Risk Scoring System based on IncRNA Expression for Predicting Survival in Hepatocellular Carcinoma with Cirrhosis

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Abstract

Objective: This study aims to explore the roles of long non-coding RNAs (lncRNAs) for predicting survival in hepatocellular carcinoma (HCC) patients with cirrhosis. Methods: A set of lncRNAs differentially expressed between HCC patients with or without cirrhosis was identified using expression profiles of The Cancer Genome Atlas database, and these lncRNAs were screened for their risk scoring system to predict recurrence-free survival (RFS) or overall survival (OS). Predictive ability of risk scoring systems was confirmed using uni- and multivariate Cox analyses while adjusting for clinical features. Predictive lncRNAs were analyzed by functional enrichment analysis. Results: Our screen identified 22 lncRNAs that were upregulated in the presence of cirrhosis and 59 that were downregulated. To predict OS of HCC patients with cirrhosis, a risk scoring system was developed with four lncRNAs (LINC02086, LINC00880, LINC01549 and AC136475.3); to predict RFS in these patients, the risk scoring system contained five lncRNAs (SH3RF3-AS1, AC104117.3, AC136475.3, LINC00239 and MRPL23-AS1). All risk scoring systems were associated with an area under the receiver operating characteristic curve > 0.7. Based on uni- and multivariate Cox analyses, the risk scoring system could serve as a significant independent predictor for OS in HCC patients with cirrhosis. Functional enrichment analysis suggested that the lncRNAs in the risk scoring systems are involved primarily in the pathway of Wnt signal and cytokine-cytokine receptor interaction. Conclusion: Risk scoring systems based on lncRNAs can effectively predict OS of HCC patients with cirrhosis. The system should be further developed and validated in larger, preferably multi-site patient populations.

Keywords: Hepatocellular carcinoma- cirrhosis- long non-coding RNA- survival

Introduction

Liver cancer remains the sixth most frequent cancer globally, with approximately 841,000 new diagnosed cases and 782,000 deaths annually. Hepatocellular carcinoma (HCC) is the most frequent common liver cancer, accounting for 75–85% of liver cancers (Bray et al., 2018; Forner et al., 2018; Kulik and El-Serag, 2019). Despite significant improvements in diagnosing and treating HCC, its heterogeneity means that it continues to be associated with relatively low rates of recurrence-free survival (RFS) and overall survival (OS) (Bruix et al., 2014; Fong and Tanabe, 2014). Currently no biomarkers have proven effective for predicting prognosis, reflecting the complicated nature of the disease. Further efforts are needed to identify biomarkers that can predict prognosis and guide treatment.

Long non-coding RNAs (lncRNAs), located in the cytoplasm and nucleus of eukaryotic cells, are non-coding RNAs longer than 200 nt (Ponting et al., 2009). These molecules play critical roles in the progression and occurrence of malignant cancers (Huarte, 2015; Schmitt and Chang, 2016; Huang et al., 2018; Wu et al., 2018a). For example, lncRNA-TUBB2A and KRTAP5-AS1 compete with endogenous RNAs to modulate the function of claudin-4, affecting prognosis of patients with gastric cancer (Song et al., 2017). The lncRNA Inc-EGFR stimulates T-regulatory cell differentiation, thereby promoting HCC immune evasion (Jiang et al., 2017), while IncRNA-6195 inhibits α-enolase activity and thus HCC progression (Yu et al., 2018).

An important risk factor for developing HCC is hepatic cirrhosis, which is characterized by the appearance of regenerative nodules surrounded by fibrous bands. Cirrhosis occurs secondary to chronic liver injury, and it can lead to portal hypertension or HCC(Schuppan and Afdhal, 2008). Since the clinical characteristics and prognosis of HCC patients can differ depending on

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whether cirrhosis is present (Grazi et al., 2003; Gassmann et al., 2010; Wang et al., 2013; Techathuvanan et al., 2015), we wanted to examine whether some lncRNAs are differentially expressed in the presence or absence of cirrhosis, and whether these might influence risk of recurrence or death. Using expression profile data in The Cancer Genome Atlas (TCGA) database, we developed a risk scoring system based on lncRNA levels and showed that it could predict survival of HCC patients with cirrhosis.

