Original Article

Overexpression of SPHK1 associated with targeted therapy resistance in predicting poor prognosis in renal cell carcinoma

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Background: Sphingosine kinase 1 (SPHK1) is a key enzyme that catalyzes the phosphorylation of sphingosine. Recent studies reported SPHK1 to be associated with renal cell carcinoma (RCC) progression by inducing targeted therapy resistance. However, the expression and the clinical significance of SPHK1 on RCC in those having received targeted therapy have not been elucidated. The present study explored the expression of SPHK1 in RCC tissues from targeted therapy recipients, the correlation of SPHK1 with clinicopathological parameters, and the effect of SPHK1 on RCC patient prognosis.

Methods: Differential gene expression analysis of RCC treated with and without targeted therapy was performed. The correlations of SPHK1 expression with clinical parameters of RCC were examined. Gene set enrichment analysis (GSEA) was performed to clarify the potential role of SPHK1 associated with targeted therapy resistance. The value of SPHK1 as a diagnostic marker for RCC was also evaluated. The Kaplan-Meier method was applied to analyze the correlation between SPHK1 expression and patient survival rate by using the clinical data from patients with RCC.

Results: Significant overexpression of SPHK1 was detected in RCC treated with targeted therapy. SPHK1 expression was closely correlated with RCC progression-related clinicopathological parameters. Therefore, elevated SPHK1 could effectively diagnose RCC and distinguish RCC with an advanced clinical stage and a high pathological grade. SPHK1 was associated with the stemness of RCC cells via the activation of the Wnt, Hedgehog, or Notch signaling pathways in targeted drug-treated or untreated RCC. Survival analysis of a large cohort of RCC samples indicated overexpression of SPHK1 to be inversely correlated with the overall and disease-free survival of patients with RCC.

Conclusions: Our study indicated that SPHK1 associated with targeted therapy resistance could serve as a potential prognostic marker and a valuable biomarker of response to angiogenic agents in RCC.

Keywords: Renal cell carcinoma (RCC); sphingosine kinase 1 (SPHK1); targeted therapy resistance; prognosis

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Introduction

Renal cell carcinoma (RCC) is the sixth most frequently diagnosed cancer in male patients and the ninth in female patients (1). RCC also represents the most lethal urologic malignancy, causing an estimated 175,098 deaths globally in 2018 (2). Localized RCC can be treated with partial or radical nephrectomy with favorable outcomes (3). However, about 30% of the patients who receive surgical treatment eventually develop metastasis that is almost incurable (4).

Based on recent advances in molecular mechanisms driving RCC progression, targeted therapy, including tyrosine kinase inhibitors, mammalian target of rapamycin inhibitors, and anti-vascular endothelial growth factor (VEGF) monoclonal antibodies, has been established and widely applied in the treatment of advanced RCC. Targeted therapy has been reported to significantly prolong the overall survival time of patients with metastatic RCC from 18.3 to 26.4 months and improve the quality of life (5). However, patients with advanced RCC eventually become resistant to targeted therapy. Mechanisms underlying the resistance of targeted therapy are unclear, and biomarkers indicating treatment efficacy are still lacking.

A recent study has shown that sphingolipid metabolism plays an important role in sunitinib resistance (6). Sphingosine (Sph) is associated with cell growth arrest and apoptosis and is a ubiquitous component of the cell membrane. Sph can be phosphorylated by sphingosine kinase (SPHK1or SPHK2) to form sphingosine-1-phosphate (S1P), which is associated with the suppression of apoptosis (7). Notably, overexpression of SPHK1 was reported to induce overproduction of S1P, which is a vital step in sunitinib resistance of RCC (8). Therefore, the role of SPHK1 in the targeted therapy resistance of RCC has received considerable attention (9). However, the expression and the clinical significance of SPHK1 on patients with RCC receiving targeted therapy have not been elucidated. In particular, the value of SPHK1 as a diagnostic marker in RCC as well as the impact of SPHK1 on the survival of patients with RCC is unclear. The present study explores the functional role as well as the clinical application of SPHK1 in RCC and concludes that SPHK1 is a potential diagnostic and prognostic marker that predicts poor outcomes of RCC. We present the following article in accordance with the REMARK reporting checklist (available at https://tcr.amegroups.com/article/view/10.21037/tcr-22-417/rc).

Highlight box

Key findings

- SPHK1 was overexpressed in RCC treated with targeted therapy. SPHK1 expression was correlated with RCC progression–related clinicopathological parameters. Elevated SPHK1 could effectively diagnose RCC and distinguish RCC with an advanced clinical stage and a high pathological grade. Overexpression of SPHK1 was inversely correlated with overall and disease-free survival of patients with RCC.

What is known and what is new?

- SPHK1 is associated with RCC progression by inducing targeted therapy resistance.
- SPHK1 expression could distiguish RCC with an advanced clinical stage as well as a high pathological grade and predict the outcomes of patients with RCC.

What is the implication, and what should change now?

- SPHK1 could serve as a potential diagnostic marker and a valuable prognostic marker in RCC.

Methods

Data acquisition and preprocessing

RNA-sequencing–based messenger RNA (mRNA) expression data and clinical parameters from 533 RCC samples containing 72 matched normal kidney tissues were downloaded from The Cancer Genome Atlas (TCGA) database (http://cancergenome.nih.gov/). Among the 533 patients with RCC, 52 patients received targeted therapy; detailed information on these patients is shown in *Table 1*. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013).

