*Research Article*

# A Survival Model Based on the ASB Genes and Used to Predict the Prognosis of Kidney Renal Clear Cell Carcinoma

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Kidney renal clear cell carcinoma (KIRC) is increasing in incidence worldwide, with poor and unpredictable patient prognosis limited by diagnostic and therapeutic approaches. New genes are urgently needed to improve this situation. The ankyrin repeat and suppressor of the cytokine signaling (SOCS) box (ASB) family are a promising class of tumorigenesis-related genes. We examined the expression and mutation of 18 ASB genes in various tumors for this study. The findings revealed that ASB genes exhibit significant copy number variation (CNV) and single nucleotide variation (SNV). There were substantial variations in ASB gene expression in different tumor tissues, and different levels of methylation of ASB genes affected the gene expression and tumor progression. By applying LASSO regression analysis, we established a KIRC survival model based on five ASB genes (ASB6, ASB7, ASB8, ASB13, and ASB17). Additionally, ROC curve analysis was used to assess the survival model's accuracy. Through univariate and multivariate COX regression analysis, we demonstrated that the model's risk score might be an independent risk factor for individuals with KIRC. In summary, our KIRC survival model could accurately predict patients' future survival. Further, we also quantified the survival model through a nomogram. This series of findings confirmed that ASB genes are potential predictive markers and targeted therapies for KIRC. Our KIRC survival model based on five ASB genes can help more clinical practitioners make accurate judgments about the prognosis of KIRC patients.

## 1. Introduction

According to the GLOBOCAN 2020 study released by the International Agency for Research on Cancer, kidney cancer has become increasingly severe worldwide in recent years. Kidney cancer reported 431,288 incidences and 179,368 fatalities in 2020. The proportion of incidences and fatalities of kidney tumors in Asia is 36.3% and 44.7%, the highest in the world [1]. One of the most frequent forms of renal neoplastic diseases, accounting for 80% and 90% of renal malignancies, is renal cell carcinoma (RCC). One of RCC's deadliest and most prevalent pathological subtypes, kidney renal clear cell carcinoma (KIRC), accounts for around 75% of RCC cases [2]. Studies related to KIRC have shown that the incidence of the disease is significantly higher in men than in women. In addition to gender factors,

obesity, hypertension, poor lifestyle and dietary habits, and chronic kidney disease are all risk factors for developing KIRC [3].

Regarding clinical management, limited KIRC is usually treated with surgical approaches such as partial and radical nephrectomy, supplemented by radiotherapy and targeted therapy to improve treatment success [4]. Patients with surgical treatment usually experience tumor recurrence after 36 months, along with metastasis to other sites such as the lung and liver. Patients with tumor recurrence and metastasis have lower treatment efficiency and less sensitive and specific diagnostic methods. These factors contribute to the poor quality of life and poor prognosis of KIRC patients and place a severe burden on their families and the social healthcare system [5, 6]. The search for diagnostic and therapeutic genes highly associated with the occurrence of

KIRC is crucial. Relevant scientific researchers are pursuing it to increase the KIRC patients' quality of survival.

The ankyrin repeat and suppressor of the cytokine signaling (SOCS) box (ASB) family are the most prominent member of the SOCS box protein superfamily and a member of the E3 ubiquitin ligase family [7]. There are 18 members of the ASB family, named ASB1-ASB18. Each ASB family member binds to more than one protein, and each protein binds specifically to only one ASB family member [8]. ASB family members share a common structural feature, consisting of two structural domains, a variable number of ankyrin repeats at the N-terminus and the SOCS box at the C-terminus [9]. To create an E3 ubiquitin ligase complex, SOCS attaches to Cullin protein and engages in interactions with Elongin B/C. This complex recognizes substrates of substrates and is involved in the degradation of the proteasome [10]. ASB family members are potential tumor-associated genes that work with various target substrates through their two specific structurals. It involves regulating cell proliferation and differentiation, altering the cell cycle, and promoting cellular carcinogenesis, affecting the development and prognosis of normal cells or tumors [11]. However, the progress of tumor research related to ASB genes has been plodding. Many biological functions of ASB genes and the mechanisms associated with tumorigenesis have not been clearly described in the current literature.

In this investigation, we thoroughly examined the activation and alterations of 18 ASB family members in 32 different tumor types, investigated the coexpression relationships among ASB family members, and analyzed the relevant pathways in which ASB family members play a role in tumorigenesis. Through LASSO regression, we established a KIRC survival model based on five ASB genes, and the accuracy of this survival model was validated using ROC curve analysis. Moreover, it quantified the entire survival prediction model by nomogram to facilitate more clinical practitioners to make accurate judgments affecting KIRC sufferers' chances of survival.

## 2. Methods and Materials

*2.1. Data Collection.* The TCGA database provided the initial data for this investigation [12]. We downloaded the raw data of CNV and SNV from this database for 32 different tumor types, analyzed the raw data using Perl language, and visualized the data results through TBtools software [13]. Meanwhile, the GSCALite database [14] was used to investigate the associations between methylation and the activation of ASB genes, the link between methylation and survival, and the degree to which ASB genes activate and inhibit conventional pathways. The R/Bioconductor software from TCGA Biolinks was used to retrieve the RNA-seq data from KIRC used in this work [15].

Moreover, the data of ASB gene expression in standard kidney specimens and KIRC specimens were analyzed using the "Limma" package. We use the "Pheatmap" package to visualize the analysis results as a heat map. Furthermore, the TCGAbiolinks database was used to retrieve and evaluate various pathological parameters, including M (tumor

metastasis), T (tumor size), tumor grade, stage, age, fustat, and futime. The "Glmnet" and "Survival" packages were used to conduct LASSO regression analysis. The univariate and multivariate Cox analysis of clinical variables was also performed using the "Survival" package.

*2.2. PPI Network and Coexpression Analysis of ASB Genes.* This study analyzed the interactions between 18 ASB genes using the STRING online tool (https://www.string-db.org/) [16]. Meanwhile, we used the Cytoscape tool to visualize the data results [17]. We also performed coexpression analysis of the 18 ASB genes by the "Corrplot" package.

