SPANDIDOS PUBLICATIONS

ONCOLOGY LETTERS 20: 509-516, 2020

High KIAA1522 expression predicts a poor prognosis in patients with hepatocellular carcinoma

YONGZHENG XU, CHUANDONG SUN, BING HAN, YUE XI, MAO ZHANG, JING YANG and ZONGKAI CHEN

Department of Hepatobiliary and Pancreatic Surgery, The Affiliated Hospital of Qingdao University, Qingdao, Shandong 266003, P.R. China

Received July 28, 2019; Accepted January 30, 2020

DOI: 10.3892/ol.2020.11588

Abstract. Hepatocellular carcinoma (HCC) is a highly malignant tumor associated with a poor prognosis, and the molecular mechanisms remain poorly understood. KIAA1522 expression is upregulated in various types of tumor tissue; however, its function remains unknown in HCC. Bioinformatics analysis was undertaken using Oncomine, OncoLnc and other databases, in order to determine KIAA1522 expression in HCC and to analyze its association with postoperative prognosis. Reverse transcription-quantitative PCR was performed to detect KIAA1522 mRNA expression in primary HCC and adjacent normal tissues, while KIAA1522 protein expression was assessed via immunohistochemical staining. KIAA1522 expression and clinicopathological characteristics of primary HCC were evaluated, and their association with patient prognosis was analyzed. The Oncomine database results indicated that KIAA1522 expression in HCC and normal liver tissues was significantly different. RT-qPCR analysis demonstrated that KIAA1522 mRNA expression was significantly higher in HCC tissues compared with that in adjacent normal tissues. Immunohistochemical analysis indicated that expression rate of KIAA1522 protein was significantly higher in primary HCC tissues compared with that in normal liver tissues. The OncoLnc database results demonstrated that KIAA1522 expression was significantly associated with short-term survival. Kaplan-Meier survival analysis indicated that high KIAA1522 protein expression was significantly associated with short-term survival for patients with HCC. Multivariate Cox regression analysis demonstrated that tumor size, Tumor-Node-Metastasis stage and high KIAA1522 protein expression were independent predictors of a poor prognosis in patients with primary HCC. Furthermore, high KIAA1522 expression was significantly associated with postoperative survival time in primary HCC, and thus may be a potential molecular marker for prognosis in patients with this cancer type.

Correspondence to: Dr Chuandong Sun, Department of Hepatobiliary and Pancreatic Surgery, The Affiliated Hospital of Qingdao University, 16 Jiangsu Road, Qingdao, Shandong 266003, P.R. China
E-mail: sunchuandong@hotmail.com

Key words: KIAA1522, hepatocellular carcinoma, bioinformatic analysis, multivariate Cox regression analysis, prognosis

Introduction

Hepatocellular carcinoma (HCC) is the most common type of liver cancer and a malignant tumor that is detrimental to human health (1,2). Geographically, the incidence rates in North and West Africa, and East and Southeast Asia are high. In China, the annual incidence of HCC is 20.37/100,000, which accounts for 50% of all reported HCC cases worldwide (3), and its mortality rate (15.2%) was reported to be the second highest among all cancer-associated mortalities in 2018 (4,5). Previous studies have reported that the 5-year survival rate of patients with HCC is only 3-5%, and ~600,000 patients die from HCC annually in China (3,6). Although patients with HCC can be treated using various measures, including surgical resection, liver transplantation, local treatment and chemotherapy, the prognosis of HCC remains poor (7,8). This is due to high recurrence and metastasis, which are the predominant causes of mortality in patients with HCC (9). Regarding patients with severe early disease who undergo liver resection or liver transplantation, >80% exhibit recurrence following surgery, which affects their quality of life (10). A number of factors have predictive prognostic value for HCC, including tumor size, tumor differentiation, vascular invasion, marginal resection, and novel immunological and tissue biomarkers (11-15). Currently, different biomarkers, such as α-fetoprotein (AFP) and des-γ-carboxyprothrombin, are being implemented as a base to establish a series of diagnostic and long-term prognostic guidelines for HCC (16). However, recent studies have failed to provide sufficient unbiased data to establish these biomarkers as effective monitoring, diagnostic and prognostic tools, due to their poor specificity and sensitivity (16,17). Thus, the development of novel molecular markers that can be used to accurately determine the prognosis or recurrence of HCC has critical clinical application value, and selecting appropriate treatment plans for patients is beneficial to improve the survival rate of HCC.

KIAA1522 is a newly cloned, large protein-coding gene of unknown function (18). Information from the Gene Expression Atlas (http://www.ebi.ac.uk/gxa) and The

European Bioinformatics Institute databases (https://www.ebi.ac.uk) demonstrates that KIAA1522 mRNA expression is upregulated in various tumor tissues, such as lung and breast cancer, suggesting that KIAA1522 may play a role in tumorigenesis and cancer development (18). However, to the best of our knowledge, KIAA1522 expression in HCC has not yet been fully investigated. Thus, the present study performed bioinformatics analysis, using the Oncomine (www.oncomine.org) and OncoLnc (www.oncolnc.org) databases, in order to analyze KIAA1522 expression and assess its association with the clinical prognosis of HCC. KIAA1522 expression in HCC and adjacent normal tissues was detected via immunohistochemical staining and reverse transcription-quantitative (RT-q)PCR analysis. Additionally, the association between KIAA1522 expression and the clinicopathological characteristics and prognosis of HCC was analyzed by combining clinical pathology and follow-up data.