Differential gene expression analysis

Differential gene expression analysis between benign and tumor tissues and an analysis of the correlations between SPHK1 expression and clinical parameters were performed. A Student 2-tailed *t*-test was used to compare the differences between the 2 groups.

Receiver operator characteristic (ROC) curve analysis

ROC curves were drawn, and the area under the curve (AUC) was calculated to detect the optimal cutoff value that yielded the highest total accuracy for discriminating

Table 1 SPHK1 expression in patients with RCC who received targeted therapy in TCGA data set

Sample ID	Targeted drug name	Status: 1, dead; 0, alive	Follow-up time after treatments (days)	Relative SPHK1 mRNA level (RNA-seq)
TCGA-B2-5639	Torisel	0	343	4.6125
TCGA-CJ-6028	Interferon, sunitinib, sorafenib	1	1,584	4.7953
TCGA-B0-5115	Afinitor (everolimus)	0	586	4.8949
TCGA-CW-5585	Sunitinib	0	46	5.1595
TCGA-AK-3436	Sunitinib	0	1,968	5.1768
TCGA-CJ-4644	Intron A, capecitabine, Avastin, Tarceva	1	174	5.1897
TCGA-BP-4165	Sunitinib	0	450	5.3496
TCGA-B0-5694	Pazopanib	1	52	5.4014
TCGA-CZ-5462	Sunitinib	1	296	5.59
TCGA-B0-5094	Torisel	1	209	5.6235
TCGA-CZ-5456	Pazopanib	0	434	5.7285
TCGA-BP-4161	Sunitinib	0	6	5.826
TCGA-BP-4329	Interferon, temsirolimus	1	655	5.89
TCGA-CJ-5681	Il-2, gemcitabine, Avastin	1	446	6.0614
TCGA-CJ-4871	Alpha, interferon	0	2,325	6.0802
TCGA-BP-5189	Temsirolimus, bevacizumb	1	162	6.2733
TCGA-CZ-5461	Sunitinib	1	285	6.2835
TCGA-CJ-5676	Pazopanib	0	2,507	6.3162
TCGA-CW-5590	Sunitinib	1	847	6.3229
TCGA-CZ-5464	Pazopanib, sunitinib	0	85	6.7428
TCGA-CJ-4875	Nexavaar	0	1,270	6.7605
TCGA-BP-5178	Sorafenib	1	1,516	6.7752
TCGA-CZ-5454	Sunitinib	1	685	6.7937
TCGA-CZ-4861	Sorafenib-Nexavar	1	488	6.9014
TCGA-B8-4153	Pazopanib	0	167	7.0922
TCGA-BP-5009	Sunitinib, everolimus, bevacizumb, pazopannib	1	754	7.1455
TCGA-BP-4974	Sunitinib, sorafenib, gefitinib	1	164	7.1572
TCGA-BP-4169	Interferon, axitinib	1	665	7.2167
TCGA-CZ-4860	Sorafenib	1	152	7.3383
TCGA-A3-3317	Sorafenib	0	886	7.3697
TCGA-BP-4787	Sunitinib, sorafenib, temsirolimus	1	427	7.4124
TCGA-CJ-6033	Avastin, gemcitabine, INF	1	192	7.4247
TCGA-B8-5162	Sunitinib	0	79	7.4347
TCGA-CJ-4881	Temsirolimus	0	83	7.6278

Table 1 (*continued*)

Table 1 (*continued*)

Sample ID	Targeted drug name	Status: 1, dead; 0, alive	Follow-up time after treatments (days)	Relative SPHK1 mRNA level (RNA-seq)
TCGA-BP-4985	Sunitinib	1	645	7.6505
TCGA-CZ-5469	Sunitinib	1	723	7.7489
TCGA-CJ-4904	Nexavaar	0	1,497	7.7709
TCGA-BP-4342	Sorafenib	1	503	7.8796
TCGA-CJ-5680	Avastin, IL-2, Tarceva	1	715	7.9609
TCGA-BP-4804	Sunitinib	0	171	7.9621
TCGA-BP-4338	Sunitinib, sorafernib, everolimus	0	1,094	8.0024
TCGA-CJ-4890	Sorafenib, sunitinib, tipifarnib, interferon	0	2,016	8.1113
TCGA-CJ-4638	Gemictibine, 5-Flu, IL-2, Avastin	1	361	8.1354
TCGA-CJ-4869	Nexavaar, sunitinib	0	1,787	8.1495
TCGA-CJ-4888	Sunitinib	1	991	8.3866
TCGA-BP-4354	Sunitinib, sorafenib, temsirolimus, gefitinib	1	1,024	8.6894
TCGA-CJ-4868	Avastin, proleukin, gemcitabine	1	589	8.7285
TCGA-B0-4837	Tyrosine kinase inhibitor	1	159	8.9445
TCGA-CJ-4637	Temsirolimus, Roferon-a, Intron A, sunitinib, sorafenib, nab-rapamycin, Avastin, AZD, pazopanib, borzomib	1	2,180	8.9846
TCGA-CJ-4900	Tarceva, Avastin	1	1,366	9.2858
TCGA-CJ-4895	Tarceva, Avastin	1	1,155	9.5056
TCGA-B0-5107	Sunitinib	1	916	9.8113

SPHK1, sphingosine kinase 1; RCC, renal cell carcinoma; TCGA, The Cancer Genome Atlas; IL, interleukin.

between the different clinical classifications of recurrence or nonrecurrence.

