MEDICAL SCIENCE MONITOR

MOLECULAR BIOLOGY

e-ISSN 1643-3750
© Med Sci Monit, 2018; 24: 9442-9464
DOI: 10.12659/MSM.913401

Received: 2018.09.28
Accepted: 2018.12.12
Published: 2018.12.29

Distinct Diagnostic and Prognostic Values of Kinesin Family Member Genes Expression in Patients with Breast Cancer

Authors' Contribution:
Study Design A
Data Collection B
Statistical Analysis C
Data Interpretation D
Manuscript Preparation E
Literature Search F
Funds Collection G

ACE 1 Xiaowei Song
CDE 2 Tengfang Zhang
ACD 3 Xiangkun Wang
ABC 3 Xiwen Liao
DF 3 Chuangye Han
DE 3 Chengkun Yang
EF 1 Ka Su
F 1 Wenlong Cao
C 4 Yizhen Gong
F 1 Zhu Chen
E 3 Quanfa Han
AEG 1 Jiehua Li

1 Department of Gastrointestinal Gland Surgery, The First Affiliated Hospital of Guangxi Medical University, Nanning, Guangxi, P.R. China
2 Department of Cardiothoracic Surgery, The First Affiliated Hospital of Guangxi Medical University, Nanning, Guangxi, P.R. China
3 Department of Hepatobiliary Surgery, The First Affiliated Hospital of Guangxi Medical University, Nanning, Guangxi, P.R. China
4 Department of Colorectal Surgery, The First Affiliated Hospital of Guangxi Medical University, Nanning, Guangxi, P.R. China

Corresponding Author: Jiehua Li, e-mail: lijiehua01@sina.com
Source of support: The current research was supported by the Guangxi Zhuang Autonomous Region Higher Education Research Project (No.: KY2015ZD031)

Background: This study investigated the diagnostic and prognostic values of kinesin superfamily proteins (KIFs) in breast cancer (BC) patients.

Material/Methods: All data were obtained from the Cancer Genome Atlas. DESeq was run to test for differentially expressed KIF genes. Patients were divided into high- and low-expression groups according to the median expression values of each KIF genes. Survival data were calculated using the Cox proportional hazard model. Comprehensive survival analysis was performed to evaluate the prognostic value of the prognostic signature. Gene set enrichment analysis (GSEA) was conducted to identify associated gene ontology and KEGG pathways.

Results: Bioinformatics analysis showed that all KIF genes were significantly enriched during DNA replication and the cell cycle, and co-expressed with each other. Thirteen KIF genes were differentially expressed in cancer and adjacent tissues, and high levels of *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* genes were significantly correlated with poor overall survival (OS). GSEA showed that BC patients with high expression of *KIF15, KIF20A, KIF23, KIF2C* and *KIF4A* were enriched in the cell cycle process, P53 regulation pathway and mismatch repair. Combinations of low expression of *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* were more highly correlated with favorable OS. Nomograms showed that the *KIF4A* risk score provided the maximum number of risk points (range 0–100), whereas other genes made a lower contribution.

Conclusions: We conclude that 13 KIF genes are differentially expressed in BC tumor tissues, and *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* are associated with prognostic factors in BC.

MeSH Keywords: Breast Neoplasms • Diagnosis • Kinesin • Prognosis • RNA

Full-text PDF: https://www.medscimonit.com/abstract/index/idArt/913401

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Background

Breast cancer (BC) remains the highest occurring cancer in women, in addition to being the third most frequent malignancy globally. In 2012, around 1.7 million persons worldwide had BC and nearly 500,000 died from the disease [1–4]. One among eight or 10 women will develop BC in their lifetime. BC mortality has decreased in North America as well as the European Union, but is still increasing in South America, Africa, and Asia. BC is the most common cause of cancer mortality in developing countries, compared with lung cancer in developed countries [5–7]. Genetic aspects and environmental exposure play a significant part in the etiology of BC [8,9]. The Human Genome Project has led to increasing attention being paid to cancer genetic susceptibility. A genetic factor which dysfunction amid normal tissues and tumors in the genome are the major potential sources of prognostic and diagnostic biomarkers [10]. Also, genes whose expression is interlinked with survival of BC might be prognostic biomarkers, as well as therapeutic targets [11–14]. As in other malignant neoplastic diseases, BC is considered to have dysfunction of numerous gene signaling pathways as well as networks that have an impact on tissue homeostasis.

There are 45 kinesin superfamily proteins (KIFs) with various functions in humans [15]. KIFs are involved in the molecular movements of axonal transportation. KIFs are ubiquitous in eukaryotes, and some are involved in transportation of vesicles and organelles inside cells [16,17]. KIF genes have been shown to play a crucial role in many tumors, and can be used to predict cancer diagnosis and prognosis [18,19] . It has been shown that the KIF family of genes is linked with BC [20,21], nevertheless, the joint analysis linked with multiple KIF family genes regarding BC have rarely been recorded. Complete examination of the diagnostic and prognostic values of KIF genes in BC requires additional investigation. The purpose of the present study was to explore the diagnostic and prognostic values of KIF gene expression in BC patients, on the basis of bioinformatics evaluation.

