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Expression and Significance of KLKB1 in Hepatocellular Carcinoma based on Multiple Gene Databases

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Abstract

Background: The expression and mechanism of plasma kallikrein B1 (KLKB1) in hepatocellular carcinoma were analyzed by deeply mining the gene information in Oncomine and other databases. Methods: The BioGPS database was used to analyze the expression of the KLKB1 gene in normal tissues. Collect the information about the study of KLKB1 in Oncomine database and analyze its expression level in hepatocellular carcinoma. Kaplan Meier method was used to analyze the relationship between the expression level of KLKB1 and the survival of patients with liver cancer, and to explore its clinical significance. The proteins related to KLKB1 gene were collected by GeneCards database, and the network diagram of KLKB1 related proteins was drawn by string to analyze the physiological process of protein enrichment. Result: The relevant results of the BioGPS database analysis showed that KLKB1 is under-expressed in normal liver tissue. Three studies on the differential expression of KLKB1 gene between liver cancer and normal liver tissues were collected in Oncomine database. Data analysis showed that the expression of KLKB1 gene in liver cancer tissues was significantly lower than that in normal liver tissues (P < 0.05). Kaplan Meier survival analysis showed that the overall survival time of liver cancer patients with low expression of KLKB1 gene was significantly shorter than that of patients with low expression, and the prognosis of patients with high expression was better (P< 0.05). GeneCards database collected 25 proteins related to KLKB1, including HGF, TFPI2 and C1QBP. The enrichment analysis results of related proteins showed that they were mainly enriched in physiological processes such as coagulation process, protein activation process, positive regulation of fibrinolysis. Conclusion: KLKB1 gene may play a role in the occurrence and development of liver cancer by regulating the process of hemagglutination. Low expression suggests that the prognosis of patients with liver cancer is poor. Targeting KLKB1 may be a potential tool for tumor diagnosis and treatment.

Keywords

Hepatocellular Carcinoma; KLKB1; Multiple Gene Databases.

1. Introduction

Hepatocellular carcinoma (HCC) is the second most common cause of cancer-related death worldwide [1]. Although great progress has been made, its diagnostic and screening capabilities also limited [2]. Therefore, looking for molecular indicators that can be used for early diagnosis and

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predict the prognosis of HCC patients is of great clinical significance to improve the early diagnosis rate and prognosis of patients with HCC.

Plasma kallikrein B1 (KLKB1) is a serine protease that can circulate in plasma by binding to high molecular weight kininogen or as a free protein [3]. Recently, accumulating studies have found that KLKB1 dysregulation is related to a great number of malignant tumors, which has been found to be involved in the occurrence and development of tumor by more and more studies, and is expected to become a new target for tumor diagnosis and treatment [4-7]. The purpose of the present study was to explore the relationship between KLKB1 expression level and clinical prognosis of HCC patients.

2. Material and Methods

2.1 BioGPS Database Analysis

The expression of KLKB1 gene in normal human tissues was analyzed by using the BioGPS database platform (http://biogps.org).

2.2 Oncominedatabase Analysis

Oncomine database (https://www.oncomine.org/resource/login.html) was used to predict the KLKB1 mRNA expression levels in HCC and normal tissues. The settings for this research study were: (1) Gene: KLKB1 (2) Analysis Type: Hepatocellular Carcinoma vs. Normal Analysis (3) Threshold By: P-Value: 1E-6, Fold Change: 4, Gene Rank: Top 10%.

2.3 Kaplan-meier Plotter Database Analysis

An online survival analysis was performed by using the Kaplan-Meier Plotter database http:// kmplot. com/analysis/). The screening conditions were as follows: (1) Cancer: Ovarian Cancer; (2) Gene: PAX8; (3) Survival: PFS, OS.

2.4 Predicting the Targets of KLKB1

GeneCards database (https://www.genecards.org/)was used to gather the information on KLKB1-associated target genes [8]. The targets of KLKB1 were put into STRING (https://string-db. org/cgi/input.pl) to build the PPI network interaction.

2.5 GO and KEGG Pathway Enrichment Analysis

We used DAVID to perform GO function and KEGG pathway enrichment analysis for the DEGs from the Venn package, and FDR < 0.05 was considered statistically significant. GO enrichment analysis included biological processes (BP), molecular functions (MF), and cellular components (CC).