Survival analysis

The Kaplan-Meier method was applied to analyze patient survival, and the log-rank test was used to determine the statistical significance between the 2 groups. X-tile software was used to generate an optimal cutoff point to dichotomize SPHK1 mRNA into high and low categories using a Monte Carlo P value <0.05.

Gene set enrichment analysis (GSEA) based on Kyoto Encyclopedia of Genes and Genomes pathway analysis

GSEA was performed using the curated gene sets (C2) of the Molecular Database version 4.0 on the Broad Institute website (http://www.Broad.Mit.Edu/gsea/).

Statistical analysis

All statistical analyses were performed with GraphPad Prism 7 (GraphPad Software, La Jolla, CA, USA). A P value <0.05 was considered statistically significant.

Results

The expression of SPHK1 in RCC tissues from targeted therapy recipients

Based on the analysis of gene expression profile data from TCGA database, RCC tissues expressed a significantly higher level of SPHK1 when compared with benign renal tissues (*Figure 1A*). We further evaluated the expression of

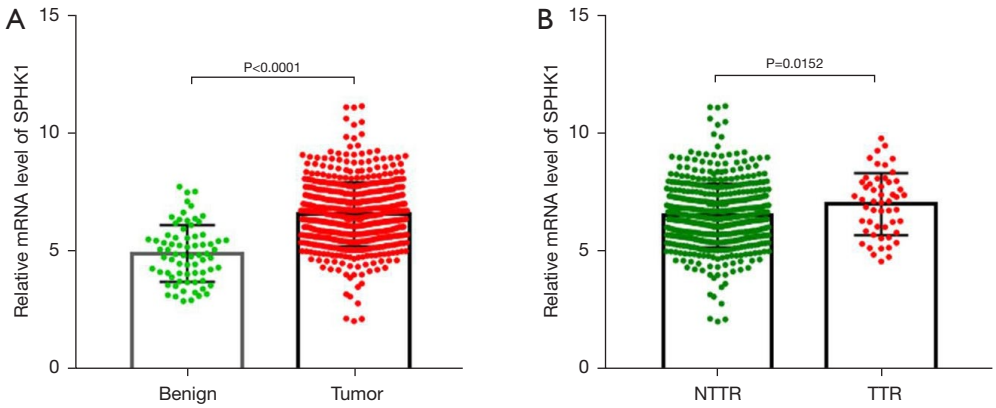

Figure 1 The expression of SPHK1 in RCC tissues from targeted therapy recipients. (A) Relative SPHK1 mRNA expression in benign kidney tissues (n=72) compared with that in RCC tissues (n=533) according to *t*-test (P<0.0001). (B) Relative SPHK1 mRNA expression in RCC tissue from NTTR (n=481) compared with that in TTR (n=52) according to *t*-test (P=0.0152). SPHK1, sphingosine kinase 1; RCC, renal cell carcinoma. NTTR, nontargeted therapy recipients; TTR, targeted therapy recipients.

SPHK1 in RCC tissues from 52 targeted therapy recipients. Notably, patients with RCC who received targeted therapy expressed a higher level of SPHK1 compared to other patients with RCC (*Table 1* and *Figure 1B*). These data indicated overexpression of SPHK1 in RCC tissues and an even higher level of SPHK1 in RCC tissues from targeted therapy recipients.

The association between SPHK1 expression and RCC progression

We further determined the correlations between the mRNA level of SPHK1 and the clinicopathological characteristics of patients with RCC. Elevated SPHK1 expression correlated with a higher RCC T stage (*Figure 2A*), positive lymph node metastasis (*Figure 2B*), positive distant metastasis (*Figure 2C*), clinical stage (*Figure 2D*), and pathological grade (*Figure 2E*). Notably, the SPHK1 level was higher in patients with RCC with recurrence compared with those without recurrence (*Figure 2F*). Collectively, these results indicated that SPHK1 expression was closely correlated with clinicopathological parameters (*Table 2*) and could be a potential biomarker to predict RCC progression.

Overexpression of SPHK1 associated with activation of signaling pathway regulating RCC cell stemness and drug resistance

GSEA was performed to delineate the functional role of SPHK1 associated with targeted therapy resistance. In RCC, elevated SPHK1 correlated with the stemness of RCC cells (*Figure 3A*). SPHK1 activated the Wnt, Hedgehog, and VEGF signaling pathways (*Figure 3B-3D*), which could enhance the stemness of RCC cells. Furthermore, we performed GSEA based on RNA-sequencing data from RCC treated with targeted therapy. Consistently, in targeted therapy–treated RCC, high expression of SPHK1 was also associated with the stemness of RCC cells (*Figure 3E*); moreover, SPHK1 was correlated with the activation of the Notch signaling pathway, VEGF signaling pathway, and cell cycle signaling pathway (*Figure 3F-3H*). These data indicated that the overexpression of SPHK1 could activate signaling pathways associated with cancer cell stemness.