Materials and Methods

Datasets on HCC patients
Expression profiles of lncRNAs and mRNAs from HCC patients, together with the corresponding clinical information, were taken from TCGA (version 09-14-2017 for HCC) via the UCSC Xena server (https://xenabrowser.net/datapages/). Patient data were included in the present study if their HCC was confirmed by histology, complete lncRNA and mRNA expression profiles were available, cirrhosis status was known, and sufficient data were available to determine OS and RFS. After applying these criteria, 77 HCC patients with cirrhosis and 130 without cirrhosis were selected (Table 1). This study was prepared in accordance with TCGA guidelines (https://cancergenome.nih.gov/publications/publication-guidelines). No ethics approval was required for this study since the data came from TCGA.

Identification of lncRNAs differentially expressed between HCC patients with or without cirrhosis
After removal of lncRNAs showing zero expression in more than 50% of patients, the “edgeR” package in R (Robinson et al., 2010) was used to identify lncRNAs differentially expressed between patients with or without cirrhosis, defined as those associated with \[\log2\text{fold change} (\log2FC) > 1\] and false discovery rate (FDR) < 0.05. Volcano and cluster heat maps were created by “gplots” and “heatmap” packages in R.

LncRNA-based risk scoring systems
Firstly, the normalized expression values of multiple samples of the same patient were averaged. Then, univariate Cox analysis was performed to screen differentially expressed lncRNAs for a significant relationship with OS or RFS. Those lncRNAs associated with \(p < 0.05\) were included in subsequent multivariate Cox regression, and the best model was selected using the method of backward stepwise. A risk scoring system was defined using a linear combination of the lncRNA expression levels, each multiplied by a regression coefficient \(\beta\):

\[
\text{Risk score} = (\beta_1 \cdot \text{lncRNA1 level}) + (\beta_2 \cdot \text{lncRNA2 level}) + (\beta_3 \cdot \text{lncRNA3 level}) + (\beta_4 \cdot \ldots \text{level}) + \ldots
\]

Using this formula, a risk score was calculated for each patient, and the ability of this score to predict survival was evaluated by time-dependent receiver operating characteristic (ROC) curves in three years. Patients were divided into groups at high or low risk based on the median risk score, and shown on a non-cluster heat map. Kaplan–Meier survival curves were compared between patients at high or low risk. All these analyses were performed using R/Bioconductor (version 3.4.4).

Validation of the prognostic significance of risk scoring systems
Uni- or multivariate analyses were used to verify associations between risk score and survival. If these analyses did not return any significant results, stratified analyses were conducted to identify factors that might be affecting the results, based on the chi-squared test. All these analyses were carried out using SPSS 16.0 (IBM, Chicago, IL, USA), and the threshold for significance was a two-sided \(p < 0.05\).

Co-expression and functional analyses of lncRNA-related mRNAs
Based on data from the 207 HCC patients, mRNAs whose expression co-varied with that of lncRNAs in the risk scoring system were identified based on a two-sided \(|\text{Pearson correlation coefficient}| > 0.40\) and a \(z\)-test \(p < 0.01\) (Fan and Liu, 2016). These mRNAs were analyzed for Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment using the “clusterProfiler” package in R (Yu et al., 2012). Differences in enrichment were considered significant when associated with \(p < 0.05\).

Results

LncRNAs differentially expressed in the presence or absence of cirrhosis in HCC patients
Our analysis included the lncRNA expression profiles in 77 HCC patients with cirrhosis and 130 HCC without cirrhosis obtained from the TCGA database. A total of 81 differentially expressed lncRNAs were identified, 22 (27.16%) of which were up-regulated in cirrhosis and 59 (72.84%) down-regulated. Table 2 shows the first 20 up- and down-regulated lncRNAs, together with the corresponding values for \log2FC, \(p\), and FDR. Figure 1 shows all differentially expressed lncRNAs plotted according to \(-\log10\) (FDR) and \log2FC, while Figure 2 shows a heatmap indicating the relative specificity of differentially expressed lncRNAs.