3. Results

3.1 KLKB1 Gene Expression in Normal Human Tissues

Results from the BioGPS database analysis showed that KLKB1 was under-expressed in normal liver tissue (Figure 1).

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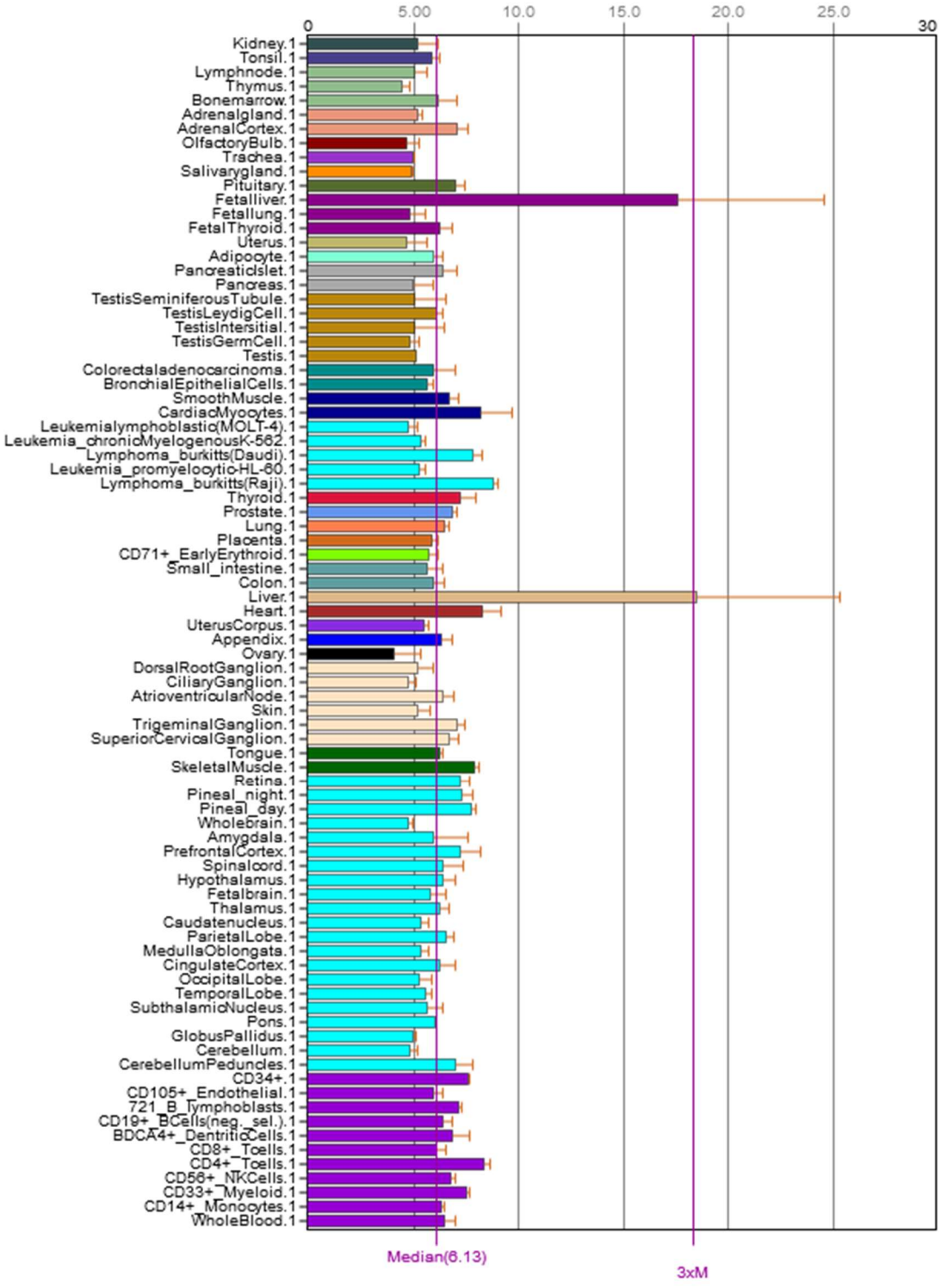

Figure 1. Expression of KLKB1 in normal human tissues

3.2 Expression of KLKB1 in Tumor Types

Oncomine data showed that there were 12 low expression level stumor types presenting statistically significant KLKB1 expression levels (Figure 2). A total of 3 studies involving the comparison of KLKB1 expression in liver cancer tissues and normal tissues were conducted (Table 1).