Material and Methods

Bioinformatics analysis of KIF genes

For analysis of the biological pathways and significance of the KIF family genes, a set of functional enrichment analysis for the KIF family was performed using the Database for Annotation, Visualization and Integrated Discovery (DAVID 6.8, *https://david.ncifcrf.gov/home.jsp*, accessed August 3, 2018). Enriched P values <0.05 were statistically significant. Gene–gene interactions of KIF family genes were investigated via GeneMANIA (*http://www.genemania.org/*, accessed August 9, 2018) [22]. Protein–protein interactions were examined by the Search Instrument for the Retrieval of Interacting Genes/Proteins (STRING, *https://string-db.org/*, accessed August 9, 2018) [23]. We also applied the Cytoscape (version 3.6.1) Biological Networks Gene Ontology (BiNGO) instrument for performing Gene Ontology (GO) evaluation on the KIF gene family [24].

Data source

The knowledge of the clinical of BC patients and RNA sequence based on the patients were gathered by the Cancer Genome Atlas (TCGA) database (*https://cancergenome.nih.gov/*, accessed June 7, 2018). Using the edgeR package in R, we normalized mRNA sequencing data and examined mRNA expression in normal tissues and BC. Genes with an accustomed P value <0.01 and $|\log_2$ fold-change (FC)| >2 were considered to be significantly different in BC and adjacent tissues. We regarded the genes as being differentially expressed mRNA (DEM). Clinical characteristics of patients with BC included gender, ethnicity (Asian, black, white or other), age at diagnosis (<65 or ≥65 years), and tumor stage.

Survival analysis

For each KIF DEM, patients were divided into low- and high-expression groups according to the median expression values of each KIF genes. By utilizing survival curves by Kaplan-Meier analysis with log-rank test, we assessed the prognostic significance of every clinical aspect as well as DEM from a criterion of P<0.05. The Cox proportional hazards model was utilized for evaluating the comparative risk in such differentially expressed genes on overall survival (OS). The mRNAs significantly associated with OS in the Cox proportional hazards model were considered to be prognostic mRNAs.

Correlation analysis

Pearson correlation coefficient was assessed for identifying correlations between the prognostic mRNAs.

Joint-effects analysis and nomograms

To assess thoroughly the prognostic model, joint analysis and nomograms were performed on the KIF DEM prognostic signature. On the basis of previous survival analysis, we divided the combined genes into high-, intermediate- and low-risk groups, completed survival analysis on 3 groups of patients, and established a Cox regression model. In addition to the joint analysis, we examined the predictive prognostic value of the risk scoring using nomograms to assess the correlation among clinical status as well as risk score within BC OS. The possible implication of risk scoring on the basis of predicting clinical characteristics has similarly been discovered. C-index

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

and calibration curve were considered with bootstrap self-sampling and internal verification.

Gene set enrichment analysis (GSEA)

The core concept in GSEA is to utilize a predefined group of genes (mainly by previous experimental outcomes or functional annotations) for ranking the genes in accordance with the extent of differential expression within the 2 types of samples, then verifying that the pre-established group of genes is supplemented at the bottom or top in the sorting table. To explore the differences in pathways as well as biological functions in the low- and high-expression sets of such prognostic KIF genes, GSEA (*http://software.broadinstitute.org/gsea/index.jsp*, accessed August 9, 2018) [25,26] was used to explore potential KEGG pathway and GO analysis within the Molecular Signatures Database (MSigDB) of c2 (curated gene sets) and c5 (GO gene sets) [27]. The criteria for significant enrichment gene sets in GSEA were: $P<0.05$ and false discovery rate (FDR) <0.25.