The diagnostic value of SPHK1 in RCC

SPHK1 expression was closely associated with RCC development; therefore, we next evaluated the diagnostic value of SPHK1 in RCC. SPHK1 mRNA expression was an effective marker for differentiating RCC tissue from benign tissue (*Figure 4A*). Furthermore, higher SPHK1 expression could be a potential indicator for with higher T stage in patients with RCC (T3-4) rather than T1-2 (*Figure 4B*), positive lymph node metastasis (N1) rather than N0 (*Figure 4C*), positive distant metastasis (M1) rather than M0 (*Figure 4D*), higher clinical stage (S3-4) rather than S1-2 (*Figure 4E*), higher pathological grade (G3-4) rather than G1-2 (*Figure 4F*), and recurrence rather than without recurrence (*Figure 4G*). These findings indicated that

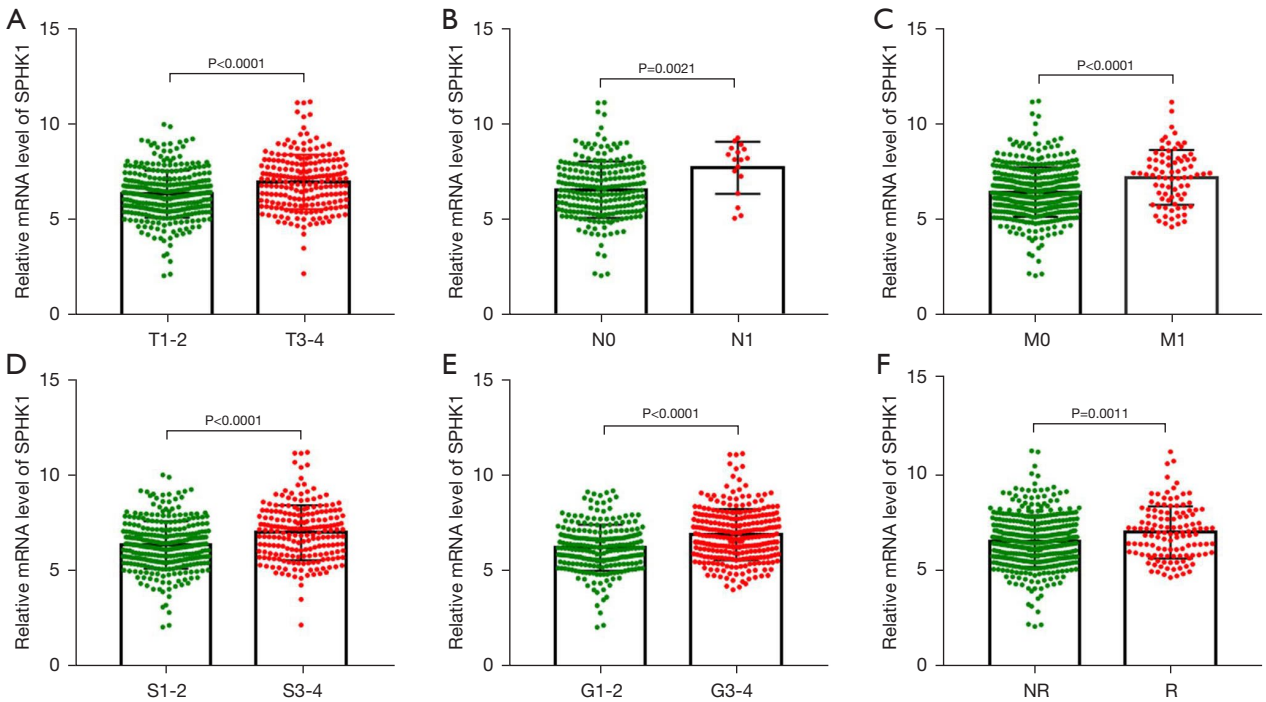

Figure 2 The association between SPHK1 expression and RCC progression. (A) SPHK1 mRNA expression in early T stage RCC (T1-2, n=342) compared with that in advanced T stage RCC (T3-4, n=191) according to *t*-test (P<0.0001). (B) Relative SPHK1 mRNA expression RCC with no lymph node metastasis (N0, n=240) compared with that in metastatic lymph node RCC (N1, n=16) according to *t*-test (P=0.0021). (C) Relative SPHK1 mRNA expression in localized RCC (M0, n=422) compared with that in metastatic RCC (M1, n=79) according to *t*-test (P<0.0001). (D) Relative SPHK1 mRNA expression in early-stage RCC (S1-2, n=324) compared with that in advanced-stage RCC (S3-4, n=207) according to *t*-test (P<0.0001). (E) SPHK1 mRNA expression in low-grade RCC (G1-2, n=243) compared with that in high-grade RCC (G3-4, n=282) according to *t*-test (P<0.0001). (F) Relative SPHK1 mRNA expression in nonrecurrent RCC (NR, n=409) compared with that in recurrent RCC (R, n=124) according to *t*-test (P=0.0011). SPHK1, sphingosine kinase 1; RCC, renal cell carcinoma.