*2.3. Construction of Regression Models and Evaluation of Risks.* We analyzed the correlation between the levels of gene activation and overall survival (OS) in KIRC using a univariate Cox model. Meanwhile, we explored the correlation between the expression of 18 ASB genes in KIRC by coexpression analysis. We processed the correlation data using LASSO analysis to exclude overfitting genes, reduce variables, and optimize the model. Besides, we used COX regression analysis to identify several ASB genes with the most potent predictive power for KIRC prognosis. We calculated patients' risk scores based on the risk score index.

$$\text{risk score} = \sum_{i=1}^{n} \left[ \text{Exp}_i * \text{Coe}_i \right], \tag{1}$$

where N denotes the number of genes, $\text{Coe}_i$ denotes the coefficient of $\text{gene}_i$, and $\text{Exp}_i$ denotes the expression of $\text{gene}_i$.

Using the average risk score, we classified the patients into high-risk and low-risk groups based on their risk scores. The difference in survival between the two groups was then evaluated using Kaplan–Meier (KM) survival curves. In addition, we also assessed the accuracy of the future 1, 3, and 5 year survival models for KIRC patients by time-related ROC curve analysis.

*2.4. Statistical Analysis.* The "SURVMINER" package was used to calculate risk scores. The patients were classified into high-risk and low-risk groups based on their median risk scores. We performed a statistical analysis of the data by the R Studio software package, and $P < 0.05$ was regarded as statistically significant.

## 3. Results

*3.1. Genetic Alterations of ASB Genes in 32 Tumors.* To learn more about how ASB genes are related to cancer, we comprehensively reviewed the relevant literature and summarized 18 highly relevant ASB genes. Using the TCGA database, we identified the CNV and SNV of these 18 ASB genes in different cancer types. We downloaded the CNV and SNV information for the 18 ASB genes in 32 different tumor types from the TCGA database. We performed validation analysis of these raw data using the R language, and the data results were visualized by TBtools software (Figures 1(a) and 1(b)). We discovered various degrees of
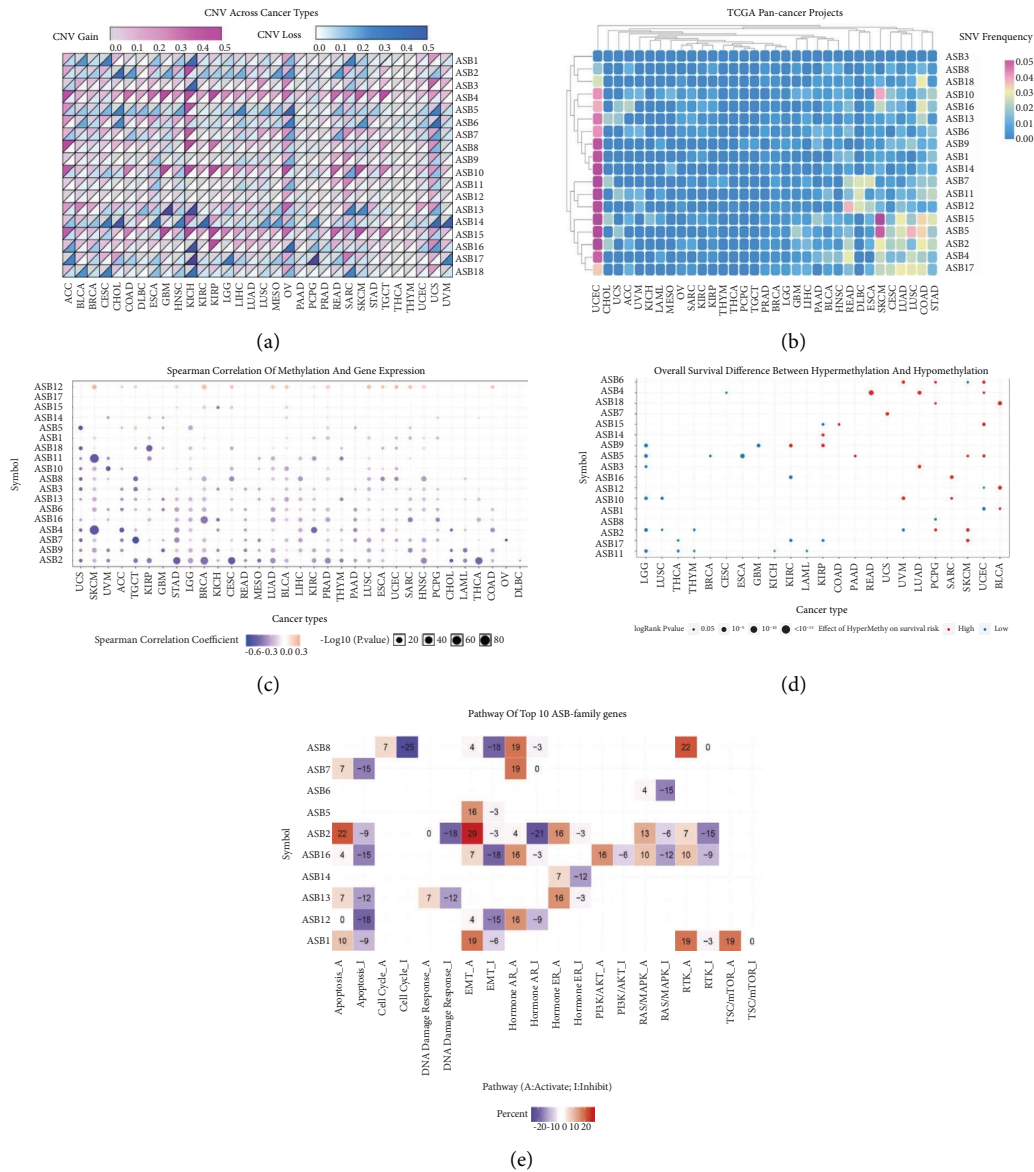
(a)



(b)



(c)



(d)



(e)

FIGURE 1: Heritability variations of ASB gene family in 32 cancers. (a) Copy number variation (CNV) gain or loss of ASB genes in pan-cancer. Pink represents CNV gain, blue represents CNV loss, and darker colors represent more robust gain or loss. (b) The change of single nucleotide variation (SNV) of ASB gene in pan-cancer, the color changes from blue to pink as the mutation increases. (c) Correlation between the degree of ASB gene methylation and gene expression. (d) Correlation between the degree of ASB gene methylation and overall survival. High levels of methylation are denoted by the colors, red for high-risk factors and blue for low-risk factors. (e) The action pathways of 10 ASB genes are analyzed, with red representing activation and blue representing suppression.