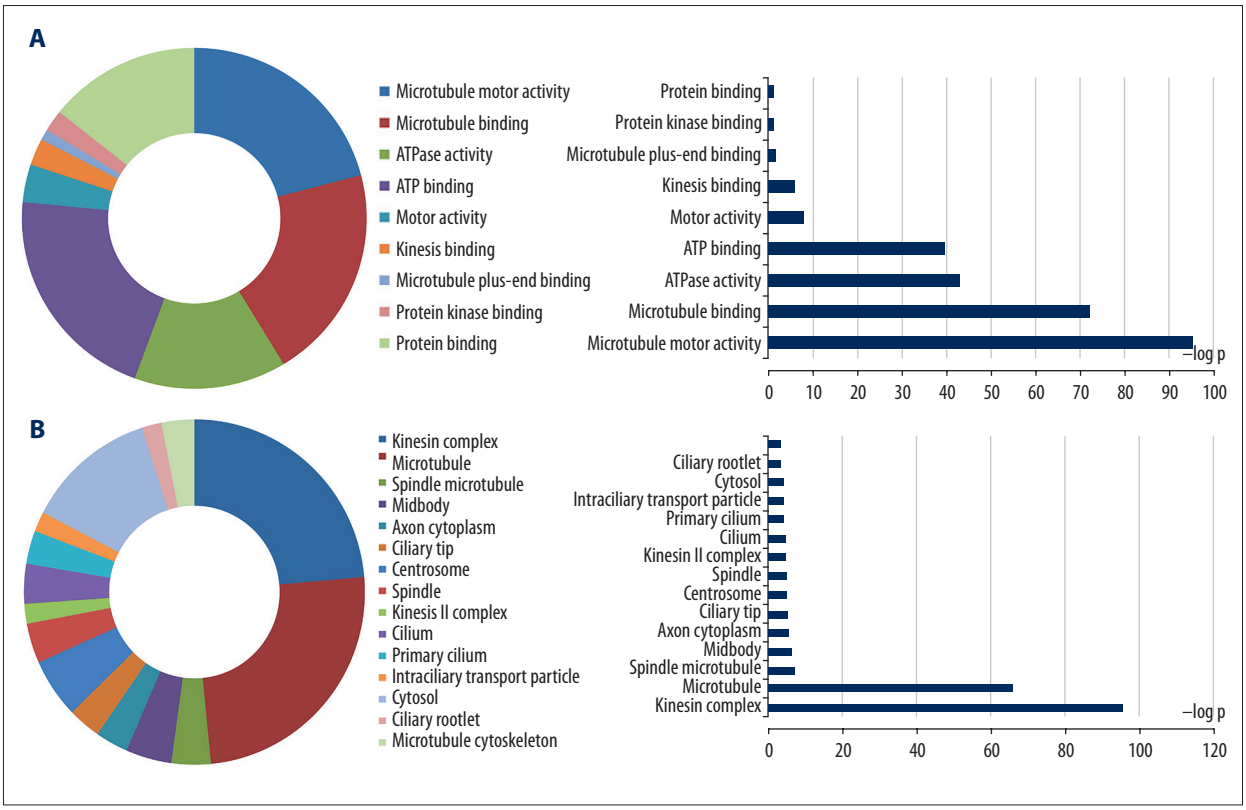
Statistical analysis

Log-rank assessment was utilized for comparing clinical aspects as well as univariate survival analysis of KIF genes. Clinicopathological parameters statistically linked to OS ($P<0.05$) were included in multivariate Cox proportional hazard regression models to adjust. Hazard ratios (HRs) and 95% confidence intervals (CIs) were used to assess the relative risk in many patients with BC. Multiple testing with the Benjamini–Hochberg procedure was used to control the FDR in GSEA. Statistical analysis was performed using SPSS 22.0 and R 3.5.1 software. $P<0.05$ was considered to be statistically significant.

Results

Bioinformatics analysis of the KIF family genes

Enrichment analysis of GO terms for the KIF family genes, performed with DAVID, showed that KIF genes had suggestive enrichment for microtubule-based movement, and biological functions mainly included mitotic metaphase plate congression, mitotic cytokinesis, mitotic spindle assembly, cell division, mitotic spindle midzone assembly, and positive regulation of cytokinesis (Figure 1). Gene–gene and protein–protein interaction networks confirmed that the KIF genes had solid protein homology as well as co-expression with one another at the protein as well as gene levels (Figure 2A, 2B). The focused KIF genetic acyclic graph constructed by BiNGO in Cytoscape similarly showed that the main biological roles were cell cycle progression, cellular processes, and microtubule-based processes (Figure 2C).



Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

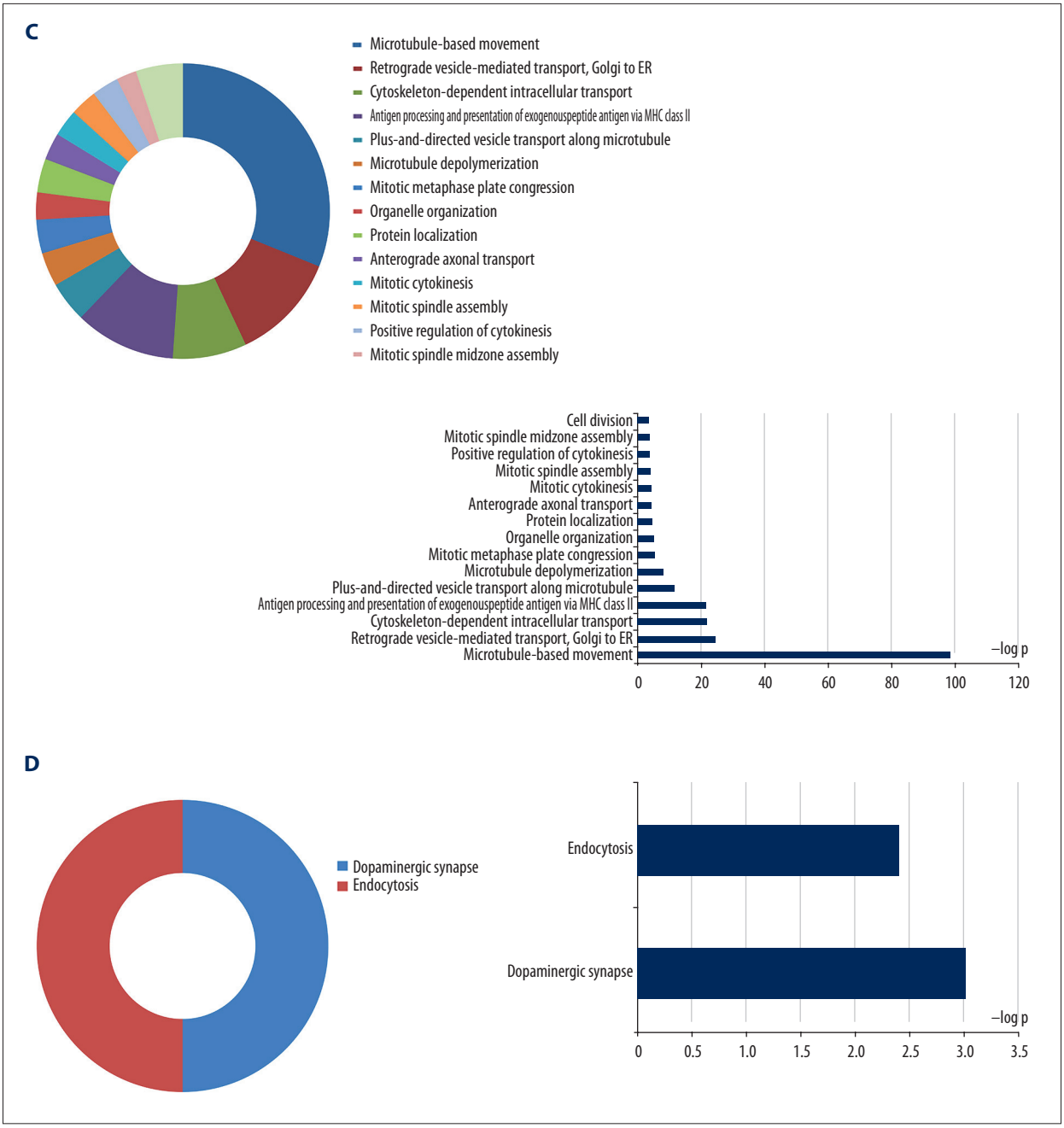


Figure 1. GO term and KEGG analysis of all the KIF family genes. GO term enrichments of KIF genes: (**A**) for MF; (**B**) for CC; (**C**) for BP. (**D**) KEGG enrichments of KIF genes. GO – gene ontology; KEGG – Kyoto Encyclopedia of Genes and Genomes; KIF – kinesin; MF – molecular function; CC – cellular component; BP – biological process.

Patient characteristics influencing differential KIF expression in BC

The $|\log_2 FC|$ of KIF family is shown in a histogram (Figure 3A). Thirteen KIF family genes met the standard of FDR <0.05 together with $|\log_2 FC| \geq 2$ (Table 1). A scatter plot produced using TCGA showed the difference in expression of the 13 KIF genes in invasive BC tissues compared with normal breast tissues (Figure 3B). Therefore, only the remaining 13 mRNAs were included in the step function screening to investigate the optimal combination, and all the mRNA expression data were $\log_2$ transformed for further analysis. The KIF genetic ROC analysis in the TCGA cohort specified that every KIF gene was highly accurate for discriminating normal breast and tumor tissues (area under the curve for the ROC curves in 11 KIF genes remained >0.9; Figure 4, Table 1).