Materials and methods

Bioinformatics analysis. The Chen Liver dataset (19) was searched within the Oncomine database with the following specifications: Gene, KIAA1522; analysis type, cancer vs. normal analysis; and cancer type, liver cancer. KIAA1522 expression levels in HCC and adjacent normal liver tissues were analyzed in the database and plotted using GraphPad Prism software (version 6; https://www.graphpad.com). Subsequently, the dataset [which comprised 217 datapoints from The Cancer Genome Atlas (TCGA) database (https://tcga-data.nci.nih.gov/tcga)] was searched for within the OncoLnc database with the following parameters: KIAA1522; liver hepatocellular carcinoma; lower percentile, 30%; and upper percentile, 30%. A survival curve was generated and plotted using GraphPad Prism 6.

Patients and samples. A total of 79 paraffin-embedded primary HCC tissue samples, from 64 males and 15 females (median age, 55 years; age range, 24-73 years) were surgically resected and collected between January 2013 and December 2013 at The Affiliated Hospital of Qingdao University (Qingdao, China). The inclusion criteria were as follows: Postoperative pathology confirmed as HCC and Tumor-Node-Metastasis (TNM) staging (20), TnN0M0. The exclusion criteria were as follows: Received any anticancer treatment prior to surgery; serious complications or mortality within 30 days following surgery; non-tumor associated mortalities, and incomplete clinical, pathological and surgical data.

According to the eighth edition of the 2017 American Joint Committee on Cancer TNM staging system (www.cancerstaging.org), there were 39 cases in clinical stage I, 10 cases in clinical stage II and 30 cases in clinical stage IIIa. The pathological differentiation grade, based on the World Health Organization tumor histological classification criteria (21), were as follows: A total of 3 cases with high differentiation, 50 cases with moderate differentiation and 26 cases with poor differentiation. Liver function grading was performed using the Child-Pugh grading standard (22), which was scored according to the most recent clinical data prior to surgery, and included 78 cases of grade A and 1 case of grade B.

RT-qPCR. Total RNA was extracted from surgically resected HCC tissues using TRIzol® reagent (Invitrogen; Thermo fisher Scientific, Inc.) and reversed transcribed into first-strand cDNA using the First-Strand Synthesis System for RT-PCR (Takara Biotechnology Co., Inc.). qPCR was subsequently performed using the SYBR Green kit (Takara Biotechnology Co., Inc.). The following primer sequences were used for qPCR: KIAA1522 forward, 5'-TGGATGAGCACCAGGACAAC-3' and reverse, 5'-GTCCGGGAGGACTGGATACT-3'; and β-actin forward, 5'-CCTCTCCCAAGTCCACACAG-3' and reverse, 5'-GGGCACGAAGGCTCATCATT-3'. The following thermocycling conditions were used for qPCR: 24 cycles of initial denaturation at 98˚C for 30 sec; 30 cycles of 98˚C for 15 sec, 72˚C for 30 sec; and a final extension at 72˚C for 30 sec, and then cooled at 4˚C for 15 sec. Relative mRNA expression levels were measured using the $2^{-\Delta\Delta Cq}$ method (23). All experiments were performed in triplicate.

Immunohistochemistry. A total of 79 paraffin-embedded primary HCC tissue samples were fixed in 4% formaldehyde for 24 h at room temperature and embedded in paraffin. Paraffin-embedded samples were cut into 4-um-thick sections. Tissue samples were subsequently deparaffinized in xylene at room temperature for 15 min, and rehydrated in a descending ethanol series (anhydrous ethanol I, 5 min; anhydrous ethanol II, 5 min; 95% ethanol, 3 min; 90% ethanol, 3 min; 80% ethanol, 2 min and 70% ethanol, 2 min). Following antigen retrieval with 10 mM citrate buffer (pH 6; Beijing g-clone Biotechnology Co., Ltd.; http://www.g-clone.com) at 100˚C for 10-15 min, tissue sections were blocked with 3% hydrogen peroxide for 10 min at 37˚C to inhibit endogenous peroxidase activity. Tissue samples were subsequently incubated with rabbit anti-human KIAA1522 antibody (1:200; cat. no. PAB22604; Abnova), overnight at 4˚C in a humidified chamber. The PV-9000 Two-step Immunohistochemistry kit (OriGene Technologies, Inc.) was used to detect primary antibodies. Tissue sections were dehydrated in a descending ethanol series, washed twice with running water and mounted. The slides were subsequently stained with 3,3'-diaminobenzidine for 10 min at room temperature, and counterstained with haematoxylin at room temperature for 1 min.

Simultaneously, two pathologists assessed the pathological sections in a blinded manner, and when the results varied, a third person assessed the pathological sections to discuss the differing observations. HCC tissue samples were observed in 10 randomly selected fields of view under high power light microscope (magnification, x200). The degree of staining of positive cells was scored as follows: 0, no staining; 1, pale yellow; 2, brownish yellow; and 3, brown. The percentage of positive cells were scored as follows: 0, <5%; 1, 6-25%; 2, 26-50%; 3, 51-75% and 4, >75%. The two scores were added together, and all cases were divided into the low and high expression groups. Total scores <2 were assigned to the low expression group, while scores ≥2 were assigned to the high expression group.