Risk scoring system based on lncRNA expression and OS prediction
Univariate Cox analysis of patients with cirrhosis identified five lncRNAs (LINC02086, LINC00880, LINC01549, AC136475.3 and HOXA-AS3) significantly associated with OS (\(p < 0.05\)). Of these, multivariate regression analyses identified the first four as independent prognostic indicators of OS (Table 3). The resulting risk scoring system was

\[
\text{Risk score} = (0.335 \cdot \text{LINC02086}) + (0.372 \cdot \text{LINC00880}) + (0.161 \cdot \text{LINC01549}) + (-0.309 \cdot \text{AC136475.3}).
\]

In this scoring system, higher expression of LINC02086 and LINC00880 predicted worse OS (\(\beta > 0\)), while higher expression of AC136475.3 and LINC01549 predicted better OS (\(\beta < 0\)). Based on their risk scores, patients with cirrhosis were
After obtaining these promising results, we wished to check whether risk scoring based on lncRNAs could predict OS independently of clinicodemographic characteristics of the patients. This analysis is important given the heterogeneity of clinical presentation of HCC, and the range of factors that can influence prognosis. First we conducted univariate Cox regression to identify which clinical features were significantly associated with OS, and then we subjected this subset of factors to classified as being at low or high risk of poor OS using the median risk score as a cut-off (Figure 3A). Kaplan–Meier curves showed that high-risk patients showed significantly lower OS rates at 3 years (65.8% vs 89.1%) and 5 years (20.6% vs 82.2%) (Figure 4A). The area under the ROC curve (AUC) for the risk scoring system was 0.818 (Figure 5A).

Table 1. Clinicopathological Characteristics of 207 HCC Patients with or without Cirrhosis

| Clinicopathological characteristics | Patients (n=207) | n   | %    |
|------------------------------------|-----------------|-----|------|
| Age                                |                 |     |      |
| ≤60                                |                 | 96  | 46.4 |
| >60                                |                 | 111 | 53.6 |
| BMI                                |                 |     |      |
| <25                                |                 | 94  | 45.4 |
| ≥25                                |                 | 103 | 49.8 |
| Not reported                       |                 | 10  | 4.8  |
| Race                               |                 |     |      |
| Non-Asian                          |                 | 121 | 58.5 |
| Asian                              |                 | 80  | 38.6 |
| Not reported                       |                 | 6   | 2.9  |
| AFP                                |                 |     |      |
| ≤20ng/mL                           |                 | 109 | 52.7 |
| >20ng/ml                           |                 | 76  | 36.7 |
| Not reported                       |                 | 22  | 10.6 |
| Gender                             |                 |     |      |
| Female                             |                 | 66  | 31.9 |
| Male                               |                 | 141 | 68.1 |
| Alcohol consumption                |                 |     |      |
| NO                                 |                 | 148 | 71.5 |
| YES                                |                 | 50  | 24.2 |
| Not reported                       |                 | 9   | 4.3  |
| Hepatitis B or C                   |                 |     |      |
| NO                                 |                 | 94  | 45.4 |
| YES                                |                 | 104 | 50.3 |
| Not reported                       |                 | 9   | 4.3  |
| Cirrhosis status                   |                 |     |      |
| NO                                 |                 | 130 | 62.8 |
| YES                                |                 | 77  | 37.2 |
| Histologic grade                   |                 |     |      |
| G1-2                               |                 | 133 | 64.2 |
| G3-4                               |                 | 72  | 34.8 |
| Not reported                       |                 | 2   | 1.0  |
| New tumor event                    |                 |     |      |
| NO                                 |                 | 97  | 46.9 |
| YES                                |                 | 101 | 48.8 |
| Not reported                       |                 | 9   | 4.3  |
| Pathologic stage*                  |                 |     |      |
| Stage I+II                         |                 | 155 | 74.9 |
| Stage III+IV                       |                 | 41  | 19.8 |
| Not reported                       |                 | 11  | 5.3  |
| Cancer status                      |                 |     |      |
| Tumor free                         |                 | 116 | 56   |
| With tumor                         |                 | 84  | 40.6 |
| Not reported                       |                 | 7   | 3.4  |
| Family cancer history              |                 |     |      |
| NO                                 |                 | 111 | 53.6 |
| YES                                |                 | 68  | 32.9 |
| Not reported                       |                 | 28  | 13.5 |
| Residual tumor                     |                 |     |      |
| R0                                 |                 | 192 | 92.7 |
| non-R0                             |                 | 13  | 6.3  |
| Not reported                       |                 | 2   | 1.0  |
| Vascular invasion                  |                 |     |      |
| Negative                           |                 | 138 | 66.7 |
| Positive                           |                 | 60  | 29   |
| Not reported                       |                 | 9   | 4.3  |

BMI, Body mass Index; AFP, Alpha fetoprotein; *TNM staging systems.