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

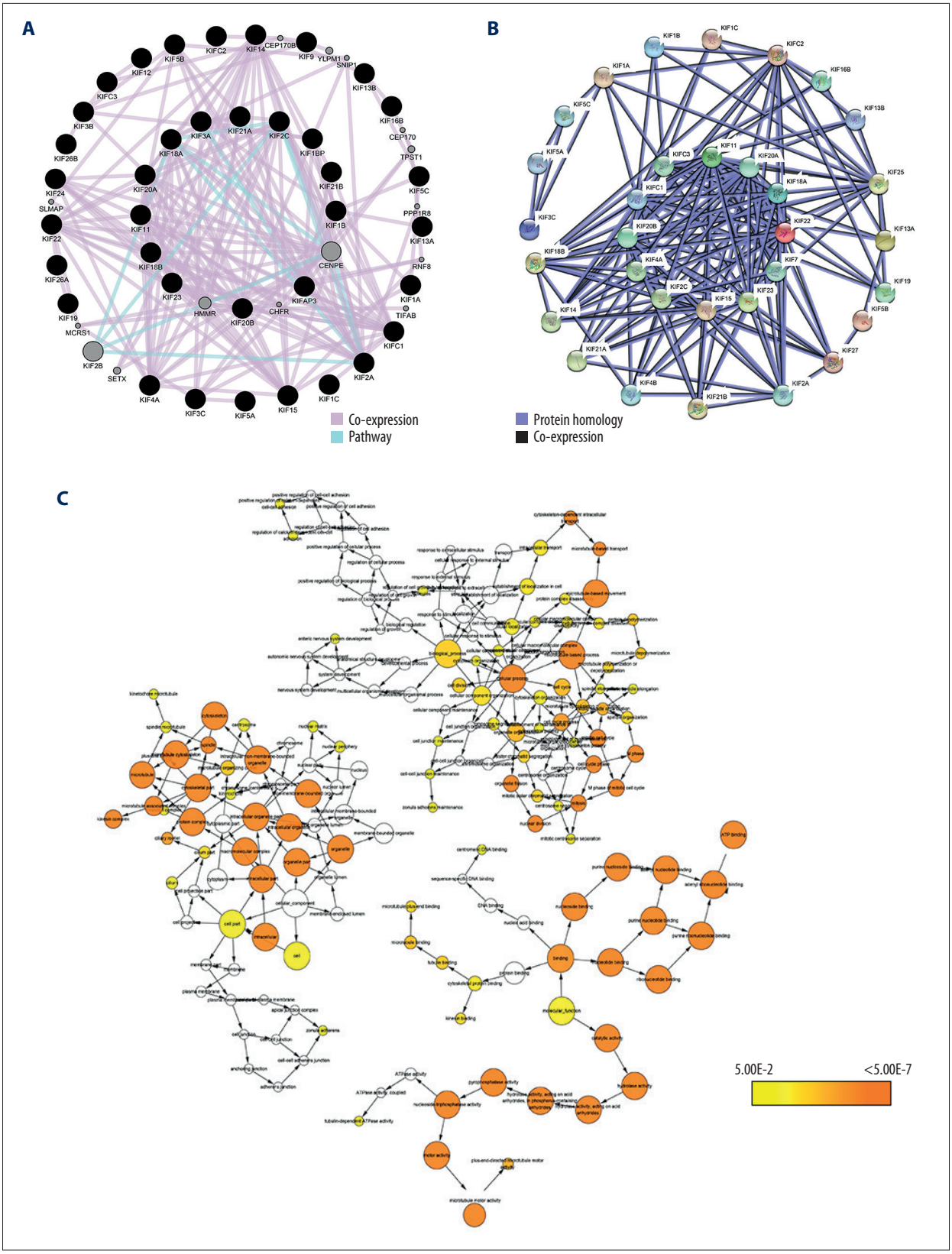


Figure 2. Protein–protein and gene–gene interaction networks of KIF genes. (**A**) GeneMANIA interaction networks. (**B**) Protein–protein interaction networks; (**C**) BiNGO analysis. KIF – kinesin; BiNGO – Biological Networks Gene Ontology tool.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

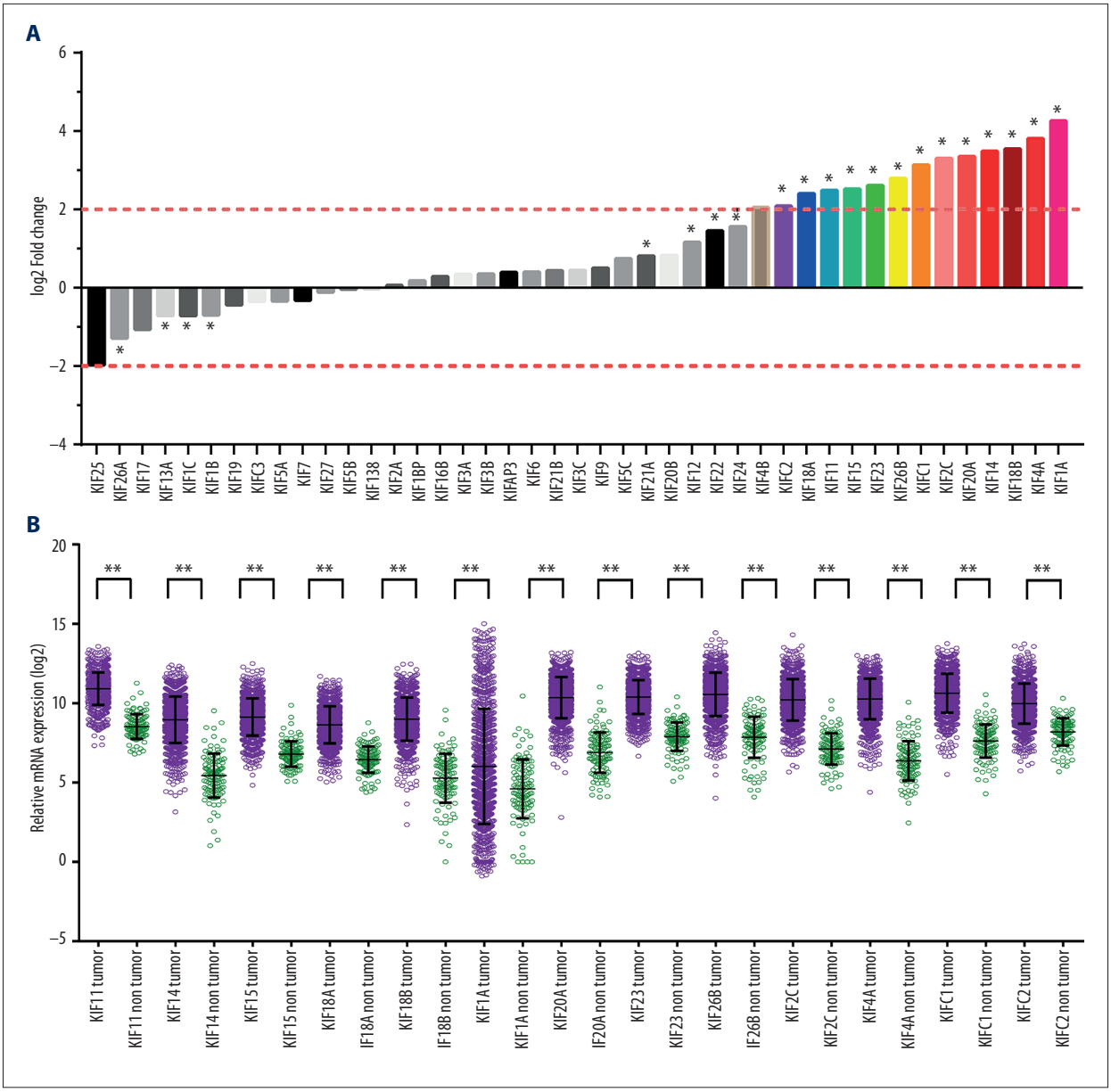


Figure 3. (**A**) Expression of KIF family genes. (**B**) Gene expression distribution of KIF genes in TCGA. * *P*<0.01. KIF – kinesin; TCGA – the Cancer Genome Atlas.