Case follow-up. Patients who met the study criteria were closely followed via outpatient review and telephone. The regular follow-up plan was set as follows: Every 3 months for 1 year post-surgery, and every 6 months between 2 and

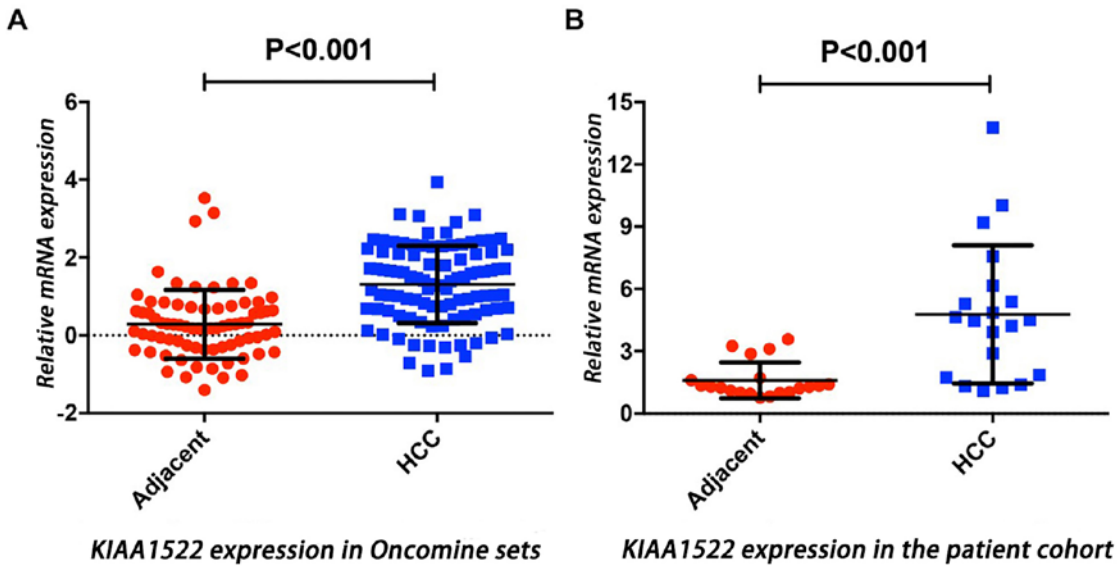

Figure 1. Identification of KIAA1522 as an overexpressed gene in HCC (A) KIAA1522 mRNA expression in the Oncomine sets. (B) KIAA1522 expression in the patient cohort assessed in the present study. HCC, hepatocellular carcinoma.

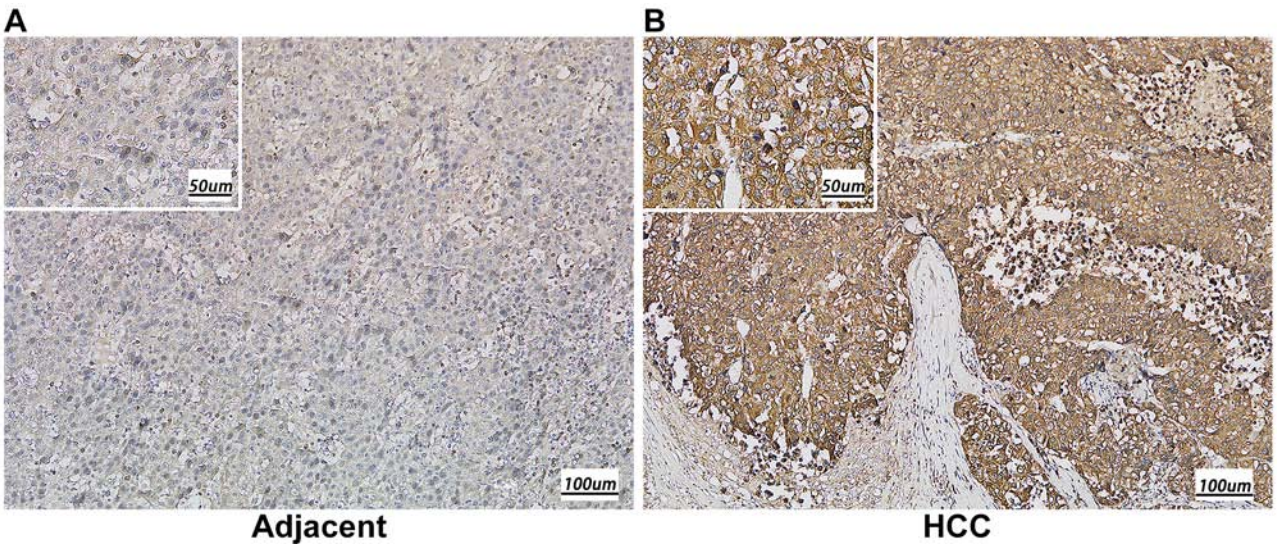

Figure 2. KIAA1522 protein expression levels detected via immunohistochemistry. KIAA1522 expression in (A) adjacent and (B) HCC tissues. HCC, hepatocellular carcinoma.

3 years post-surgery. The review included liver function, AFP, abdominal ultrasound, chest radiograph, enhanced CT or MRI and needle biopsy, among other examinations. Recurrence was defined by imaging studies, or biopsy-confirmed new lesions in the liver or outside the liver. Disease-free survival (DFS) time was defined as the time from the date of surgery to the time of recurrence or follow-up. Overall survival (OS) time was defined as the time from the date of surgery to the time of mortality or follow-up. Both DFS and OS were calculated on a monthly basis, and the follow-up deadline was December 2017.