copy number gain and loss of ASB genes in 32 distinct tumor types. In KICH, ACC, and KIRP, specific ASB genes exhibited greater gain mutation frequencies. In various cancers, the gain mutation frequencies were greater for ASB4, ASB10, and ASB15. In contrast, the loss mutation frequencies of ASB genes were greater in OV, UCS, and KICH. In various cancers, the loss mutation frequencies of ASB14, ASB17, and ASB18 were greater. The ASB gene exhibited varying degrees of variation in 32 distinct tumor types when we looked into its single nucleotide variants. Among them, UCEC showed a significant frequency of single nucleotide variations in the ASB gene. In addition, we

also explored the relationship between the degree of gene methylationand gene expression in different tumors. The exploration results showed that the methylation level of most ASB genes negatively correlated with ASB gene expression in different tumors. Among them, the relationship between the methylation levels of ASB11 and ASB4 and gene expression showed a significantnegative correlation in SKCM. That is, the higher the methylation level of ASB11 and ASB4 genes in SKCM, the lower the expression of both genes. Similar to this phenomenon, the degree of ASB2 gene methylation showed a significant negative correlation with gene expression in four tumors: STAD, BRCA, CESC, and THCA
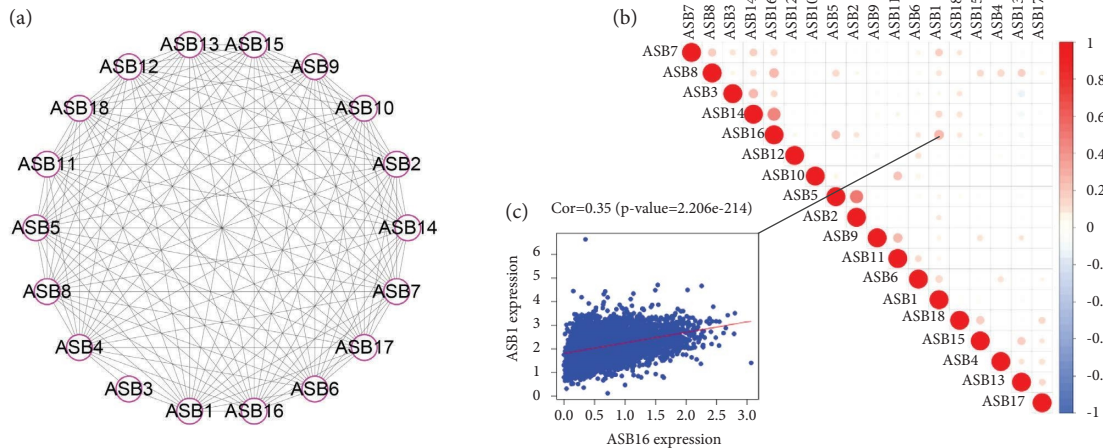
FIGURE 2: PPI network of ASB genes. (a) Results of PPI network analysis between ASB genes. (b) Coexpression analysis between ASB genes. Blue denotes a negative connection, whereas red indicates a positive association. (c) Analysis of the coexpression of ASB1 and ASB16.

(Figure 1(c)). Meanwhile, we further explored the differences between the degree of gene methylation and survival risk in different tumors. The results showed that in pancancer, different methylation degrees of the ASB gene would bring different survival risks, which is closely related to the prognosis of patients. Among them, the hypermethylation of the ASB4 gene posed a higher survival risk associated with poor patient prognosis in READ. In contrast to this phenomenon, the hypermethylation of the ASB5 gene can bring a relatively low survival risk associated with a good prognosis in ESCA (Figure 1(d)).

### 3.2. Functional Analysis of the Linkage and Action Pathways among ASB Gene Families.
To investigate the association between these 18 ASB genes, we performed protein-protein interaction (PPI) analysis on these 18 genes through the STRING website, and the data results were visualized by Cytoscape software (Figure 2(a)). To further investigate the correlation between the genes, Using the "Corrplot" package, we examined the 18 ASB genes' coexpression (Figure 2(b)). It showed a positive association between the ASB1 and ASB16 genes with a Pearson correlation coefficient (PCC) of 0.35 (Figure 2(c)). As a whole, it seems that most of the ASB genes were likewise positively correlated with each other and had a strong association. Meanwhile, we examined the traditional pathways of ASB genes to learn more about the function of ASB genes in carcinogenesis. The investigation revealed that 10 ASB genes either inhibit or activate the signaling pathways. These signaling pathways include apoptosis, cell cycle, DNA damage response, epithelial-mesenchymal transition (EMT), estrogen receptor (AR, ER), PI3K/AKT, RAS/MAPK, RTK, and TSC/ mTOR during tumorigenesis (Figure 1(e)). Among them, these genes, including ASB2, ASB16, ASB12, and ASB1, have significant inhibitory effects on apoptosis and can vigorously promote the activation of EMT.

### 3.3. Differences in ASB Gene Activation and Prognosis in Various Cancers.
We retrieved the data of 18 distinct human cancers from the database to investigate the variations in

ASB gene expression in various tumors. Moreover, we analyzed the raw data using the *R* language and visualized the analysis results through TBtools software (Figure 3(a)). The analysis showed that these 18 ASB genes were expressed to varying degrees in 18 different types of cancer. Among them, the genes ASB18, ASB6, ASB3, ASB7, and ASB14 had higher $Log_2$ (FC) values in a variety of tumors, suggesting that the expression of these genes was significantly higher in tumor tissues than in normal tissues. In contrast, the genes ASB11, ASB5, and ASB2 had lower $Log_2$ (FC) values in multiple tumors, suggesting that the expression of these ASB genes in tumor tissues was significantly lower than that in normal tissues. We further analyzed the raw data to explore the role of ASB genes in different tumors. In total, we explored the roles played by 18 ASB genes in 25 different tumor types. Based on the value of the hazard ratio of ASB genes in a particular tumor, we defined ASB genes with a hazard ratio greater than one as risk genes for that tumor. In contrast, we defined ASB genes with a hazard ratio of less than one as protective genes for that tumor. Finally, we visualized the results of our analysis by TBtools software (Figure 3(b)). Following a pan-cancer examination of ASB genes, we found that these genes function as risk factors in most cancers and that increased ASB gene expression is associated with a poorer patient prognosis. However, in KIRC, the expression of many ASB genes was positively linked with patient prognosis, indicating that many ASB genes had a protective effect on cancer and that the greater the expression of ASB genes, the better the prognosis of patients. Based on this fascinating discovery, we thoroughly looked into the relationship between the KIRC and ASB genes.