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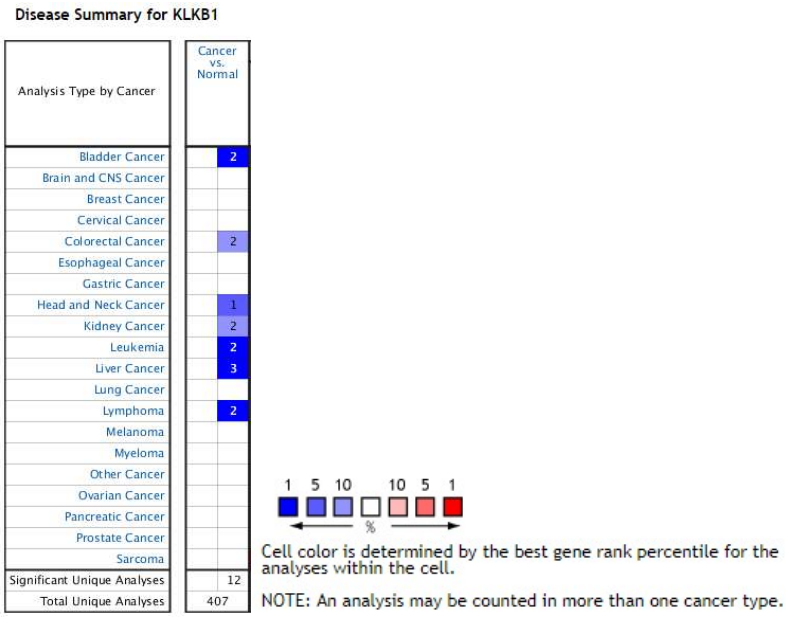


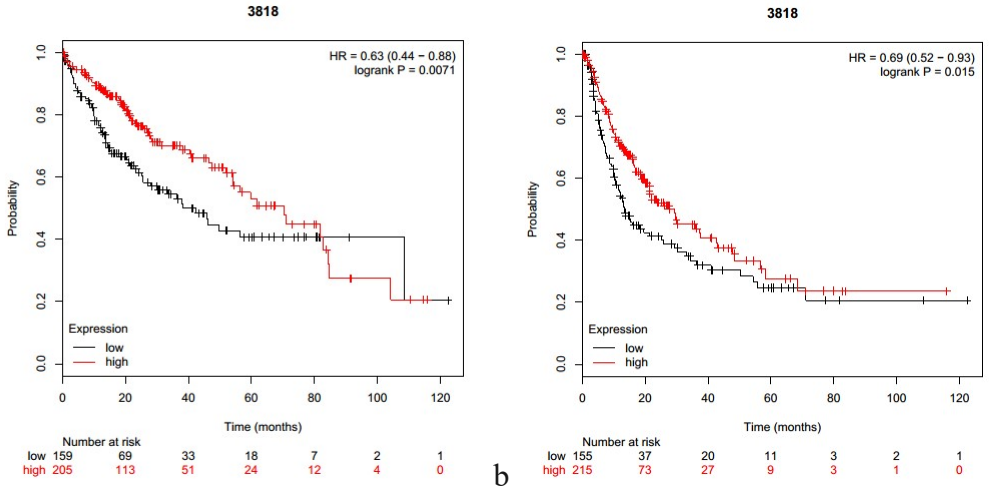
Figure 2. Expression data for KLKB1 in a variety of normal and cancerous human tissues in Oncomine database.