Table 1. The difference expression between BC patients and normal breast tissues.

Gene name	log2 fold change	P value	FDR	AUC	95%CI	P value
KIF4A	3.815	0.000	0.000	0.979	0.969–0.988	0.000
KIFC1	3.134	0.000	0.000	0.968	0.956–0.980	0.000
KIF18B	3.545	0.000	0.000	0.967	0.951–0.983	0.000
KIF2C	3.302	0.000	0.000	0.967	0.954–0.980	0.000
KIF20A	3.352	0.000	0.000	0.964	0.948–0.980	0.000
KIF23	2.609	0.000	0.000	0.962	0.949–0.975	0.000
KIF11	2.488	0.000	0.000	0.961	0.945–0.977	0.000

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 1 continued. The difference expression between BC patients and normal breast tissues.

Gene name	log2 fold change	P value	FDR	AUC	95%CI	P value
KIF14	3.489	0.000	0.000	0.955	0.938–0.973	0.000
KIF22	1.450	0.000	0.000	0.950	0.936–0.965	0.000
KIF15	2.519	0.000	0.000	0.943	0.924–0.962	0.000
KIF18A	2.405	0.000	0.000	0.935	0.917–0.952	0.000
KIF24	1.563	0.002	0.013	0.929	0.911–0.948	0.000
KIF26B	2.793	0.000	0.000	0.927	0.909–0.944	0.000
KIFC2	2.087	0.000	0.000	0.880	0.857–0.904	0.000
KIF26A	–1.291	0.006	0.027	0.867	0.840–0.895	0.000
KIF13A	–0.716	0.001	0.006	0.859	0.931–0.887	0.000
KIF17	–1.062	0.132	0.305	0.829	0.794–0.865	0.000
KIF25	–1.970	0.018	0.066	0.809	0.771–0.848	0.000
KIF1B	–0.693	0.002	0.013	0.797	0.763–0.832	0.000
KIF4B	2.058	0.298	0.532	0.781	0.744–0.818	0.000
KIF20B	0.825	0.027	0.092	0.765	0.730–0.800	0.000
KIF19	–0.436	0.534	0.747	0.757	0.724–0.790	0.000
KIF1C	–0.715	0.000	0.002	0.738	0.691–0.785	0.000
KIF21A	0.815	0.007	0.032	0.733	0.699–0.767	0.000
KIFAP3	0.395	0.096	0.242	0.720	0.686–0.754	0.000
KIF5A	–0.338	0.475	0.702	0.707	0.657–0.757	0.000
KIFC3	–0.346	0.261	0.486	0.704	0.664–0.743	0.000
KIF9	0.507	0.367	0.605	0.684	0.645–0.724	0.000
KIF7	–0.322	0.377	0.616	0.679	0.637–0.721	0.000
KIF3B	0.352	0.071	0.194	0.665	0.629–0.700	0.000
KIF13B	–0.027	0.815	0.927	0.626	0.592–0.659	0.000
KIF3A	0.348	0.343	0.580	0.617	0.569–0.664	0.000
KIF1BP	0.183	0.414	0.651	0.612	0.568–0.656	0.000
KIF12	1.161	0.000	0.002	0.605	0.557–0.653	0.000
KIF1A	4.263	0.000	0.000	0.604	0.565–0.644	0.000
KIF27	–0.115	0.693	0.857	0.598	0.545–0.652	0.001
KIF3C	0.444	0.182	0.381	0.568	0.532–0.604	0.018
KIF5B	–0.040	0.903	0.977	0.553	0.504–0.601	0.065
KIF16B	0.292	0.219	0.433	0.536	0.498–0.574	0.211
KIF21B	0.436	0.437	0.671	0.510	0.470–0.550	0.722
KIF6	0.406	0.772	0.904	0.509	0.471–0.547	0.765
KIF2A	0.065	0.772	0.904	0.507	0.465–0.549	0.805
KIF5C	0.742	0.023	0.083	0.506	0.463–0.550	0.826

FDR – false discovery rate; AUC – area under the curve; 95%CI – 95% confidence interval.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