### 3.4. Correlation between ASB Gene and Prognosis of Kidney Renal Clear Cell Carcinoma (KIRC).
We collected ASB gene expression data in 72 normal kidney tissues and 539 KIRC tissues. Using the "Limma" package in *R* language, we did a detailed analysis of the collected data. The analysis showed that 14 out of 18 ASB genes in KIRC specimens significantly differed from that in normal kidney specimens. Among
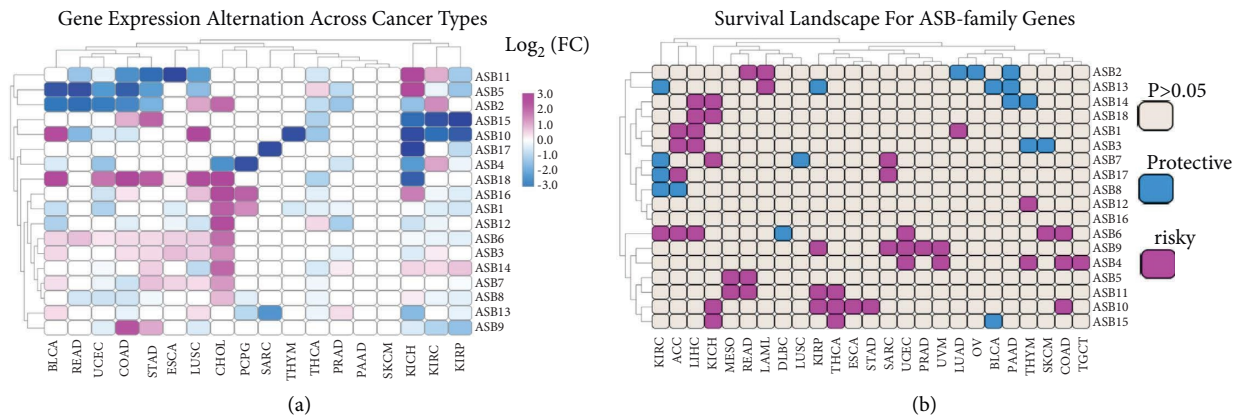
FIGURE 3: Expression of ASB genes in cancer and impact on survival (a) 18 different forms of cancer express 18 ASB genes. The $\text{Log}_2$ (FC) values are displayed in a color-coded legend on the right side. (b) Survival of 18 ASB genes in 25 different types of cancers. White represents $P > 0.05$ and red denotes danger genes, whereas blue denotes protective genes.

them, ASB2, ASB4, ASB3, ASB11, and ASB14 were significantly more expressed in KIRC specimens than in normal kidney tissues. In contrast to this phenomenon, the expression of ASB12, ASB9, ASB15, ASB10, ASB6, ASB1, ASB8, ASB5, and ASB16 genes in KIRC tissues was significantly reduced compared to their expression in normal kidney tissues (Figure 4(a)).
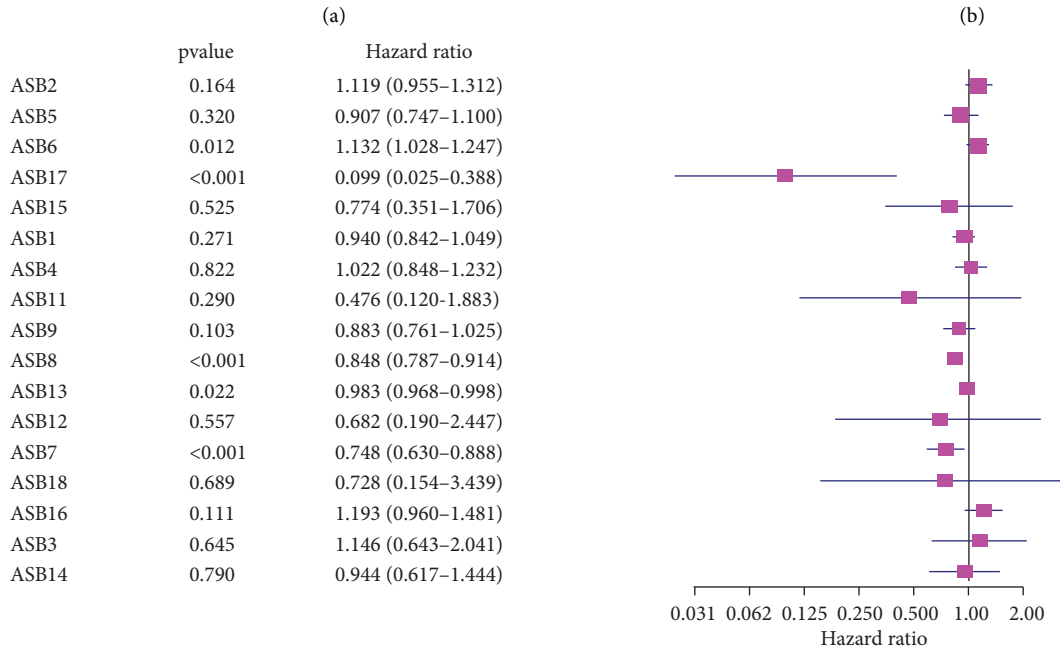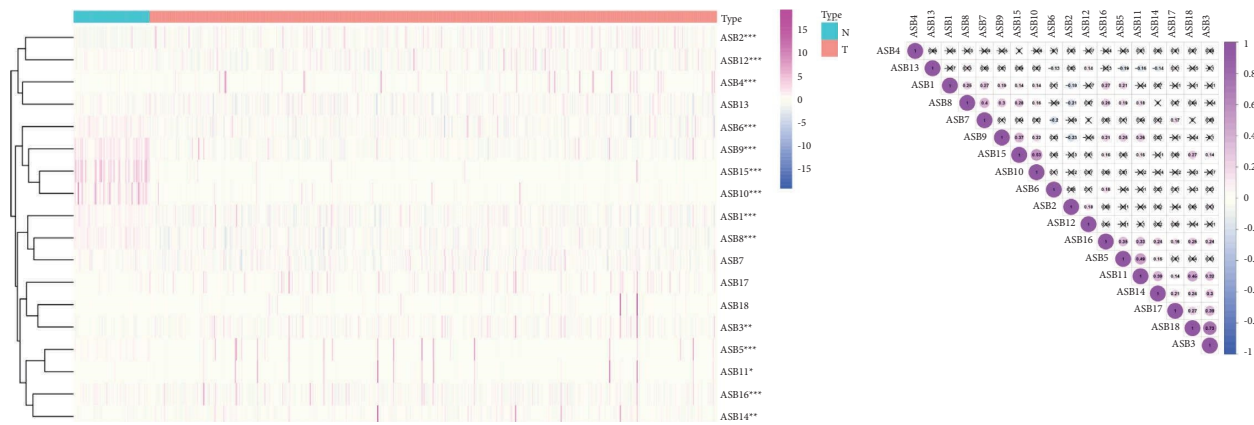
Meanwhile, to investigate the correlation between ASB genes in KIRC patients, we did a coexpression analysis of 18 ASB genes in KIRC patients (Figure 4(b)). The results suggested that although a small number of ASB genes were negatively correlated, most ASB genes were positively correlated with each other and had a strong association. To further clarify the relationship between ASB genes and KIRC prognosis, we analyzed the activation of ASB genes in KIRC using univariate Cox regression analysis. The findings demonstrated a worse prognosis in KIRC was related to elevated ASB6 gene expression, in contrast to high expression of four ASB genes, ASB17, ASB8, ASB13, and ASB7, respectively, which resulted in KIRC patients having a good prognosis (Figure 4(c)). Their findings were in line with the pictures depicting the relative link between the expression of genes and patient survival time in the study of all types of cancer (Figure 3(b)). It is suggested that ASB6 is a risk factor for tumor development and ASB17, ASB8, ASB13, and ASB7 are protective factors that inhibit tumor development during KIRC development.