Figure 1. Volcano Map of the Differentially Expressed lncRNAs in HCC Patients between with Cirrhosis or without Cirrhosis. Red spots represent up-regulated genes, and green spots represent down-regulated genes.

Figure 2. Heatmap Based on the Differentially Expressed lncRNAs in HCC Patients between with Cirrhosis or without Cirrhosis.
Univariate analysis of patients with cirrhosis identified risk score and age as significantly associated with OS, but not body mass index (BMI), ethnicity, alpha fetoprotein (AFP), gender, hepatitis, alcohol consumption, cancer status, histology grade, new tumor event, pathology stage, family cancer history, residual tumor, or vascular invasion. Multivariate Cox regression confirmed that age was an independent risk factor for OS. Table 2 presents the top 20 up-regulated and down-regulated lncRNAs in HCC patients with cirrhosis compared to those without cirrhosis. Table 3 lists the four lncRNAs significantly correlated with OS of HCC patients with cirrhosis in the best statistical model. The risk score rises gradually from left to right in the Non-Cluster Risk Heat Map (Figure 3).
independent predictor of poor OS [hazard ratio (HR) 2.86, 95%CI 1.09-7.56], as was risk score (HR 4.08, 95%CI 1.43-11.68) (Table 4).

**Risk scoring system based on lncRNA expression and RFS prediction**

Univariate analysis identified the following 11 lncRNAs as significantly correlated with RFS of patients with cirrhosis: SH3RF3-AS1, AC104117.3, AC136475.3, LINC00239, MRPL23-AS1, LINC00494, LINC01970, MEG3, Z93930.3, MIR9-3HG and TRBV11-2. Multivariate analysis showed the first five to be independent prognostic indicators of RFS (Table 5). The resulting risk scoring system was:

Risk score = (-0.2730 * SH3RF3-AS1) + (-0.2463 * MRPL23-AS1) + (-0.2425 * LINC00239) + (-0.2497 * AC136475.3) + (-0.3609 * AC104117.3), in which higher expression of all five lncRNAs was associated with better RFS ($\beta < 0$).

Based on their risk scores, patients with cirrhosis were classified as being at low or high risk of poor RFS using the median risk score as cut-off (Figure 3B). Kaplan–Meier curves showed that high-risk patients showed significantly lower RFS rates at 3 years (25.3% vs 66.8%) and 5 years (16.9% vs 57.3%) (Figure 4B). The AUC of the risk scoring system was 0.819 (Figure 5B).

As we did above for OS, we wished to check whether...
risk scoring based on lncRNAs could predict RFS independently of clinicodemographic characteristics of the patients. For patients with cirrhosis, our analysis showed that AFP and vascular invasion, but not risk score, correlated significantly with RFS in univariate analysis (Table 6). Frequencies of patients with certain clinicodemographic characteristics were stratified into groups at low or high risk of RFS; this analysis identified...
significant associations of the risk score with AFP, new tumor event and cancer status (Table 7).

Table 6. Univariate Cox Regression Analysis for RFS in HCC Patients with Cirrhosis