Statistical analysis. Statistical analysis was performed using SPSS software (version 24.0; IBM Corp.). The χ^2, continuity correction χ^2 and Fisher's exact probability tests were used to determine the associations between KIAA1522 expression and clinicopathological characteristics in HCC. The independent sample t-test was used to analyze the difference between KIAA1522 expression in adjacent and HCC tissues. Kaplan-Meier survival analysis and the log-rank test were used to compare the association between KIAA1522 expression and postoperative recurrence and survival in patients with HCC. The Cox proportional hazard model was used to select variables by the forward logistic regression method for univariate and multivariate analyses of recurrence and survival. $P<0.05$ was considered to indicate a statistically significant difference.

Results

KIAA1522 expression in HCC and adjacent normal tissues. Analysis of the Chen Liver dataset within the Oncomine database demonstrated that KIAA1522 mRNA expression

Table I. KIAA1522 protein expression in HCC and adjacent normal tissues.

	KIAA1522			
Tissue	Negative, n (%)	Positive, n (%)	Total, n	P-value
HCC	8 (10.1)	71 (89.9)	79	<0.01[a]
Adjacent normal	67 (84.8)	12 (15.2)	79	

[a]Paired χ^2 test; P<0.05. HCC, hepatocellular carcinoma.

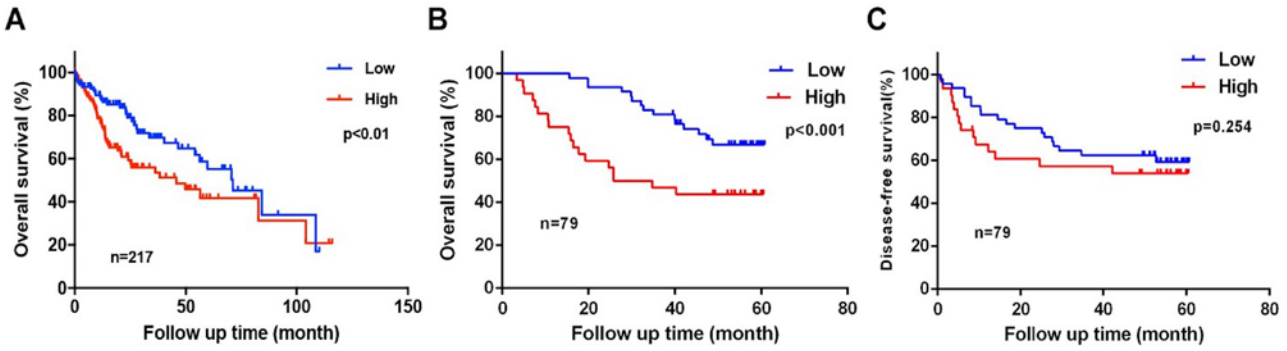

Figure 3. Kaplan-Meier curves depicting the association between KIAA1522 expression levels and postoperative prognosis. (A) OS rates of patients with low and high KIAA1522 expression levels in the OncoLnc database. (B) OS rates of patients with low and high KIAA1522 expression levels in HCC. (C) Disease-free survival rates of patients with low and high KIAA1522 expression levels in HCC. HCC, hepatocellular carcinoma; OS, overall survival.

was significantly higher in HCC tissues compared with that in adjacent normal liver tissues (P<0.01; Fig. 1A). This observation was validated in the patient cohort assessed in the present study via RT-qPCR analysis of the matched HCC and adjacent non-tumor tissues, which also demonstrated that KIAA1522 mRNA expression was significantly higher in HCC tissues compared with that in adjacent normal tissues (P<0.001; Fig. 1B). Subsequently, immunohistochemistry was performed to detect KIAA1522 expression in 79 paraffin-embedded human HCC tissues, including 50 cases of clinical stage I, 19 cases of clinical stage II and 10 cases of clinical stage IIIa. The results indicated that KIAA1522 protein expression was predominantly localized in the cytoplasm, and the high expression rate of KIAA1522 protein was significantly higher in the HCC tissues (89.9%) compared with the adjacent normal tissues (15.2%) (P<0.01; Fig. 2 and Table I).

Association between KIAA1522 expression and clinicopathological characteristics in HCC. The association between KIAA1522 expression and the clinicopathological characteristics in HCC are presented in Table II. Notably, KIAA1522 expression was not demonstrated to be significantly associated with any of the assessed clinicopathological characteristics in HCC, including age, sex, Child-Pugh classification, tumor number and size, degree of differentiation and clinical stage.

Association between KIAA1522 expression and postoperative prognosis in HCC. The KIAA1522 dataset, including 217 TCGA primary liver cancer datapoints, was downloaded from the OncoLnc database and a survival curve was generated, which demonstrated that high KIAA522 expression was closely associated with low OS time of patients with HCC (P<0.01; Fig. 3A). Survival analysis was performed on the 79 clinical cases from the present study using the Kaplan-Meier method and log-rank test, in order to determine the association between KIAA1522 expression in HCC and OS time following surgery. The results demonstrated that the mean OS time was 49.700 months (95% CI, 45.320-53.950). The mean OS time in the low expression group was 59.200 months (95% CI, 57.405-60.907), while the mean OS time in the high expression group was 36.600 months (95% CI, 27.607-43.636). The mean OS time in the high expression group was significantly decreased compared with the low expression group (P<0.001; Fig. 3B).

