Y., Chen, S., Wang, L., Yu, Z., Luo, G., and Li, Y. (2018). The c-Myc-regulated IncRNA NEAT1 and paracasplices modulate imatinib-induced apoptosis in CML cells. Mol. Cancer 17, 130.

63. van den Ende, T., van den Boorn, H.G., Hoonhout, N.M., van Etten-Jamaludin, F.S., Shao, Y., Song, X., Jiang, W., Chen, Y., Ning, Z., Gu, W., and Jiang, J. (2019). Identification and validation of methylation-driven genes prognostic signature for recurrence of laryngeal squamous cell carcinoma by integrated bioinformatics analysis. Cancer Cell Int. 20, 472.

64. Jiang, Y., and Luo, Y. (2020). LINC01354 Promotes Osteosarcoma Cell Invasion by Up-regulating Integrin ß1. Arch. Med. Res. 51, 115–123.

65. Li, R., Yang, Y.E., Yin, Y.H., Zhang, M.Y., Li, H., and Qu, Y.Q. (2019). Methylation and transcriptome analysis reveal lung adenocarcinoma-specific diagnostic biomarkers. J. Transl. Med. 17, 324.

66. Li, J., He, M., Xu, W., and Huang, S. (2019). LINC01354 interacting with hnRNPD contributes to the proliferation and metastasis in colorectal cancer through activating Wnt/beta-catenin signaling pathway. J. Exp. Clin. Cancer Res. 38, 161.

67. Yang, G., Yang, C., She, Y., Shen, Z., and Gao, P. (2019). LINC01354 enhances the proliferation and invasion of lung cancer cells by regulating miR-340-5p/ATF1 signaling pathway. Artif. Cells Nanomed. Biotechnol. 47, 3737–3744.

68. Ma, X., Mo, M., Tan, H.J.J., Tan, C., Zeng, X., Zhang, G., Huang, D., Liang, J., Liu, J., and Qiu, X. (2020). LINC02499, a novel liver-specific long non-coding RNA with potential diagnostic and prognostic value, inhibits hepatocellular carcinoma cell proliferation, migration, and invasion. Hepatol. Res. 50, 726–740.

69. Hu, J., Chen, Z., Bao, L., Zhou, L., Hou, Y., Liu, L., Xiong, M., Zhang, Y., Wang, B., Tao, Z., and Chan, K. (2020). Single-Cell Transcriptome Analysis Reveals Intratumoral Heterogeneity in cRCC, which Results in Different Clinical Outcomes. Mol. Ther. 28, 1658–1672.

70. Kawamura, E., Maruyama, M., Abe, J., Sudo, A., Takeda, A., Takeda, S., Yokota, T., Kinugawa, S., Harashima, H., and Yamada, Y. (2020). Validation of Gene Therapy for Mutant Mitochondria by Delivering Mitochondrial RNA Using a MITO-Porter. Mol. Ther. Nucleic Acids 20, 687–698.

71. Easley, N.B., Nace, R.A., Russell, S.J., and Schulze, A.J. (2020). Oncolytic Activity of Targeted Picornaviruses Formulated as Synthetic Infectious RNA. Mol. Ther. Oncolytics 17, 484–495.

72. Gupta, S.C., Awashtee, N., Rai, V., Chava, S., Gunda, V., and Chapallangudi, K.B. (2020). Non-long coding RNAs and nuclear factor-kB crosstalk in cancer and other human diseases. Biochem. Biophys. Acta Rev. Cancer 1873, 188316.

73. Iwai, N., Yasui, K., Tomie, A., Teraski, K., Kitaichi, T., Suda, T., Yamada, N., Doihi, O., Seko, Y., et al. (2018). Oncogenic miR-96-5p inhibits apoptosis by targeting the caspase-9 gene in hepatocellular carcinoma. Int. J. Oncol. 53, 237–245.

74. Chen, Z., Xiang, B., Qi, L., Zhou, S., and Li, H. (2020). miR-221–23b promotes hepatocellular carcinogenesis by downregulating O6-methylguanine-DNA methyltransferase. Cancer Biol. Ther. 21, 915–926.

75. Yang, J., Cui, R., and Liu, Y. (2020). MicroRNA-212-3p inhibits paclitaxel resistance through regulating epithelial-mesenchymal transition, migration and invasion by targeting ZEB2 in human hepatocellular carcinoma. Oncol. Lett. 20, 23.

76. Ghosh, S., Bhownik, S., Majumdar, S., Goswami, A., Chakraborty, J., Gupta, S., Aggarwal, S., Ray, S., Chatterjee, R., Bhattacharyya, S., et al. (2020). The exosome encapsulated microRNAs as circulating diagnostic marker for hepatocellular carcinoma with low alpha fetoprotein. Int. J. Cancer 147, 2934–2947.

77. Liu, Y., Cao, Y., Dai, W., Wu, L., Zhao, P., and Liu, X.G. (2020). Aberrant expression of targeting ZEB2 in human hepatocellular carcinoma. Oncol. Lett. 20, 2934–2947.

78. Wang, X., Liao, X., Huang, K., Zeng, X., Liu, Z., Yu, T., Yang, C., Yu, L., and Lu, G. (2020). lncITPF Promotes Pulmonary Fibrosis by Targeting hnRNP-L Depending on Targeted Picornaviruses Formulated as Synthetic Infectious RNA. Mol. Ther. Oncolytics 17, 484–495.