| Variables                  | P-value | HR    | 95% CI  |
|----------------------------|---------|-------|---------|
| Risk score (high/low)      | 0.92    | 0.93  | 0.23 3.79 |
| Age (≥60/≤60)              | 0.11    | 0.33  | 0.08 1.3  |
| BMI                       | 0.94    |       |        |
| <25                       | Reference| 0.72  | 0.1 4.95  |
| ≥25                       | 0.23    | 3.79  | 2.21E+173  |
| Not reported              | 390.28 | 0     | 3.52E+226  |
| Race                      | 0.35    |       |        |
| Non-Asian                 | Reference| 0.22  | 0.03 1.68  |
| Asian                     | 0.72    | 3.49  | 1.97E+109  |
| Not reported              | 23.5    | 0     | 3.52E+226  |
| AFP                       | 0.03    |       |        |
| ≤20ng/mL                  | Reference| 4.48  | 0.45 44.66 |
| >20ng/ml                  | 0.03    | 0.45  | 44.66  |
| Not reported              | 45.46   | 2.18  | 947.68  |
| Gender (Male/Female)      | 0.52    |       |        |
| Female                    | Reference| 2.6   | 0.14 47.19 |
| Male                      | 0.72    | 1.3   | 6.845  |
| Hepatitis B or C          | 0.3     |       |        |
| No                        | Reference| 0.35  | 0.07 1.61  |
| Yes                       | 0.72    | 3.49  | 1.97E+109  |
| Not reported              | 0.13    | 0.07  | 1.61  |
| Alcohol consumption (Yes/No)| 0.41  | 0.42  | 0.37  |
| Histologic grade          | 0.32    |       |        |
| G1-2                      | Reference| 5.29  | 0.61 45.68 |
| G3-4                      | 0.72    | 3.79  | 2.21E+173  |
| Not reported              | 67.46   | 0     | 1.27E+173  |
| New tumor event           | 0.66    |       |        |
| No                        | Reference| 5.34  | 1.16 24.58 |
| Yes                       | 0.72    | 3.79  | 2.21E+173  |
| Pathologic stage*         | 0.1     |       |        |
| Stage I+II                | Reference| 5.34  | 1.16 24.58 |
| Stage III+IV              | 0.72    | 3.79  | 2.21E+173  |
| Not reported              | 67.46   | 0     | 1.27E+173  |
| Cancer status             | 0.1     |       |        |
| Tumor free                | Reference| 6.89  | 1.17 40.49 |
| With tumor                | 0.72    | 3.79  | 2.21E+173  |
| Not reported              | 1.46    | 0     | 7.08E+07  |
| Family cancer history     | 0.23    |       |        |
| No                        | Reference| 1.21  | 0.09 15.49 |
| Yes                       | 0.72    | 3.79  | 2.21E+173  |
| Not reported              | 0.11    | 0.09  | 1.38  |
| Residual tumor            | 0.71    |       |        |
| <0.05                     | Reference| 0.58  | 0.04 9.56  |
| Vascular invasion         | 0.71    |       |        |
| Negative                  | Reference| 3.4   | 0.39 19.61 |
| Positive                  | 0.72    | 3.49  | 1.97E+109  |
| Not reported              | 0.07    | 0     | 6.845  |

Note: BMI, Body mass index; AFP, Alpha fetoprotein; HR, Hazard ratio; CI, Confidence interval; *TNM staging systems.

Table 7. Stratified Analyses for the Risk Score of RFS in HCC Patients with Cirrhosis Using Chi-Square Test

| Variables                  | Risk score | P-value |
|----------------------------|------------|---------|
| Age (≤60/>60)              | 0.92       | 0.19    |
| BMI                       | 0.9        | 0.09    |
| <25                       | Reference  | 0.03    |
| ≥25                       | 0.03       | 0.09    |
| Not reported              | 390.28     | 0       |
| Race                      | 0.1        | 0.01    |
| Non-Asian                 | Reference  | 0.1     |
| Asian                     | 0.1        | 0.01    |
| Not reported              | 390.28     | 0       |
| AFP                       | 0.03       | 0.09    |
| ≤20ng/mL                  | Reference  | 0.03    |
| >20ng/ml                  | 0.03       | 0.09    |
| Not reported              | 390.28     | 0       |
| Gender (Male/Female)      | 0.52       | 0.01    |
| Female                    | Reference  | 0.52    |
| Male                      | 0.52       | 0.01    |
| Hepatitis B or C          | 0.3        | 0.01    |
| No                        | Reference  | 0.3     |
| Yes                       | 0.3        | 0.01    |
| Not reported              | 390.28     | 0       |
| Alcohol consumption       | 0.85       | 0.01    |
| No                        | Reference  | 0.85    |
| Yes                       | 0.85       | 0.01    |
| Histologic grade          | 0.39       | 0.01    |
| G1-2                      | Reference  | 0.39    |
| G3-4                      | 0.39       | 0.01    |
| Not reported              | 390.28     | 0       |
| New tumor event           | 0.0        | 0.01    |
| No                        | Reference  | 0.0     |
| Yes                       | 0.0        | 0.01    |
| Not reported              | 390.28     | 0       |
| Pathologic stage*         | 0.1        | 0.01    |
| Stage I+II                | Reference  | 0.1     |
| Stage III+IV              | 0.1        | 0.01    |
| Not reported              | 390.28     | 0       |
| Cancer status             | 0.1        | 0.01    |
| Tumor free                | Reference  | 0.1     |
| With tumor                | 0.1        | 0.01    |
| Not reported              | 390.28     | 0       |
| Family cancer history     | 0.49       | 0.01    |
| No                        | Reference  | 0.49    |
| Yes                       | 0.49       | 0.01    |
| Not reported              | 390.28     | 0       |
| Residual tumor            | 0.64       | 0.01    |
| <0.05                     | Reference  | 0.64    |
| Vascular invasion         | 0.08       | 0.01    |
| Negative                  | Reference  | 0.08    |
| Positive                  | 0.08       | 0.01    |