*3.5. A New KIRC Survival Model Based on the ASB Genes.* We chose a few ASB genes as subsistence genes in the findings of the univariate Cox regression model (Figure 4(c)). We reduced some unnecessary genes through LASSO regression analysis and identified the most correlated prognostic indicators. According to the minimal criterion, we selected 5 ASB genes (ASB17, ASB8, ASB13, ASB7, and ASB6) in our analysis results to establish the ASB gene-related KIRC survival model (Figures 5(a) and 5(b)). We categorized KIRC patients into high- and low-risk groups based on the average risk

score. Moreover, the Kaplan–Meier survival curve (K-M survival curve) analysis suggested that patients in the high-risk group fared worse than those in the low-risk group in terms of survival rates (Figure 5(c)). Meanwhile, to evaluate the prognostic, predictive capability of the novel survival model for KIRC patients, we analyzed the survival AUC scores of KIRC patients at 1, 3, and 5 years using the ROC curve (Figures 5(d)–5(f)). One-year survival had an AUC of 0.711, three-year survival of 0.656, and five-year survival of 0.668. The evaluation showed that the new KIRC survival model could accurately predict patients' survival for the following year. However, it was relatively inaccurate for patients' survival for the next 3 and 5 years.

*3.6. New KIRC Survival Model Correlates with Clinicopathological Features.* We examined the correlation between the clinicopathological characteristics of KIRC patients with various risk levels in the TCGA database and risk scores derived from five ASB expressions through the heat map (Figure 6(a)). The clinicopathological characteristics included M (tumor metastasis), T (tumor size), grade, stage, age, fustat, and futime. Based on the analysis results, we observed that the expression of four ASB genes, ASB17, ASB8, ASB13, and ASB7, was significantly lower in the high-risk group than in the low-risk group. In contrast to this phenomenon, the high-risk group had higher levels of ASB6 gene expression than the low-risk group.

However, age, tumor grade, tumor stage, T, M, and risk score, were all significantly linked with OS in KIRC patients according to univariate Cox regression analysis (Figure 6(b)). Moreover, age, risk score, tumor stage, and tumor grade were also found to be independent risk variables for the prognosis of KIRC patients, according to multivariate Cox regression analysis (Figure 6(c)). Finally, using *R*, we create a nomogram for predicting the risk of KIRC patients (Figure 7). The nomogram is divided into nine rows. The first row is the factional meter, and rows 2 through 5 show the age, tumor grade, tumor stage, and risk

(a)



(b)

|        | pvalue  | Hazard ratio          |
|--------|---------|-----------------------|
| ASB2   | 0.164   | 1.119 (0.955–1.312)   |
| ASB5   | 0.320   | 0.907 (0.747–1.100)   |
| ASB6   | 0.012   | 1.132 (1.028–1.247)   |
| ASB17  | <0.001  | 0.099 (0.025–0.388)   |
| ASB15  | 0.525   | 0.774 (0.351–1.706)   |
| ASB1   | 0.271   | 0.940 (0.842–1.049)   |
| ASB4   | 0.822   | 1.022 (0.848–1.232)   |
| ASB11  | 0.290   | 0.476 (0.120-1.883)   |
| ASB9   | 0.103   | 0.883 (0.761–1.025)   |
| ASB8   | <0.001  | 0.848 (0.787–0.914)   |
| ASB13  | 0.022   | 0.983 (0.968–0.998)   |
| ASB12  | 0.557   | 0.682 (0.190–2.447)   |
| ASB7   | <0.001  | 0.748 (0.630–0.888)   |
| ASB18  | 0.689   | 0.728 (0.154–3.439)   |
| ASB16  | 0.111   | 1.193 (0.960–1.481)   |
| ASB3   | 0.645   | 1.146 (0.643–2.041)   |
| ASB14  | 0.790   | 0.944 (0.617–1.444)   |



(c)

FIGURE 4: Effect of ASB gene in KIRC patients. (a) Analysis of the difference in ASB gene expression in normal kidney tissue and KIRC specimens. N represents normal kidney tissue, and T represents KIRC tissue. Blue denotes a low level of ASB gene expression, whereas red indicates a high level. (b) Coexpression analysis of 18 ASB genes in KIRC. (c) Analysis of the ASB genes' hazard ratio in KIRC.

score. The scores corresponding to rows 2 through 5 are added to produce the total score in the sixth row. We were able to forecast the survival rate of KIRC patients in the upcoming 1, 3, and 5 years based on the total score in the sixth row.

## 4. Discussion

Numerous investigations on the role of ASB genes in cancers are now being conducted. The ASB2 gene significantly affects malignant hematologic diseases, and inhibition of ASB2 gene expression inhibits the NF-κB pathway. It can produce cytotoxic effects on diffuse large B-cell lymphoma cells and promotes apoptosis of T cell acute lymphoblastic leukemia (T-All) related cells. It enhances patients' prognosis by preventing T-All cell growth [18, 19]. ASB3, ASB4, and ASB9 have been intensively studied in colorectal and

hepatocellular carcinoma. The expression of these genes is suppressed or promoted, and they play a critical part in the growth of malignancies [20–23].