Database analysis failed to provide a survival curve associated with recurrence of KIAA1522 expression in HCC. Thus, KIAA1522 expression in HCC was compared with postoperative recurrence, as described above. The results demonstrated that the overall mean DFS time was 41.100 months (95% CI, 35.691-46.480). The mean DFS time in the low expression group was 43.400 months (95% CI, 36.994-49.875), while the mean DFS time in the high expression group was 37.241 months (95% CI, 35.691-46.480). No significant difference in DFS time was observed between the two groups (P=0.254 Fig. 3C).

Univariate and multivariate Cox regression analyses of prognosis. Univariate and multivariate analyses of clinicopathological characteristics with regard to OS are presented in Table III. Factors that may influence the OS time of patients with HCC were included in the univariate Cox regression analysis. The results demonstrated that TNM staging, KIAA1522 expression, tumor size, surgical resection, hepatic capsule

Table II. Association between KIAA1522 expression and clinicopathological characteristics in patients with hepatocellular carcinoma.

Characteristic	Patient, n	KIAA1522, n (%)		χ^2	P-value
		High	Low		
Sex				0.395	0.530[a]
Male	64	37 (57.8)	27 (42.2)		
Female	15	5 (33.3)	10 (66.7)		
Age, years				0.750	0.386[a]
<50	16	8 (50.0)	8 (50.0)		
≥50	63	39 (61.9)	24 (38.1)		
Alcohol consumption				0.069	0.793[a]
Yes	21	13 (61.9)	8 (38.1)		
No	58	34 (58.6)	24 (41.4)		
HBV infection				0.000	>0.999[b]
Yes	74	44 (59.5)	30 (40.5)		
No	5	3 (60.0)	2 (40.0)		
Liver cirrhosis				0.002	0.965[a]
Yes	64	38 (59.4)	26 (40.6)		
No	15	9 (60.0)	6 (40.0)		
AFP level, ng/l				2.005	0.157[a]
≤400	54	35 (64.8)	19 (35.2)		
>400	25	12 (48.0)	13 (52.0)		
Child-Pugh grade					>0.999[c]
A	78	46 (59.0)	32 (41.0)		
B	1	0 (0.0)	1 (100.0)		
Tumor number				0.000	>0.999[b]
Single	69	41 (59.4)	28 (40.6)		
Multiple	10	6 (60.0)	4 (40.0)		
Tumor size, cm				1.114	0.286[a]
≤5	57	36 (63.2)	21 (36.8)		
>5	22	11 (50.0)	11 (50.0)		
Pathological differentiation				0.000	0.242[b]
High	3	2 (66.7)	1 (33.3)		
Middle and low	76	45 (59.2)	31 (40.8)		
Microvascular tumor thrombus				0.264	0.607[b]
Yes	27	15 (55.6)	12 (44.4)		
No	52	32 (61.5)	20 (38.5)		
Capsule invasion				3.174	0.075[a]
Yes	50	26 (52.0)	24 (48.0)		
No	29	21 (72.4)	8 (27.6)		
Surgical resection range				2.420	0.120[a]
Liver lobe	41	21 (51.2)	20 (48.8)		
Liver segment	38	26 (68.4)	12 (31.6)		
TNM stage				0.005	0.943[a]
I+II	49	29 (59.2)	20 (40.8)		
IIIa	30	18 (60.0)	12 (40.0)		

[a]Pearson's χ^2 test; [b]Continuity correction by χ^2 test; [c]Fisher's exact test. HBV, hepatitis B virus; AFP, α-fetoprotein; TNM, Tumor-Node-Metastasis.

invasion and vascular tumor thrombus had a significant impact on postoperative survival time (all P<0.05). Conversely, sex, age and AFP levels, among others, were demonstrated to have no significant effect on postoperative survival time in patients

Table III. Univariate and multivariate analyses of clinicopathological characteristics with regard to overall survival.

	Univariate		Multivariate	
Characteristic	HR (95% CI)	P-value	HR (95% CI)	P-value
Sex (female vs. male)	2.478 (0.577-10.644)	0.222		
Age (<50 vs. ≥50 years)	0.575 (0.223-1.480)	0.251		
Alcohol consumption	1.492 (0.602-3.699)	0.387		
HBV infection	1.356 (0.182-10.103)	0.655		
TBIL level (≤22 vs. >22 μmol/l)	1.067 (0.359-3.172)	0.907		
ALB level (<35vs. ≥35 g/l)	1.454 (0.338-8.102)	0.615		
ALT level (≤60 vs. >60 μ/l)	1.688 (0.681-4.183)	0.258		
AST level (≤42 vs. >42 μ/l)	1.883 (0.760-4.668)	0.172		
PLT level (<100 vs. ≥100 l)	5.188 (0.696-30.166)	0.108		
Liver cirrhosis	0.767 (0.281-2.094)	0.604		
AFP level (≤400 vs. >400 ng/l)	1.833 (0.772-4.355)	0.170		
Child-Pugh grade (A vs. B)	0.049 (0.020-5.284)	0.710		
Tumor number (single vs. multiple)	1.704 (0.573-5.068)	0.338		
Tumor size (≤5 vs. >5 cm)	4.603 (1.934-10.953)	0.001[a]	3.070 (1.107-8.512)	0.031[a]
Differentiation (high vs. middle and low)	2.506 (0.526-5.812)	0.517		
Surgical resection range (liver lobe vs. segment)	2.876 (0.967-8.566)	0.040[a]		
Capsule invasion	2.813 (1.090-7.255)	0.032[a]		
Vascular tumor thrombus	6.541 (2.363-18.101)	0.001[a]		
TNM stage (I+II vs. IIIa)	3.354 (1.388-8.102)	0.007[a]	3.116 (0.904-10.740)	0.047[a]
KIAA1522 (low vs. high)	13.294 (3.898-45.344)	0.001[a]	19.413 (5.197-72.500)	0.001[a]