Note: BMI, Body mass index; AFP, Alpha fetoprotein; *TNM staging systems.
These related mRNAs were analyzed using the KEGG signal pathway databases (Figure 6). Functional enrichment analysis showed that mRNAs strongly related to the lncRNAs in our risk scoring systems were involved mainly in the pathway of Wnt signal (Supplementary Figure 1) and cytokine-cytokine receptor interaction (Supplementary Figure 2).

Discussion

HCC is associated with high morbidity and poor prognosis, and the factors that contribute to poor outcomes are likely to vary substantially in the presence or absence of cirrhosis (Grazi et al., 2003; Gassmann et al., 2010; Wang et al., 2013; Techathuvanan et al., 2015). Therefore we developed an lncRNA-based scoring system to assess a patient’s risk of poor OS or RFS in the presence of cirrhosis. Our results establish the potential of four or five lncRNAs to serve as prognostic biomarkers with respective AUCs of 0.818 and 0.819, suggesting reasonable predictive power.

After adjusting for other clinical variables, uni- and/or multivariate Cox analyses showed the lncRNA-based scoring system to be significantly associated with OS of HCC patients with cirrhosis, suggesting the lncRNA-based scoring system can serve as a significant independent predictor. However, the system for predicting RFS was not significantly associated with RFS in univariate analysis. Therefore, we stratified patients by low or high risk score and found the following factors to influence risk: AFP, new tumor event and cancer status. Our results suggest that these factors should be taken into account when predicting RFS in HCC patients with cirrhosis. Furthermore, other clinical factors also influenced the survival of HCC patients in our sample. Our study showed that age was an independent factor for the prediction of OS in HCC patients with cirrhosis, which was similar with previous studies (Wang et al., 2013; Techathuvanan et al., 2015; Liu et al., 2018). To gain insight into the prognostic significance of our risk scoring systems, we analyzed the molecular functions of genes highly related to these lncRNAs. We found that the lncRNAs associated with prognosis in HCC are involved mainly in Wnt signaling and cytokine-cytokine receptor interactions.

Several previous studies constructed risk scoring systems to predict the prognosis of HCC patients (Gu et al., 2018; Liao et al., 2018; Ma et al., 2018; Shi et al., 2018; Sui et al., 2018; Wu et al., 2018b; Zhao et al., 2018; Bai et al., 2019; Yan et al., 2019; Ye et al., 2019; Zhang et al., 2019). However, all these risk scoring systems are based on lncRNAs differentially expressed between HCC and normal samples, while our systems are based on lncRNAs differentially expressed between HCC patients with or without cirrhosis. Therefore, our systems may be more specific for HCC involving cirrhosis. One previous study identified a four-lncRNA signature as a prognostic indicator in cirrhotic HCC (Ma and Deng, 2019), with no overlap with our four or five-lncRNA signatures. Those authors extracted their data from a different expression profile dataset, and they used only stratified analyses based on clinical characteristics to validate the prognostic significance of their risk scoring system.

In conclusion, we have defined lncRNA-based risk scoring systems that can effectively predict OS in HCC patients with cirrhosis. Our work highlights the potential usefulness of lncRNAs as prognostic biomarkers, and it provides several leads for the development of novel therapies.

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Appendix

Supplementary Table 1. LncRNA-mRNA co-expression analysis for OS in HCC with cirrhosis.

Supplementary Table 2. LncRNA-mRNA co-expression analysis for RFS in HCC with cirrhosis.

Supplementary Figure 1. Wnt signaling pathway

Supplementary Figure 2. The pathway of cytokine-cytokine receptor interaction

Conflicts of interest

The authors have no conflicts of interest to declare.
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