Table 2 Correlations between SPHK1 (sphingosine kinase 1) expression and clinicopathological characteristics

Parameter	Number of patients	SPHK1 expression (No.)		P value
		Low (n=266)	High (n=267)	
Age (years)				
<60	244	124	120	0.5519
≥60	288	141	147	
Unknown	1	1	0	
Gender				
Female	188	106	82	0.0273
Male	345	160	185	

Table 2 (*continued*)

Table 2 (*continued*)

Parameter	Number of patients	SPHK1 expression (No.)		P value
		Low (n=266)	High (n=267)	
Pathological stage				
Stage I	267	161	106	<0.0001
Stage II	57	32	25	
Stage III	123	51	72	
Stage IV	84	29	55	
Unknown	2	0	2	
T stage				
T1	273	164	109	<0.0001
T2	69	28	41	
T3	180	72	108	
T4	11	2	9	
N stage				
N0	240	119	121	0.1161
N1	16	4	12	
Unknown	277	143	134	
M stage				
M0	422	232	190	<0.0001
M1	79	26	53	
Unknown	32	24	8	
Fuhrman grade				
G1	14	10	4	<0.0001
G2	229	137	92	
G3	206	96	110	
G4	76	18	58	
Unknown	8	5	3	
Targeted therapy				
No	481	247	234	0.0424
Yes	52	19	33	
Survival status				
Alive	358	206	152	<0.0001
Dead	175	60	115	
Recurrence status				
Nonrecurrence	409	215	194	0.0256
Recurrence	124	51	73	

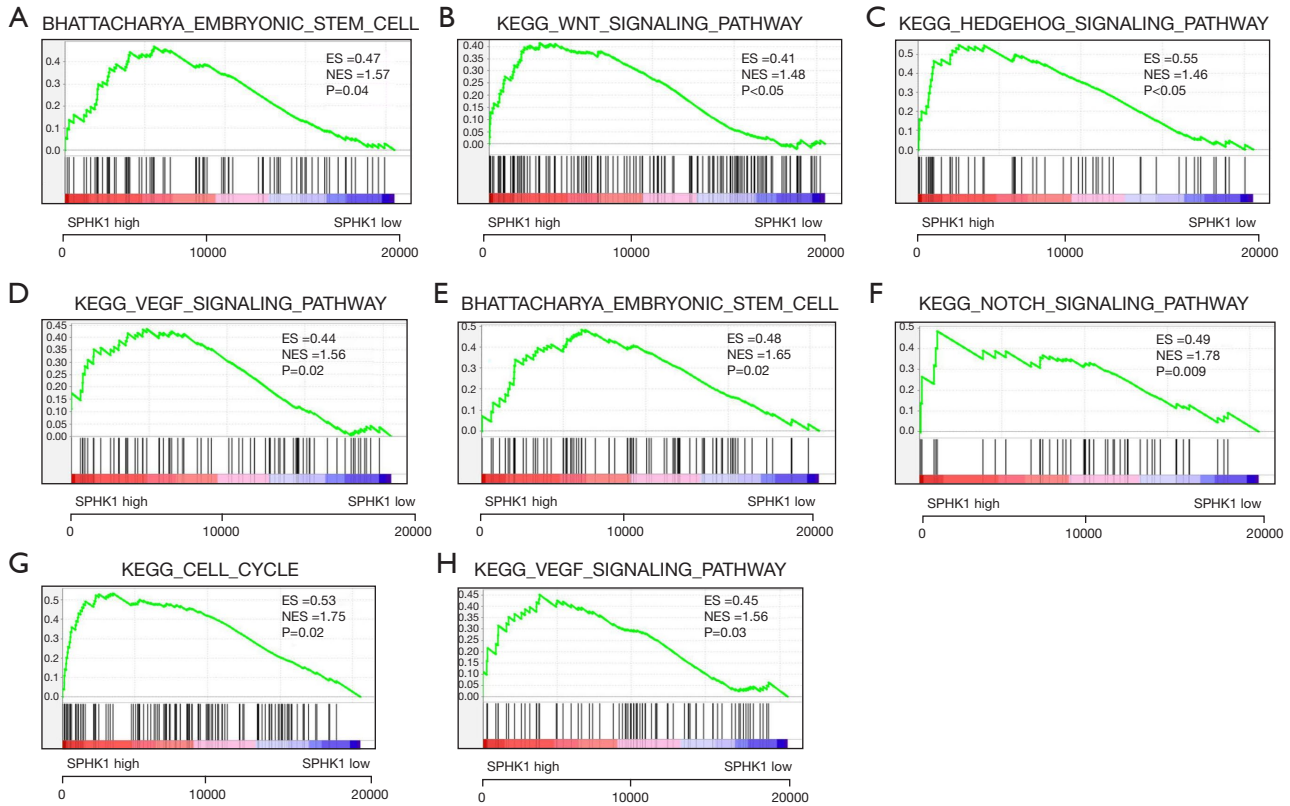

Figure 3 Overexpression of SPHK1 associated with activation of signaling pathway regulating RCC cell stemness and drug resistance. (A) GSEA showed that enrichment of stem cell signature in SPHK1 was positively correlated with the gene set from TCGA. (B) GSEA showed that high expression of SPHK1 was associated with activation of the Wnt signaling pathway. (C) GSEA showed that high expression of SPHK1 was associated with activation of the Hedgehog signaling pathway. (D) GSEA showed that high expression of SPHK1 was associated with activation of the VEGF signaling pathway. (E) GSEA showed that enrichment of the stem cell signature in SPHK1 was positively correlated with the gene set from TCGA. (F) GSEA showed that the high expression of SPHK1 was associated with activation of the Notch signaling pathway. (G) GSEA showed that high expression of SPHK1 was associated with activation of the cell cycle signaling pathway. (H) GSEA showed that high expression of SPHK1 was associated with activation of the VEGF signaling pathway. ES, enrichment score; NES, normalized Enrichment score; SPHK1, sphingosine kinase 1; RCC, renal cell carcinoma; GSEA, gene set enrichment analysis; TCGA, The Cancer Genome Atlas.

SPHK1 could be an effective diagnostic marker for RCC.