[a]P<0.05. HR, hazard ratio; CI, confidence interval; HBV, hepatitis B virus; TBIL, total bilirubin; ALB, albumin; ALT, alanine aminotransferase; AST, aspartate aminotransferase; PLT, platelet; AFP, α-fetoprotein; TNM, Tumor-Node-Metastasis.

with HCC (all P>0.05). Subsequently, multivariate Cox regression analysis was performed on the clinicopathological characteristics which demonstrated statistical significance in the univariate analysis. The results demonstrated that surgical resection range, hepatic capsule invasion and vascular tumor thrombus did not affect the OS time of patients with HCC. However, tumor size >5 cm (P=0.031), TNM clinical stage IIIa (P=0.047) and high KIAA1522 expression (P=0.001) were demonstrated to be independent risk factors affecting postoperative OS time (all P<0.05). According to the hazard ratio (HR) values, these factors were considered to have an impact on the OS time in the following order: High KIAA1522 expression, TNM clinical stage IIIa and tumor size >5 cm (Table III).

Discussion

Currently, a variety of methods, including hepatectomy, liver transplantation, radiofrequency ablation, transarterial chemoembolization and sorafenib are available for the treatment of patients with HCC at different stages (1,24). However, the long-term prognosis of HCC remains unsatisfactory. Thus, elucidating the underlying molecular mechanism of HCC and identifying essential molecules can help improve the early prediction of prognosis, and aid in the development of novel treatment strategies. Examining the expression or the prognostic value of a specific gene or panel of genes with key biological functions is one approach to identifying essential molecules. Additionally, high-throughput technology can identify biomarkers at the genomic or proteomic level (25,26), and online expression profile datasets can also be used to search for potential biomarkers (1). According to several databases, KIAA1522 expression is elevated in HCC tissues and is closely associated with the prognosis of HCC, which prompted its analysis in the present study.

Previous studies have reported that the newly cloned gene, KIAA1522, is overexpressed in several types of human cancer (18,27-29). However, the association between aberrant KIAA1522 expression and malignant tumor growth remains unclear. Xie *et al* (18) reported that KIAA1522 is overexpressed in oesophageal squamous cell carcinoma (ESCC), and the overexpression of KIAA1522 can enhance the malignant proliferative capacity and anoikis resistance by activating the ERK signaling pathway to promote tumor formation and progression. This indicates that aberrant KIAA1522 expression plays a carcinogenic role in ESCC. Furthermore, Li *et al* (28) demonstrated that KIAA1522 is a direct target of miR-125b-5p in breast cancer and is involved in tumor cell proliferation, colony formation, cell migration and cell invasion. Liu *et al* (29) indicated that the high KIAA1522 expression can be used as an independent biomarker for predicting poor survival and platinum resistance in patients with non-small cell lung cancer. KIAA1522 is involved in oncogenic KRAS signaling in lung cancer cells and may be a novel target for lung

cancer treatment. These studies demonstrate that KIAA1522 plays a key role in the proliferation, metastasis and invasion of various cancer cells. Although KIAA1522 is overexpressed in several tumor tissues, to the best of our knowledge, its association with HCC remains unknown.

The present study applied bioinformatics technology, using the Oncomine and OncoLnc databases, to determine the association between KIAA1522 expression and clinical prognosis. The results demonstrated that KIAA1522 mRNA expression was significantly higher in HCC tissues compared with that in adjacent normal tissues. Furthermore, the high KIAA1522 expression group exhibited a significantly lower OS time compared with the low expression group. Subsequently, immunohistochemical staining was performed to detect KIAA1522 protein expression levels in the 79 HCC and adjacent normal tissue samples, while RT-qPCR was performed to determine KIAA1522 mRNA expression levels. The results demonstrated that both KIAA1522 protein and mRNA expression levels were significantly higher in the HCC tissues compared with those in the adjacent normal tissues. Taken together, these results indicate that KIAA1522 is upregulated in HCC at both the molecular and protein levels.

Clinical data from 79 patients with HCC was analyzed to determine whether KIAA1522 expression levels were associated with the relevant clinicopathological characteristics. The results demonstrated that KIAA1522 protein expression in HCC was not associated with age, sex, alcoholism, cirrhosis, Child-Pugh classification, number and size of tumors, degree of differentiation and clinical stage. However, this may be inaccurate due to the small sample size used in the present study, thus further studies with larger sample sizes are required for verification. The association between KIAA1522 expression and postoperative prognosis in HCC was assessed during the follow-up period, which demonstrated that the OS time of patients in the high KIA1522 expression group was significantly lower compared with that of patients in the low expression group. No significant difference was observed for DFS time and KIAA1522 expression, indicating that KIAA1522 expression was not associated with postoperative recurrence. This may be due to the small sample size used in the present study and untimely patient postoperative review, thus future studies will aim to increase the sample size to verify this view. The association between KIAA1522 expression and postoperative prognosis, and the risk factors affecting survival and recurrence following hepatectomy were also assessed. Univariate and multivariate Cox regression analyses demonstrated that high KIAA1522 expression in HCC was significantly and positively associated with the short-term survival of patients. This suggests that KIAA1522 may play a key role in the occurrence and development of HCC and may serve as a potential molecular marker for predicting the prognosis of patients with HCC. Further investigation on the association between KIAA1522 expression and postoperative survival in HCC may provide a novel and effective approach to improve the OS time of patients with HCC.