79. Wang, X., Dohi, O., Seko, Y., et al. (2018). Oncogenic miR-96-5p inhibits apoptosis by targeting the caspase-9 gene in hepatocellular carcinoma. Int. J. Oncol. 53, 237–245.

80. Chen, Z., Xiang, B., Qi, L., Zhou, S., and Li, H. (2020). miR-221–23b promotes hepatocellular carcinogenesis by downregulating O6-methylguanine-DNA methyltransferase. Cancer Biol. Ther. 21, 915–926.

81. Yang, J., Cui, R., and Liu, Y. (2020). MicroRNA-212-3p inhibits paclitaxel resistance through regulating epithelial-mesenchymal transition, migration and invasion by targeting ZEB2 in human hepatocellular carcinoma. Oncol. Lett. 20, 23.

82. Ghosh, S., Bhownik, S., Majumdar, S., Goswami, A., Chakraborty, J., Gupta, S., Aggarwal, S., Ray, S., Chatterjee, R., Bhattacharyya, S., et al. (2020). The exosome encapsulated microRNAs as circulating diagnostic marker for hepatocellular carcinoma with low alpha fetoprotein. Int. J. Cancer 147, 2934–2947.

83. Liu, Y., Cao, Y., Dai, W., Wu, L., Zhao, P., and Liu, X.G. (2020). Aberrant expression of targeting ZEB2 in human hepatocellular carcinoma. Oncol. Lett. 20, 2934–2947.
100. Huang, Z., Yu, H., Du, G., Han, L., Huang, X., Wu, D., Han, X., Xia, Y., Wang, X., and Lu, C. (2021). Enhancer RNA lnc-CES1-1 inhibits decidual cell migration by interacting with RNA-binding protein FUS and activating PPARγ in URPL. Mol. Ther. Nucleic Acids 24, 104–112.

91. Zeng, J., Tang, Z., Zhang, Y., Tong, X., Dou, J., Gao, L., Ding, S., and Lu, J. (2021). Ozonated autolymphopheresis elevates PPAR-γ expression in CD4+ T cells and serum HDL-C levels, a potential immunomodulatory mechanism for treatment of psoriasis. Am. J. Transl. Res. 13, 349–359.

92. Yu, Q., Cheng, P., Wu, J., and Guo, C. (2021). PPARγ/NF-κB and TGF-β1/Smad pathway are involved in the anti-fibrotic effects of levo-tetracydralmanilne on liver fibrosis. J. Cell. Mol. Med. 25, 1645–1660.

93. Chen, H.X., Li, M.Y., Jiang, Y.Y., Hou, H.T., Wang, J., Liu, X.C., Yang, Q., and He, G.W. (2020). Role of the PPAR pathway in atrial fibration associated with heart valve disease: transcriptomics and proteomics in human atrial tissue. Signal Transduct. Target. Ther. 5, 4.

94. Miao, Y., Zheng, Y., Geng, Y., Yang, L., Cao, N., Dai, Y., and Wei, Z. (2021). The role of GLS1-mediated glutaminolysis/2-HG/H3K4m3 and GSH/ROS signals in Th17 responses counteracted by PPARγ agonists. Theranostics 11(4), 4531–4548.

95. Li, Z., Lou, Y., Tian, G., Wu, J., Lu, A., Chen, J., Xu, B., Shi, J., and Yang, J. (2019). Discovering master regulators in hepatocellular carcinoma: one novel MR, SEC14L2 inhibits cancer cells. Aging (Albany NY) 11(8), 12375–12411.

96. Chen, C., Cai, Z., Zhuo, Y., Xi, M., Lin, Z., Jiang, F., Liu, Z., Wan, Y., Zheng, Y., Li, J., et al. (2020). Overexpression of SLC6A1 associates with drug resistance and poor prognosis in prostate cancer. BMC Cancer 20, 288.

97. Kudo, M., Han, K.H., Ye, S.L., Zhou, J., Huang, Y.H., Lin, S.M., Wang, C.K., Ikeda, M., Chen, S.L., Chou, S.P., et al. (2020). A Changing Paradigm for the Treatment of Intermediate-Stage Hepatocellular Carcinoma: Asia-Pacific Primary Liver Cancer Expert Consensus Statements. Liver Cancer 9, 245–260.

98. Gagan, J., and Van Allen, E.M. (2015). Next-generation sequencing to guide cancer therapy. Genome Med. 7, 80.

99. Li, S., Cui, Z., Gu, J., Wang, Y., Yang, S., and Chen, J. (2021). Effect of porcine corneal stromal extract on keratocytes from SMILE-derived lenticules. J. Cell. Mol. Med. 25, 1207–1220.

100. Shannon, P., Markiel, A., Ozier, O., Baliga, N.S., Wang, J.T., Ramage, D., Amin, N., Schwikowski, B., and Ideker, T. (2003). Cytoscape: a software environment for integrated models of biomolecular interaction networks. Genome Res. 13, 2498–2504.

101. Cerami, E., Gao, J., Dogrusoz, U., Gross, B.E., Sumer, S.O., Aksoy, B.A., Jacobsen, A., Byrne, C.J., Heuer, M.L., Larsson, E., et al. (2012). The cBio cancer genomics portal: an open platform for exploring multidimensional cancer genomics data. Cancer Discov. 2, 401–404.

102. Gao, J., Aksoy, B.A., Dogrusoz, U., Dresdner, G., Gross, B., Sumer, S.O., Sun, Y., Jacobsen, A., Sinha, R., Larsson, E., et al. (2013). Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. Sci. Signal. 6, p11.