The correlation between SPHK1 expression and the disease-free survival of patients with RCC

To investigate the impact of SPHK1 on the survival of those with RCC, we used the Kaplan-Meier method and the log-rank test to perform a survival analysis. Higher SPHK1 expression was significantly associated with worse disease-free survival (*Figure 5A*). Subgroup analysis further showed that SPHK1 could be an effective prognostic marker for patients with RCC aged more than 60 years (*Figure 5B*), female patients with RCC (*Figure 5C*), RCC with a lower pathological grade (G1-2; *Figure 5D*), RCC with a lower T stage (T1-2; *Figure 5E*), and RCC without lymph node metastasis (*Figure 5F*) or distant metastasis (*Figure 5G*). From these findings, we concluded that SPHK1 is a potent prognostic marker for the disease-free survival of RCC.

The association of SPHK1 expression with the overall survival of RCC

We further explored the effect of SPHK1 on the overall survival of patients with RCC. Higher SPHK1 expression

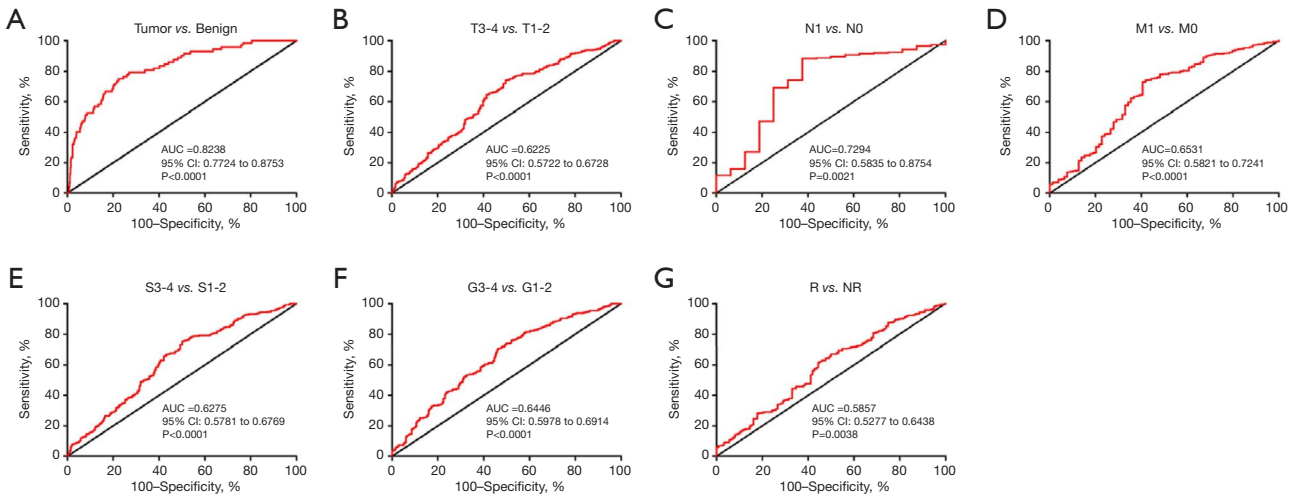

Figure 4 The diagnostic value of SPHK1 in RCC. (A) The ROC curve showed that SPHK1 could effectively differentiate RCC tissue from benign tissue. (B) The ROC curve showed that SPHK1 could effectively differentiate advanced T stage RCC from early T stage RCC. (C) The ROC curve showed that SPHK1 could effectively differentiate RCC with lymph node metastasis from RCC without lymph node metastasis. (D) The ROC curve showed that SPHK1 could effectively differentiate metastatic RCC from localized RCC. (E) The ROC curve showed that SPHK1 could effectively differentiate advanced-stage RCC from early-stage RCC. (F) The ROC curve showed that SPHK1 could effectively differentiate high-grade RCC from low-grade RCC. (G) The ROC curve showed that SPHK1 could effectively differentiate recurrent RCC from nonrecurrent RCC. SPHK1, sphingosine kinase 1; RCC, renal cell carcinoma; R, recurrence; NR, nonrecurrence; ROC, receiver operating characteristic.

was significantly associated with worse overall survival (*Figure 6A*). Subgroup analysis further showed that SPHK1 could be an effective prognostic marker for patients with RCC both younger and older than 60 years (*Figure 6B,6C*), both females and males (*Figure 6D,6E*), with a lower or higher T stage (T1-2; *Figure 6F,6G*), without lymph node metastasis (*Figure 6H*), with and without distant metastasis (*Figure 6I,6J*), with a higher clinical stage (*Figure 6K*), with a higher pathological grade (G3-4; *Figure 6L*), and with or without recurrence (*Figure 6M,6N*). From these findings, we concluded that SPHK1 was a potent prognostic marker for the overall survival of RCC.

Discussion

Targeted therapy is the mainstay of treatment for patients with advanced RCC. However, no more than 40% of patients respond to antiangiogenic agents, including sunitinib, temsirolimus, and bevacizumab (10). Furthermore, the determinants of response and resistance to targeted therapy are largely unknown. The need for biomarkers of response to treatment has become more compelling because novel systemic immunotherapy agents, including anti–programmed cell death protein 1 (PD-1) monoclonal antibody nivolumab and anti–cytotoxic T-lymphocyte–associated protein 4 (CTLA-4) monoclonal antibody ipilimumab, have already been approved for advanced RCC and can be an alternative to angiogenesis inhibitors (11). Therefore, identifying biomarkers to select the best therapy for each patient during their natural history of the disease has become a priority in precision medicine research (12).