Additionally, tumor size and TNM staging were demonstrated to be independent risk factors affecting OS time. A previous study combined a variety of liver cancer staging systems and clinicopathological factors to predict the prognosis of liver cancer (17). Based on the results of the present study, it is speculated that late TNM stages lead to a large tumor burden, which may result in micro-hepatic metastasis of HCC prior to surgery, thus affecting prognosis. This also suggests that early detection, diagnosis and surgery can effectively decrease the clinical stage of preoperative tumors, which may be an effective means to prevent postoperative recurrence and improve prognosis. The results of the present study demonstrated that a tumor size >5 cm may be an independent risk factor for the OS time of patients with HCC following surgery. This is consistent with previous finding that indicated that the prognosis patients with large liver tumors were worse than that of patients with small liver tumors (30). Zhou *et al* (31) reported that the survival rate of patients with HCC, with tumors <5 cm following surgery, is significantly higher compared with patients with tumors >5 cm. At the same time, when dissecting the pathological specimens during liver transplantation, it was demonstrated that the larger the tumor is, the higher the vascular invasion and the degree of metastasis. This finding is consistent with the results of the present study. However, the present study failed to demonstrate a significant association between KIAA1522 expression levels and TNM stage and tumor size in the HCC tissues, which may be due to the small sample size used.

Overall, the results of the present study demonstrated that KIAA1522 expression was associated with postoperative prognosis in HCC, and overexpression of KIAA1522 is associated with a poor prognosis in patients with HCC. The results of the present study demonstrated that TNM staging, tumor size and high KIAA1522 expression were all independent risk factors for postoperative survival, suggesting that KIAA1522 may serve as a novel molecular marker for predicting the survival of patients with HCC. Based on bioinformatics analysis, this preliminary small sample-level study was performed on the postoperative prognosis of 79 patients with HCC in The Affiliated Hospital of Qingdao University in 2013. A major limitation of the present study is use of a small sample size, which may have introduced bias. Thus, future studies will use larger sample sizes, gather more relevant data and aim to perform an in-depth investigation of the underlying molecular mechanism of KIAA1522 in HCC.

Acknowledgements

Not applicable.

Funding

The present study was funded by the Key Research and Development Plan of Shandong Province (grant no. 2018GSF118233), the Science and Technology Plan of Qingdao City Shinan District (grant no. 2018-4-018-YY) and the Natural Science Foundation of Shandong Province (grant no. ZR2012HQ039).

Availability of data and materials

The datasets used and/or analyzed during the present study are available from the corresponding author upon reasonable request.

Authors' contributions

CS and BH contributed to the study design. MZ, YXi, JY and ZC acquired the data. YXu, MZ and YXi performed the experiments. YXu drafted the initial manuscript. All authors read and approved the final version of the manuscript.