Interestingly, in studies related to hepatocellular carcinoma, inhibition of ASB3 gene expression can promote mitochondrial apoptosis and enhance cellular autophagy, synergistically promoting the death of hepatocellular carcinoma cells [20]. However, inhibition of ASB3 gene expression in studies related to colorectal cancer promoted proliferation and metastasis of colorectal cancer cells, which was detrimental to patient prognosis. In contrast, overexpression of the ASB3 gene prevented the proliferation of colorectal cancer cells by upregulating β-catenin and E-cadherin and downregulating N cadherin epithelial-mesenchymal transition to inhibit colorectal cancer metastasis [10]. In addition to the direct effect of ASB genes on tumor development, some ASB genes can also affect tumor
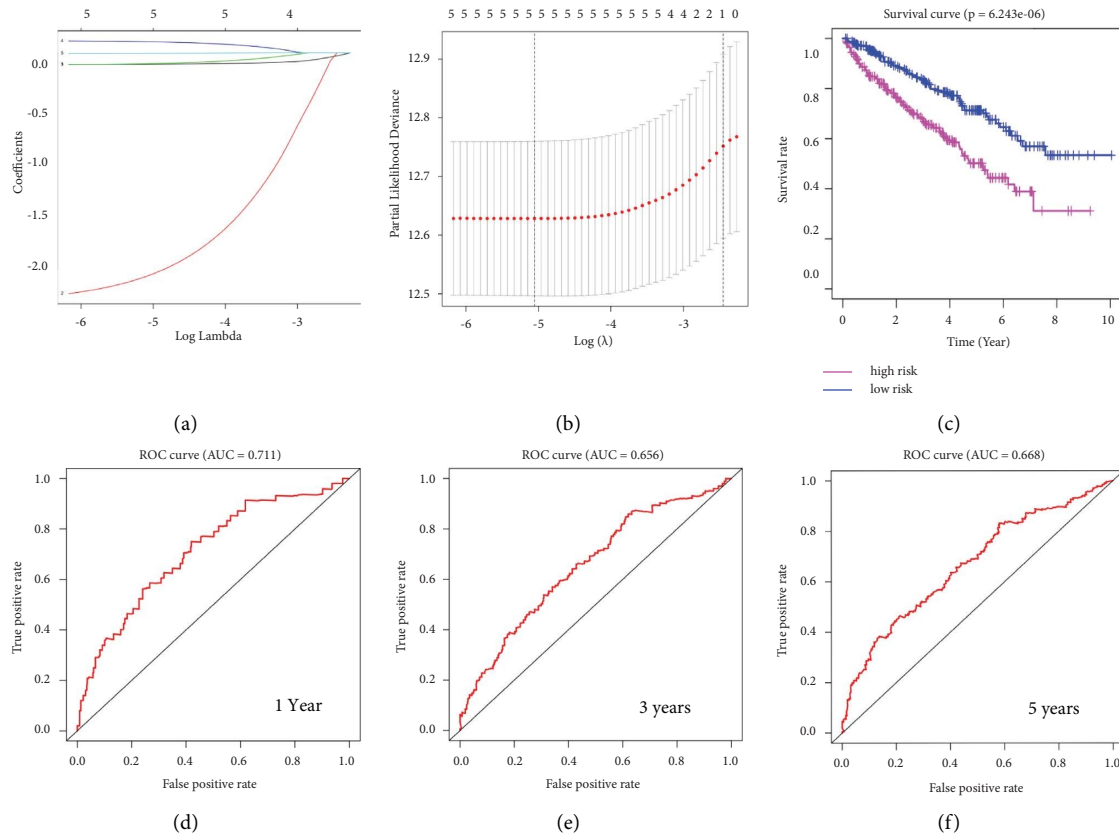
FIGURE 5: KIRC survival model based on ASB genes: (a) Lasso coefficient profiles of ASB gene family in KIRC. (b) Partial likelihood deviation marked with Log ($\lambda$). (c) Based on the KIRC patient's risk score, the Kaplan–Meier survival curve analysis displays the difference between the patients who survived in the high- and low-risk groups. The high-risk group is symbolized by red, while the low-risk group is shown by blue. (d–f) ROC curves predict survival at 1, 3, and 5 years.

growth by regulating the tumor microenvironment. Among them, the proinflammatory cytokine release and the effective control of the inflammatory response are influenced by the ASB1 gene [24]. ASB4 and ASB5 genes are associated with angiogenesis in humans [21, 25]. The expression of these genes at different levels in tumor patients can have different effects on the development of tumors. This body of literature suggests that ASB family genes may play a suppressive or promotive role in different tumors. Therefore, our pan-cancer analysis of ASB family genes using multiple bioinformatics tools in this study is crucial.

Our analysis of CNV and SNV data of 18 ASB genes in the ASB family revealed that ASB genes are mutated to varying degrees in 32 types of tumors. The results proved that ASB genes are crucial for tumorigenesis and impact how tumors develop. We also probed the ASB family genes for epigenetic changes. The results showed that the degree of methylation of most ASB genes was significantly and negatively correlated with ASB gene expression in different tumors. Exceptionally, in pan-cancer, the degree of methylation of ASB12 was positively correlated with gene expression in tumors. During the further exploration of the degree of gene methylation and survival risk, we found that different degrees of methylation levels of ASB gene in pan-cancer would bring different survival risks.

However, hypermethylation of the ASB gene leads to a higher survival risk and a poor prognosis. We reviewed previous results on ASB gene methylation, and high methylation of the ASB1 gene is an important marker of cardiac deterioration [26]. That anxiety is brought on by the ASB1 gene's epigenetic control [27]. However, we did not find any studies in which epigenetic changes in the ASB gene were associated with tumors. Our results suggest that the role of ASB gene methylation is to suppress the expression of ASB genes. Those different ASB genes may play a protective or risk gene role in tumor development, which provides new ideas for basic research and inspires us to investigate the effect of ASB genes in tumor development.