Recent studies have provided evidence to support the finding that SPHK1 is associated with anticancer-drug resistance in several types of cancer, including RCC, colorectal cancer, and prostate cancer (8,9,13,14). We identified SPHK1 as a highly expressed protein in RCC treated with targeted therapy. Elevated SPHK1 was associated with a higher clinical stage and pathological grade as well as a high risk of tumor recurrence. SPHK1 is known to phosphorylate Sph to S1P, which has been implicated in resistance to the antiangiogenic therapy of RCC. The SPHK1–S1P pathway is highly unregulated in TKI-resistant RCC. Since patients with advanced RCC treated with targeted therapy eventually become resistant, SPHK1 could be a critical regulator of acquired targeted therapy resistance. Further studies have shown that SPHK1

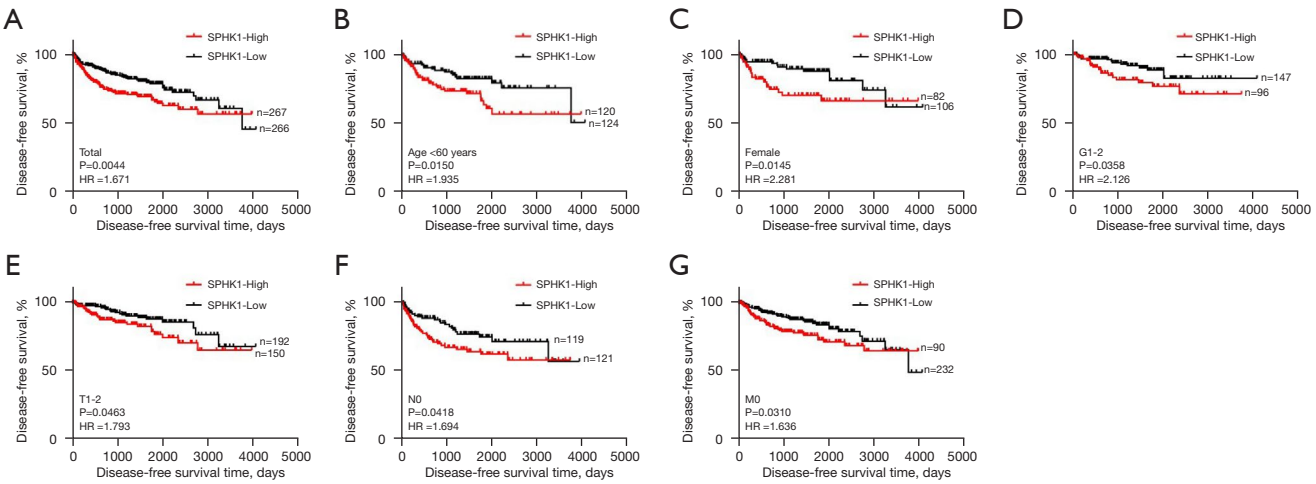

Figure 5 The correlation between SPHK1 expression and disease-free survival of RCC. (A) Kaplan-Meier survival analysis of the association between SPHK1 expression and disease-free survival of total patients with RCC. (B) Kaplan-Meier survival analysis of the association between SPHK1 expression and disease-free survival of patients with RCC younger than 60 years. (C) Kaplan-Meier survival analysis of the association between SPHK1 expression and disease-free survival of female patients with RCC. (D) Kaplan-Meier survival analysis of the association between SPHK1 expression and disease-free survival of patients with low-grade RCC. (E) Kaplan-Meier survival analysis of the association between SPHK1 expression and disease-free survival of patients with early T stage RCC. (F) Kaplan-Meier survival analysis of the association between SPHK1 expression and disease-free survival of patients with RCC and no lymph node metastasis. (G) Kaplan-Meier survival analysis of the association between SPHK1 expression and disease-free survival of localized patients with RCC. X-tile was used to generate an optimal cutoff point to dichotomize SPHK1 mRNA into high and low classificaitions using a Monte Carlo P value <0.05. The long-rank test was used to evaluate the differences between groups; the P value, hazard ratio, and 95% confidence interval are shown. HR, hazard ratio; SPHK1, sphingosine kinase 1; RCC, renal cell carcinoma.

can activate stemness-associated signaling pathways, including Wnt, Hedgehog, and Notch, in both targeted agents in treated or untreated RCC, indicating that SPHK1 induces targeted therapy resistance by enhancing RCC cell stemness (15-17).

Identifying the biomarkers for diagnosis, prognosis prediction, follow-up, and monitoring treatment for patients with RCC is still challenging (18,19). We assessed the predictive value of SPHK1 for the prognosis of patients with RCC. SPHK1 expression could effectively differentiate RCC tissue from benign tissue. Higher SPHK1 expression in RCC indicated a higher clinical stage and pathological grade as well as metastatic disease. Notably, elevated expression of SPHK1 was associated with a worse disease-free survival rate of patients with RCC. However, lower SPHK1 expression was correlated with better outcomes. Our study provided a novel diagnostic marker and a new prognostic biomarker, SPHK1, for patients with RCC.