Ethics approval and consent to participate

The present study was approved by the Ethics Committee of The Affiliated Hospital of Qingdao University and written informed consent was obtained from all patients prior to the study start.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Llovet JM, Schwartz M and Mazzaferro V: Resection and liver transplantation for hepatocellular carcinoma. Semin Liver Dis 25: 181-200, 2005.
2. Torre LA, Bray F, Siegel RL, Ferlay J, Lortet-Tieulent J and Jemal A: Global cancer statistics, 2012. CA Cancer J Clin 65: 87-108, 2015.
3. Grazi GL, Ercolani G, Pierangeli F, Del Gaudio M, Cescon M, Cavallari A and Mazziotti A: Improved results of liver resection for hepatocellular carcinoma on cirrhosis give the procedure added value. Ann Surg 234: 71-78, 2001.
4. El-Serag HB and Rudolph KL: Hepatocellular carcinoma: Epidemiology and molecular carcinogenesis. Gastroenterology 132: 2557-2576, 2007.
5. Feng RM, Zong YN, Cao SM and Xu RH: Current cancer situation in China: Good or bad news from the 2018 Global Cancer Statistics? Cancer Commun (Lond) 39: 22, 2019.
6. Jemal A, Bray F, Center MM, Ferlay J, Ward E and Forman D: Global cancer statistics. CA Cancer J Clin 61: 69-90, 2011.
7. Desjardins LA: Hepatocellular carcinoma. Clin J Oncol Nurs 6: 107-108, 2002.
8. Maluccio M and Covey A: Recent progress in understanding, diagnosing, and treating hepatocellular carcinoma. CA Cancer J Clin 62: 394-399, 2012.
9. Wang L, Yao M, Dong Z, Zhang Y and Yao D: Circulating specific biomarkers in diagnosis of hepatocellular carcinoma and its metastasis monitoring. Tumour Biol 35: 9-20, 2014.
10. Contratto M and Wu J: Targeted therapy or immunotherapy? Optimal treatment in hepatocellular carcinoma. World J Gastrointest Oncol 10: 108, 2018.
11. Ji F, Fu SJ, Shen SL, Zhang LJ, Cao QH, Li SQ, Peng BG, Liang LJ and Hua YP: The prognostic value of combined TGF-β1 and ELF in hepatocellular carcinoma. BMC Cancer 15: 116, 2015.
12. Ng KK, Lo CM, Liu CL, Poon RT, Chan SC and Fan ST: Survival analysis of patients with transplantable recurrent hepatocellular carcinoma: Implications for salvage liver transplant. Arch Surg 143: 68-74, 2008.
13. Kim DY, Paik YH, Ahn SH, Youn YJ, Choi JW, Kim JK, Lee KS, Chon CY and Han KH: PIVKA-II is a useful tumor marker for recurrent hepatocellular carcinoma after surgical resection. Oncology 72 (Suppl 1): 52-57, 2007.
14. Fu SJ, Qi CY, Xiao WK, Li SQ, Peng BG and Liang LJ: Glypican-3 is a potential prognostic biomarker for hepatocellular carcinoma after curative resection. Surgery 154: 536-544, 2013.
15. Guo Y, Liang X, Lu M, Weng T, Liu Y and Ye X: Mammalian target of rapamycin as a novel target in the treatment of hepatocellular carcinoma. Hepatogastroenterology 57: 913-918, 2010.
16. Marrero JA, Feng Z, Wang Y, Nguyen MH, Befeler AS, Roberts LR, Reddy KR, Harnois D, Llovet JM, Normolle D, *et al*: Alpha-fetoprotein, des-gamma carboxyprothrombin, and lectin-bound alpha-fetoprotein in early hepatocellular carcinoma. Gastroenterology 137: 110-118, 2009.
17. Cristea CG, Gheonea IA, Săndulescu LD, Gheonea DI, Ciurea T and Purcarea MR: Considerations regarding current diagnosis and prognosis of hepatocellular carcinoma. J Med Life 8: 120-128, 2015.
18. Xie ZH, Yu J, Shang L, Zhu YQ, Hao JJ, Cai Y, Xu X, Zhang Y and Wang MR: KIAA1522 overexpression promotes tumorigenicity and metastasis of esophageal cancer cells through potentiating the ERK activity. Onco Targets Ther 10: 3743-3754, 2017.
19. Chen X, Cheung ST, So S, Fan ST, Barry C, Higgins J, Lai KM, Ji J, Dudoit S, Ng IO, *et al*: Gene expression patterns in human liver cancers. Mol Biol Cell 13: 1929-1939, 2002.
20. Edge SB and Compton CC: The American Joint Committee on Cancer: The 7th Edition of theAJCC cancer staging manual and the future of TNM. Ann Surg Oncol 17: 1471-1474, 2010.
21. Maiolino P, Restucci B, Papparella S, Paciello O and De Vico G: Correlation of nuclear morphometric features with animal and Human World Health Organization international histological classifications of canine spontaneous seminomas. Vet Pathol 41: 608-611, 2004.
22. Durand FO and Valla D: Assessment of the prognosis of cirrhosis: Child-Pugh versus MELD. J Hepatol 42 (Suppl): S100-S107, 2005.
23. Livak KJ and Schmittgen TD: Analysis of relative gene expression data using real-time quantitative PCR and the 2(-Delta Delta C(T)) Method. Method 25: 402-408, 2001.
24. Franssen B, Alshebeeb K, Tabrizian P, Marti J, Pierobon ES, Lubezky N, Roayaie S, Florman S and Schwartz ME: Differences in surgical outcomes between hepatitis B- and hepatitis C-related hepatocellular carcinoma: A retrospective analysis of a single North American center. Ann Surg 260: 650-658, 2014.
25. Zhu CQ, Ding K, Strumpf D, Weir BA, Meyerson M, Pennell N, Thomas RK, Naoki K, Ladd-Acosta C, Liu N, *et al*: Prognostic and predictive gene signature for adjuvant chemotherapy in resected non-small-cell lung cancer. J Clin Oncol 28: 4417-4424, 2010.
26. Zhu CQ, Pintilie M, John T, Strumpf D, Shepherd FA, Der SD, Jurisica I and Tsao MS: Understanding prognostic gene expression signatures in lung cancer. Clin Lung Cancer 10: 331-340, 2009.
27. Nagase T, Kikuno R, Ishikawa K, Hirosawa M and Ohara O: Prediction of the coding sequences of unidentified human genes. XVII. The Complete Sequences of 100 New cDNA clones from brain which code for large proteins in vitro. DNA Res 7: 143-150, 2000.
28. Li Y, Wang Y, Fan H, Zhang Z and Li N: miR-125b-5p inhibits breast cancer cell proliferation, migration and invasion by targeting KIAA1522. Biochem Biophys Res Commun 504: 277-282, 2018.
29. Liu YZ, Yang H, Cao J, Jiang YY, Hao JJ, Xu X, Cai Y and Wang MR: KIAA1522 is a novel prognostic biomarker in patients with non-small cell lung cancer. Sci Rep 6: 24786, 2016.
30. Takamoto T, Sano K, Hashimoto T, Ichida A, Shimada K, Maruyama Y and Makuuchi M: Practical contribution of virtual hepatectomy for colorectal liver metastases: A propensity-matched analysis of clinical outcome. J Gastrointest Surg 22: 2037-2044, 2018.
31. Zhou XD, Tang ZY, Ma ZC, Fan J, Wu ZQ, Qin LX, Zhou J, Yu Y, Sun HC and Qiu SJ: Twenty-year survivors after resection for hepatocellular carcinoma-analysis of 53 cases. J Cancer Res Clin Oncol 135: 1067-1072,