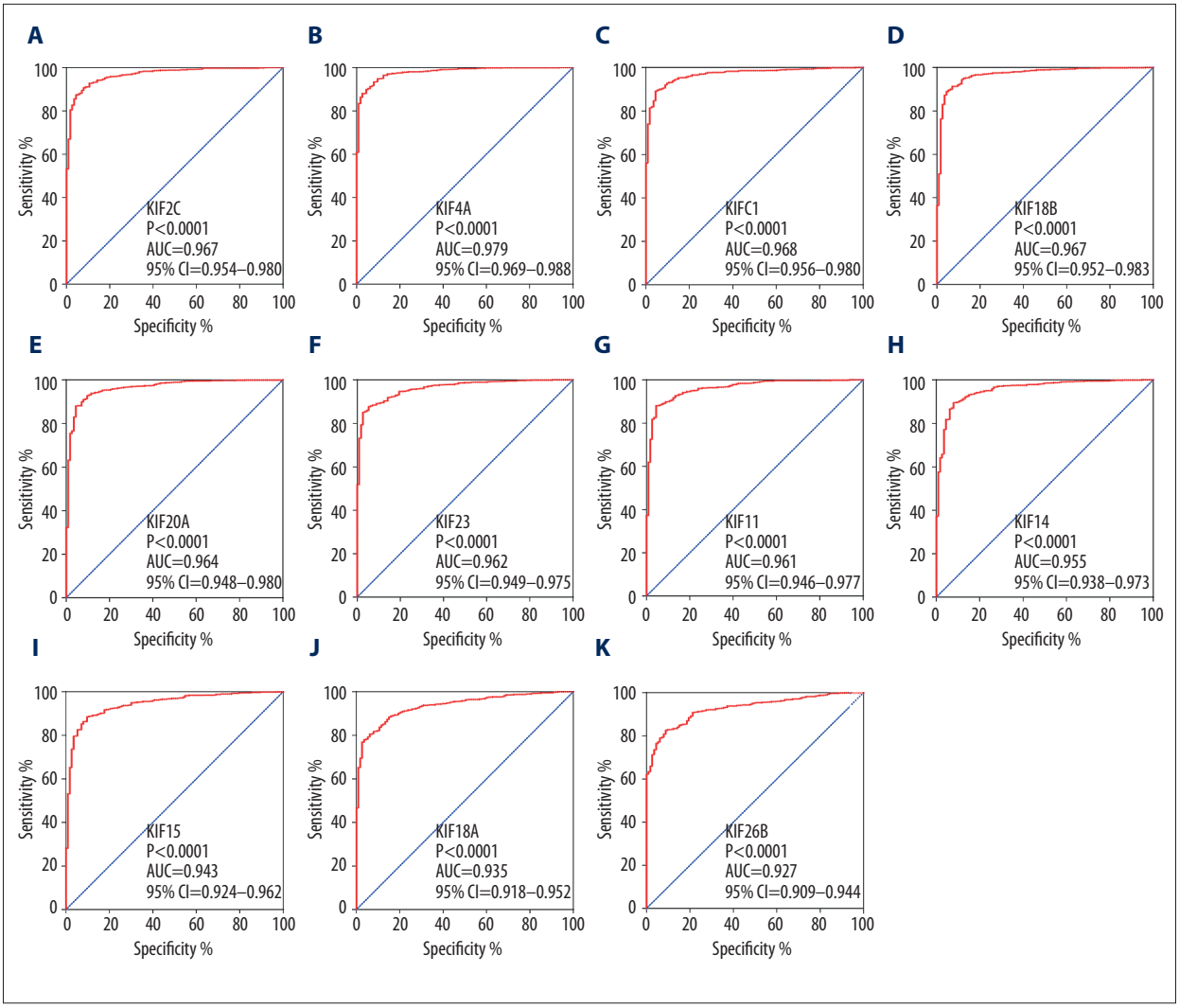


Figure 4. ROC curves (AUC>0.9) of KIF gens for distinguishing BC tumor tissue and adjacent normal tissues in TCGA. ROC curves of *KIF2C* (**A**), *KIF4A* (**B**), *KIFC1* (**C**), *KIF18B* (**D**), *KIF20A* (**E**), *KIF23* (**F**), *KIF11* (**G**), *KIF14* (**H**), *KIF15* (**I**), *KIF18A* (**J**), and *KIF26B* (**K**). KIF – kinesin; TCGA – the Cancer Genome Atlas; BC – breast cancer; AUC – area under the curve; ROC – receiver operating characteristic.

Survival analysis and association analysis

In the TCGA invasive BC cohort, patients with advanced tumor stage and age ≥65 years had an increased risk of invasive BC mortality (Table 2). Other patient characteristics, including gender and race, within the TCGA cohort did not show a significant association with OS of invasive BC.

Survival analysis of the 13 differentially expressed KIF genes is shown in Table 3. Patients with low expression of *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* genes in the TCGA invasive BC cohort had an extended OS (Table 3, Figure 5A–5E). However, only the *P* values of *KIF4A* reached significance (P=0.008). It was suggested that elevated expression of *KIF15* (adjusted *P*=0.045; adjusted HR=1.422; 95% CI=1.007–2.008), *KIF20A* (adjusted *P*=0.03; adjusted HR=1.467; 95% CI=1.038–2.072), *KIF23* (adjusted *P*=0.014; adjusted HR=1.54; 95% CI=1.09–2.175), *KIF2C* (adjusted *P*=0.001; adjusted HR=1.805; 95% CI=1.276–2.553) and *KIF4A* (adjusted *P*=0.001; adjusted HR=1.805; 95% CI=1.276–2.553) (Table 3) was related to poor OS within invasive BC, after adjusting for tumor stage and age.

After performing survival analysis within the TCGA cohorts, co-expression analysis of *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* in BC malignant tissues was evaluated using Pearson's correlation coefficient. The genes were co-expressed strongly with each other in the TCGA cohort (Figure 6).

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 2. Demographic and clinical data for 1055 BC patients.

Variables	Patients (n=1055)	No. of events	MST (days)	HR (95% CI)	Log-rank P
Race					0.534
White	732	109	3941	Ref	
Others	239	33	3873	1.132 (0.766–1.671)	
Missing	84				
Gender					0.854
Female	1043	148	3926	Ref	
Male	12	1	NA	0.832 (0.116–5.96)	
Age (years)					<0.001
≥65	719	88	6456	Ref	
<65	322	61	3418	2.18 (1.567–3.033)	
Missing	14				
Tumor stage					<0.001
I	175	15	3959	Ref	
II	596	65	4267	1.71 (0.974–2.999)	
III	241	43	3461	3.131 (1.738–5.641)	
IV	20	15	1034	13.481 (6.572–27.654)	
Missing	23				

MST – median survival time; HR – hazard ratio; CI – confidence interval.