Table 1. The expression of KLKB1 in different liver cancer studies identified in the Oncomine database

Database (sample size)	Comparison groups	Fold Change	*P*-value
Roessler Liver 2 (445)	Hepatocellular Carcinoma vs. Normal	-5.595	8.74E-72
Roessler Liver (43)	Hepatocellular Carcinoma vs. Normal	-5.271	2.98E-11
Wurmbach Liver(45)	Hepatocellular Carcinoma vs. Normal	-4.414	7.71E-7

3.3 Relationship between KLKB1 and Survival Prognosis of HCC Patients

Kaplan-Meier Plotter database showed that the expression level of KLKB1 had a significant effect on the OS (P = 0.0071) and PFS (P = 0.015) of HCC patients. Compared with the high expression group, the total survival time of patients in the group with low KLKB1 expression was significantly reduced (Figure 3).



a b

Figure 3. The relationship between the prognosis of liver cancer and KLKB1 expression (a: OS, b: PFS)

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3.4 Identification of KLKB1-Related Target Genes.

We explored the target genes of KLKB1 using GeneCards database. We constructed the PPI network and found that KLKB1 was closely associated with KNG1, F12, TF, IGFBP3, HGF, AP2M1, HGFAC, SERPINA5, TFPI2, KIF27, MAP2K1 and MAP2K2 proteins (Figure 4).

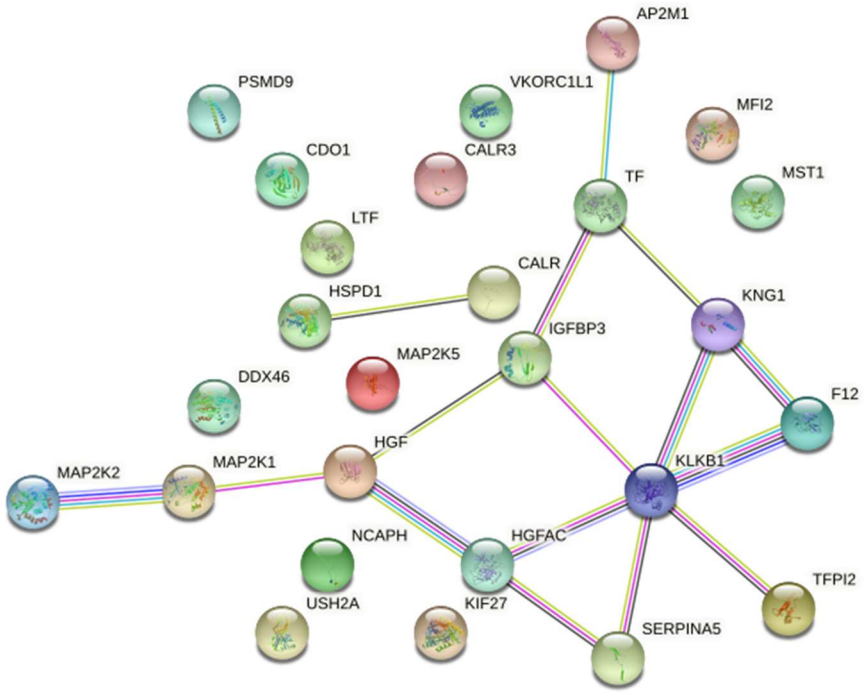

Figure 4. Network diagram of KLKB1 related proteins.

3.5 GO and KEGG Enrichment Analysis.

Table 2. Analysis of GO enrichment of KLKB1 related proteins

Category	GO-term	Description	Count in gene set	*P*-value
BP	GO: 0007596	blood coagulation	11 of 288	6.69E-11
BP	GO: 0007597	blood coagulation, intrinsic pathway	6 of 17	2.98E-11
BP	GO: 0072376	protein activation cascade	7 of 74	1.70E-9
BP	GO: 0051919	positive regulation of fibrinolysis	4 of 4	2.07E-8
BP	GO: 0030193	regulation of blood coagulation	6 of 77	1.15E-7
MF	GO:0008236	serine-type peptidase activity	10 of 207	1.83e-11
MF	GO:0004252	serine-type endopeptidase activity	9 of 180	9.62e-11
MF	GO:0140096	catalytic activity, acting on a protein	12 of 2176	0.00020
MF	GO:0004866	endopeptidase inhibitor activity	4 of 169	0.0017
MF	GO:0070008	serine-type exopeptidase activity	2 of 14	0.0030
CC	GO:0044421	extracellular region part	15 of 1375	3.27e-09
CC	GO:0005576	extracellular region	18 of 2505	4.13e-09
CC	GO:0005615	extracellular space	13 of 1134	2.48e-08
CC	GO:0031091	platelet alpha granule	5 of 91	6.36e-06
CC	GO:0031093	platelet alpha granule lumen	4 of 68	7.78e-05

BP, biological process; CC, cellular component; MF, molecular function.