The therapeutic options available for advanced RCC have expanded and now include immune checkpoint inhibitors, targeted agents, and combination strategies. Biomarkers are needed to guide the choice of therapeutic agents based on key features in each patient's tumor cells (20). Thus, identifying tissue-based predictive biomarkers for RCC has become a pressing need (21). A key finding from the present study is to identify SPHK1 as a biomarker of angiogenic agent resistance; advanced RCC with high SPHK1 expression could be resistant to these angiogenic agents, and immune checkpoint inhibitors may be a better therapy for these patients.

SPHK1 has unique advantages as a biomarker for RCC. The expression level of SPHK1 could predict the stage and grade of the disease as well as the prognosis of patients with RCC. In this study, overexpression of SPHK1 was associated with targeted therapy resistance, and the expression level of SPHK1 could predict whether the tumor was resistant to sunitinib. However, the present study was limited because it lacked real-world experimental data to support the results based on bioinformatics analysis. Further studies are required to validate the conclusions of this study.

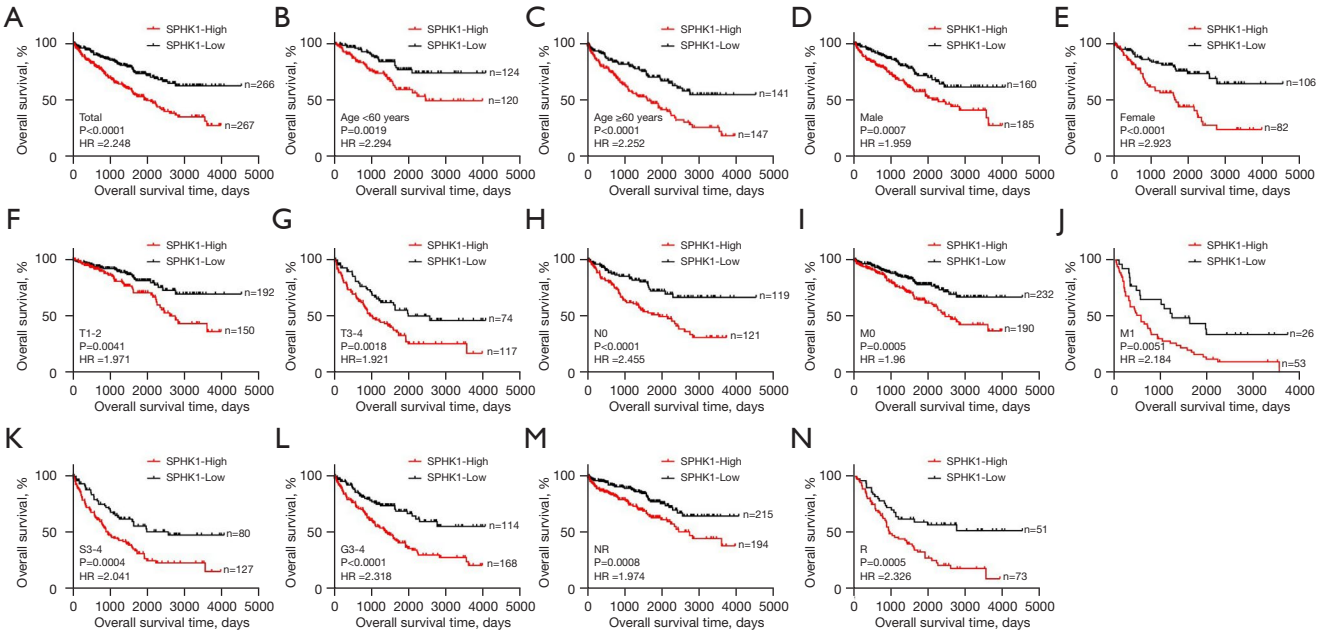

Figure 6 The association of SPHK1 expression with overall survival of RCC. (A) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of total patients with RCC. (B) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with RCC younger than 60 years. (C) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with RCC aged ≥60 years. (D) Kaplan-Meier survival analysis of the association between SPHK1 expression and the overall survival of male patients with RCC. (E) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of female patients with RCC. (F) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with early T stage RCC. (G) Kaplan-Meier survival analysis of the association between SPHK1 expression and the overall survival of patients with advanced T stage RCC. (H) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with RCC and no lymph node metastasis. (I) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with localized RCC. (J) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with metastatic RCC. (K) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with advanced-stage RCC. (L) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with high-grade RCC. (M) Kaplan-Meier survival analysis of the association between SPHK1 expression and overall survival of patients with nonrecurrent RCC. (N) Kaplan-Meier survival analysis of the association between SPHK1 expression and the overall survival of patients with recurrent RCC. X-tile was used to generate an optimal cutoff point to dichotomize SPHK1 mRNA into high and low classifications using a MonteCarlo P value <0.05. The long-rank test was used to evaluate the differences between groups. The P value, hazard ratio, and 95% confidence interval are shown. HR, hazard ratio; SPHK1, sphingosine kinase 1; RCC, renal cell carcinoma; R, recurrence; NR, nonrecurrence.

Conclusions

Overexpression of SPHK1 is associated with RCC development and antiangiogenic agent resistance. Elevated SPHK1 expression predicts poor outcomes in patients with RCC as well as angiogenic agent resistance. We can conclude that SPHK1 is a valuable biomarker of response to targeted therapy and an effective prognostic marker for RCC.

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Footnote

Reporting Checklist: The authors have completed the REMARK reporting checklist. Available at https://tcr.amegroups.com/article/view/10.21037/tcr-22-417/rc

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://tcr.amegroups.com/article/view/10.21037/tcr-22-417/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013).

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