We performed correlation analysis between each gene member of the ASB gene family, and the results showed a high degree of linkage between ASB gene families. Different ASB genes do not exist alone in the development of various tumors. However, they interact with each other and play synergistic or antagonistic roles in influencing the course of tumor development. Meanwhile, we are a result of our investigation of the tumors' ASB gene activity pathways. The findings indicated that ASB genes affect tumor development by activating or inhibiting apoptosis, cell cycle, DNA damage response, EMT, AR, ER, PI3K/AKT, RAS/MAPK,
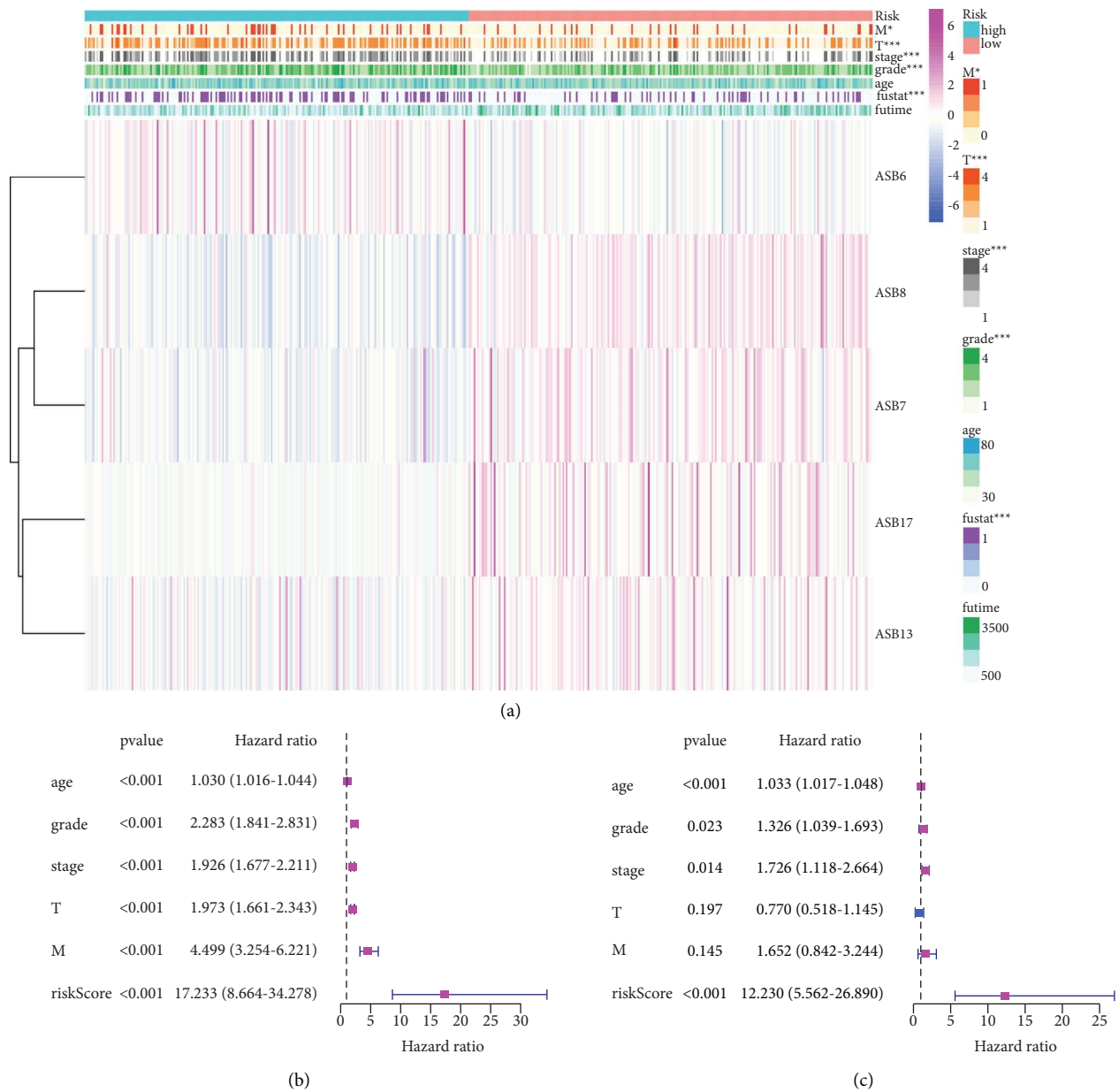
(a)



(b)



(c)

FIGURE 6: Clinical correlation analysis of the KIRC survival model. (a) Correlation analysis of risk scores with KIRC clinical case characteristics. Blue and red are symbols for up- and down-regulations, respectively. *P* values of <0.05, <0.01, and <0.001 are indicated by the symbols *, **, and ***, respectively. (b) Univariate analysis. (c) Multivariate analysis.

RTK, and TSC/mTOR pathways of action. Our results were further validated by reviewing previous basic research experiments [11, 24].

Meanwhile, we investigated the variations in the activation of the 18 ASB genes in normal tissues compared with 18 different types of tumor tissues. Our study found that ASB18, ASB6, ASB3, ASB7, and ASB14 were significantly more expressed in tumor tissues than in normal tissues. By further analysis of the data, we clarified the role of these 18 ASB genes in developing 25 different types of tumors based on their hazard ratios in tumors. The results showed that ASB genes are risk genes in most tumors. However, there are exceptions in

KIRC, where ASB genes are protective genes associated with a good prognosis for patients. This exciting study prompted us to explore the relationship between KIRC and ASB genes.

KIRC is a common and poorly prognosed type of urological tumor [3]. The study of the effect of the ASB gene on KIRC could lead to new prognostic and therapeutic targets for KIRC and bring new light to KIRC patients. In order to do this, we compared the expression of the ASB gene family's 14 genes in 72 normal and 539 malignant tissues. The results showed that the ASB genes were significantly differentially expressed in KIRC tissues compared to normal renal tissues. We also performed a coexpression analysis of 18 ASB genes in
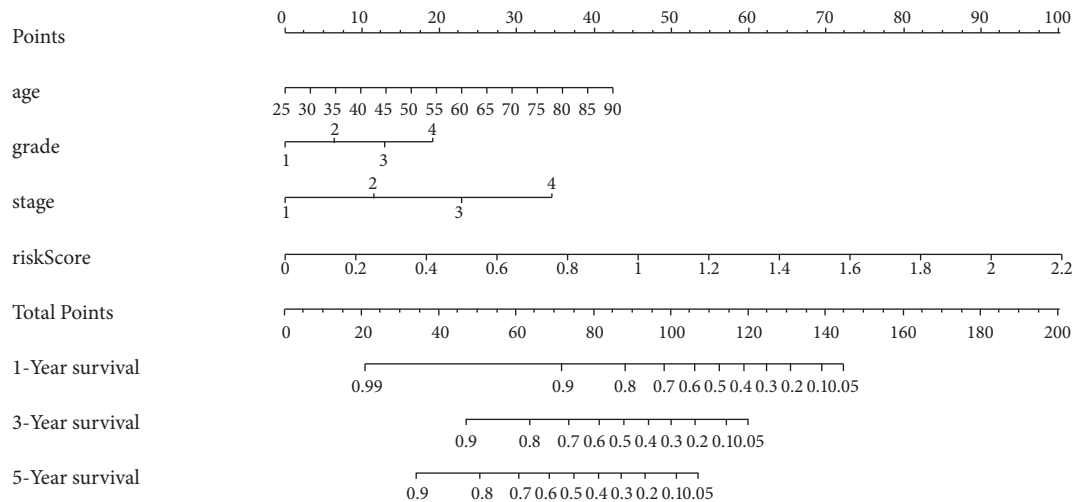
FIGURE 7: The KIRC survival model was used to create the nomogram.