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GO and KEGG pathway enrichment analyses were conducted to explore the functional characteristics of the target genes. The GO analysis results revealed that the target genes were signifcantly enriched in blood coagulation, blood coagulation, intrinsic pathway, protein activation cascade, positive regulation of fibrinolysis and regulation of blood coagulation in terms of BP. Regarding MF, the target genes were enriched in serine-type peptidase activity, serine-type endopeptidase activity, catalytic activity, acting on a protein endopeptidase inhibitor activity and serine-type exopeptidase activity. Under CC, the target genes were enriched in extracellular region part, extracellular region, extracellular space, platelet alpha granule and platelet alpha granule lumen. In addition, the KEGG analysis results showed that the target genes were signifcantly enriched in Thyroid cancer, Complement and coagulation cascades, Bladder cancer, Renal cell carcinoma and Non-small cell lung cancer. The enriched GO terms and KEGG pathways are shown in Table 2 and Table 3.

Table 3. Analysis of KEGG pathway enrichment of KLKB1 related proteins

Pathway	Description	Count in network	*P*-value
hsa05216	Thyroid cancer	2 of 36	0.0365
hsa04610	Complement and coagulation cascades	4 of 82	0.0022
hsa05219	Bladder cancer	2 of 41	0.0373
hsa05211	Renal cell carcinoma	3 of 66	0.0233
hsa05223	Non-small cell lung cancer	3 of 68	0.0233

4. Discussion

Hepatocellular carcinoma (HCC) is the leading cause of cancer-related deaths worldwide. Although many efforts for treating HCC have been made, the survival rate remains unsatisfied. KLKB1 is a serine protease, which can be combined with high molecular weight kininogen or circulated in plasma as a free protein [9]. KLKB1 can catalyze the cleavage of plasminogen into plasmin and participate in a variety of physiological and pathological processes such as endogenous coagulation cascade, inflammatory reaction, fibrinolytic system, renin angiotensin system [10-14]. Accumulating evidence indicates that KLKB1 functions as a tumor biomarker and involves in many biological processes such as tumor initiation, development, and metastasis in certain types of human cancers. Joshua Wong et al. Found that the expression of KLKB1 in lung cancer and mesothelioma cells after demethylation intervention was significantly increased. In addition, KLKB1 can interfere with the formation of new blood vessels by activating kinin B1 and B2 receptors, so as to inhibit the growth and proliferation of lung and pleural tumors, suggesting that klkb1 can regulate the occurrence and development of tumors as an tumor suppressor gene [15].

In this study, the BioGPS database was used to analyze the expression of the KLKB1 gene in normal tissues. Collect the information about the study of KLKB1 in Oncomine database and analyze its expression level in hepatocellular carcinoma. Three studies on the differential expression of KLKB1 gene between liver cancer and normal liver tissues were collected in Oncomine database. Data analysis showed that the expression of KLKB1 gene in liver cancer tissues was significantly lower than that in normal liver tissues ($P < 0.05$). Kaplan Meier survival analysis showed that the overall survival time of liver cancer patients with low expression of KLKB1 gene was significantly shorter than that of patients with low expression, and the prognosis of patients with high expression was better. GeneCards database collected 25 proteins related to KLKB1, including KNG1, F12 and TF. The enrichment analysis results of related proteins showed that they were mainly enriched in physiological processes such as coagulation process, protein activation process, positive regulation of fibrinolysis.

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In conclusion, KLKB1 gene may play a role in the occurrence and development of liver cancer by regulating the process of hemagglutination. Low expression suggests that the prognosis of patients with liver cancer is poor. Targeting KLKB1 may be a potential tool for tumor diagnosis and treatment.

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Author disclosure statement

The authors declare that no competing financial interests exist.

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