KIRC tissues. The results suggest that most ASB genes are strongly associated with each other and influence each other during the development of KIRC. Immediately, we performed a univariate regression analysis of ASB genes in KIRC and clarified whether these ASB genes were risk genes or protective genes in the development of KIRC based on hazard ratios. To further investigate the relationship between ASB genes and KIRC prognosis, we urgently need to establish a survival model based on ASB genes.

The ASB gene family has 18 gene members, and too many genes can lead to the fitting of the results. To reduce the fitting of the results, we used LASSO regression to reduce unnecessary genes [28]. We finally identified five ASB genes (ASB6, ASB7, ASB8, ASB13, and ASB17) most strongly associated with KIRC prognosis. We developed a survival model for KIRC based on these five genes to predict patient survival. Using ROC curves, we assessed how well the survival model predicted the survival rate of KIRC patients at 1, 3, and 5 years. The corresponding AUC values were 0.711, 0.656, and 0.668. According to the assessment findings, our survival model could forecast KIRC patients' survival with high accuracy for the following year but with lesser accuracy for the following three and five years. We thoroughly analyzed the expression variations between these five ASB genes and KIRC clinicopathological characteristics in the high- and low-risk groups of carcinogenesis to further support the reliability of the KIRC-related survival model that focuses on these five ASB genes. On these clinico-pathological traits, we ran univariate and multivariate COX regression analyses. According to the analysis, individuals with high-risk scores were likely older patients with more advanced tumor stages and grades. Their activation of these five ASB genes was consistent with the genes themselves. A significant link exists between the risk score of the survival model and the prognosis of KIRC, as does age, tumor stage, and tumor grade. This series of findings again demonstrate the accuracy and reliability of our survival model, which can

provide clinical practitioners with great convenience in estimating how long KIRC sufferers will live in the future.

We have carefully examined the contribution of these five ASB genes in building survival models based on the findings of prior studies. The ASB6 gene is a novel biomarker and potential therapeutic target in developing oral squamous cell carcinoma, hepatocellular carcinoma, breast cancer, and other tumors. The ASB6 gene is a risk gene in tumors, and overexpression in various tumors can reduce endoplasmic reticulum stress response, promote filopodia formation, unsuppressed tumor growth, increase metastasis of cancer cells, and poorer patient prognosis [29–32]. A study confirmed that overexpression of the ASB8 gene in lung cancer has a regulatory effect on tumor cell growth. The ASB8 gene is a protective gene that is overexpressed in lung cancer cells, which can prevent the proliferation of those cells [33]. The high expression of the ASB13 gene is proportionate to the higher overall survival rate of breast cancer patients, and the changed copy number status of the ASB13 gene is strongly associated with the high expression of the gene [34]. The ASB13 gene is protective of breast carcinogenesis by promoting the degradation of SNAI2, deregulating the transcriptional repression of YAP, and inhibiting breast cancer metastasis [35]. Being directly linked to the growth of kidney tumors, the overexpression of the ASB13 gene can prevent lung tumor cells from metastasizing, but the exact mechanism has not been elucidated [35, 36]. The ASB17 gene is mainly expressed in the testis. It has been shown in previous studies to promote apoptosis, make the NF-κB pathway active, and boost proinflammatory cytokine production. It plays a role in cellular immunological and inflammatory reactions in vitro and in vivo [37, 38]. According to this study's findings, the ASB17 gene may have a role in controlling the tumor microenvironment during carcinogenesis and consequently influence tumor growth. The findings of this study demonstrate the need for more research into the pathways that affect tumor formation.

## 5. Conclusion

In conclusion, in this study, a new KIRC survival model was constructed by a series of analyses with five ASB genes selected from the ASB gene family. Based on this survival model, the AUC values for the 1-, 3-, and 5-year ROC curve analysis were 0.711, 0.656, and 0.668. This survival model could predict the survival of KIRC patients with high accuracy for the following year but lesser for the following three and five years, and the nomogram provided in the article quantifies the survival model. It may enable more medical professionals to choose individualized treatment regimens for KIRC patients and anticipate the survival rate of KIRC individuals in the upcoming years. It can extend the review period, reduce unnecessary tests, and relieve the financial pressure on the patient's family for individuals with little risk and a high chance of survival. For high-risk individuals with a poor prognosis, timely monitoring of disease progression, necessary treatment, and enhanced patient care can be provided. However, there are still several limitations of this study. One is that more clinical data are required to confirm the model's realistic accuracy as the information used in this research is retrospective data from open sources. Second, there are still many ASB genes in the ASB gene family whose biological functions in tumors have not been validated by basic experiments. More research is needed to improve the relevant experiments.

## Abbreviations

ACC: Adrenocortical carcinoma
BRCA: Breast invasive carcinoma
CESC: Cervical squamous cell carcinoma and endocervical adenocarcinoma
KICH: Kidney chromophobe
KIRC: Kidney renal clear cell carcinoma
KIRP: Kidney renal papillary cell carcinoma
OV: Ovarian serous cystadenocarcinoma
SKCM: Skin cutaneous melanoma
STAD: Stomach adenocarcinoma
UCS: Uterine carcinosarcoma
UCEC: Uterine corpus endometrial carcinoma
THCA: Thyroid carcinoma
TCGA: The Cancer Genome Atlas
LASSO: Least absolute shrinkage and selection operator
OS: Overall survival
CNV: Copy number variations
SNV: Single nucleotide variation
AUC: Area under the curve
TME: Tumor microenvironment.

## Data Availability

Data used to support the results of this study are available to readers from the authors.

## Conflicts of Interest

The authors declare that no commercial or financial relationships could be considered conflicts of interest in this study.

## Authors' Contributions

WZ and TH created the concepts and study techniques. DX, LD, and XY carried out data collection and analysis. DX prepared the manuscript; LD and XY made revisions, and WZ checked the final draft. Each author agreed to the contributions made in this work and reviewed and approved the final manuscript before submission.

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