Effect of combinations of KIF gene expression on OS

Based on KIF gene survival analysis, *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* were screened as prognostic genes by multivariate survival analysis. A joint-effects model was utilized for determining the combined influence of the 5 KIF genes on OS of BC patients. The diverse groups for this analysis were generated in accordance with expression of *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* (Tables 4–7). The Kaplan-Meier estimator with a log-rank evaluation was administered to evaluate the prognostic significance of the gene expression combinations represented by each group. Two selected groups showed that the BC patients with high expression of *KIF20A* and *KIF4A* or high expression of *KIF2C* and *KIF4A* had poor OS (Table 8). Within the evaluation of low *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and low *KIF4A* expression, the combinations in groups 4, 7, 10, 13, 16, 19, 22, 25 and 28 were highly correlated with favorable OS (all $P<0.05$; Table 8). In the analysis of high expression of *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A*, the combinations in groups 3, 6, 9, 12, 15, 18, 21, 24, 27 and 30 were highly correlated with poor OS (all $P<0.05$; Table 8).

GSEA

GSEA of the prognostic genes *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* was performed within the TCGA cohorts. The expression profiles of the genome-wide dataset in the TCGA-based cohorts were divided into 2 groups in accordance with the median prognostic *KIF* genetic values. GSEA outcomes of the TCGA cohort are shown in Figures 7A–7L, 8A–8L and 9A–9F), which suggested that their elevated expression remained linked with mismatch repair, P53 regulation pathway, and cell cycle progression.

Nomogram analysis

The nomogram was driven from rms as well as its supplementary packages on the base of information of patients having BC with comprehensive clinical evidence within TCGA. It showed that, among the 5 *KIF* genes, *KIF4A* had the greatest sum of risk points (ranging between 0 and 100), while the other genes made a considerably lower contribution (Figure 10A). By examining the conformity and discrimination using the nomogram model, bootstrap analysis on the bases of 1000 resampling tests had a C-index of 0.76 and 95% CI of 0.70–0.82. The discrimination is suitable. The calibration curve showed that the general point was close to the ideal curve of 45 degrees, indicating good compliance (Figure 10B, 10C).

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 3. Prognostic survival analysis according to the high or low level of 13 diagnostic KIF genes and OS.

Gene	Patients (n=1055)	Events	MST (days)	Crude HR (95% CI)	Crude P	Adjusted HR* (95% CI)*	Adjusted P*
KIF11							
High	527	82	4456	**1**		**1**	
Low	528	67	3736	1.106 (0.801–1.529)	0.54	1.402 (0.991–1.982)	0.056
KIF14							
High	527	76	7455	**1**		**1**	
Low	528	73	3736	1.028 (0.745–1.417)	0.869	1.187 (0.848–1.663)	0.318
KIF15							
High	527	80	6593	**1**		**1**	
Low	528	69	3736	1.068 (0.773–1.476)	0.688	1.422 (1.007–2.008)	**0.045**
KIF18A							
High	527	82	3945	**1**		**1**	
Low	528	67	3736	1.105 (0.799–1.527)	0.546	1.367 (0.971–1.924)	0.073
KIF18B							
High	527	78	3959	**1**		**1**	
Low	528	71	3736	1.143 (0.829–1.577)	0.415	1.224 (0.875–1.713)	0.238
KIF1A							
High	527	74	3945	**1**		**1**	
Low	528	75	3926	1.001 (0.726–1.38)	0.996	1.046 (0.748–1.461)	0.794
KIF20A							
High	527	87	3959	**1**		**1**	
Low	528	62	3736	1.273 (0.917–1.766)	0.148	1.467 (1.038–2.072)	**0.03**
KIF23							
High	527	83	3959	**1**		**1**	
Low	528	66	3736	1.23 (0.889–1.701)	0.21	1.54 (1.09–2.175)	**0.014**
KIF26B							
High	527	69	3472	**1**		**1**	
Low	528	80	3959	1.067 (0.771–1.475)	0.696	1.194 (0.848–1.682)	0.309
KIF2C							
High	527	84	3959	**1**		**1**	
Low	528	65	3926	1.341 (0.97–1.855)	0.075	1.805 (1.276–2.553)	**0.001**
KIF4A							
High	527	90	3941	**1**		**1**	
Low	528	59	3926	1.557 (1.121–2.162)	**0.008**	1.805 (1.276–2.553)	**0.001**
KIFC1							
High	527	82	4456	**1**		**1**	
Low	528	67	3492	1.106 (0.798–1.534)	0.545	1.273 (0.902–1.796)	0.17
KIFC2							
High	527	71	4456	**1**		**1**	
Low	528	78	3669	0.983 (0.712–1.358)	0.919	0.915 (0.653–1.281)	0.603

* Adjusted for age (stratified by 65 years) and tumor stage. KIF – kinesin; OS – overall survival; MST – median survival time; HR – hazard ratio; CI – confidence interval.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

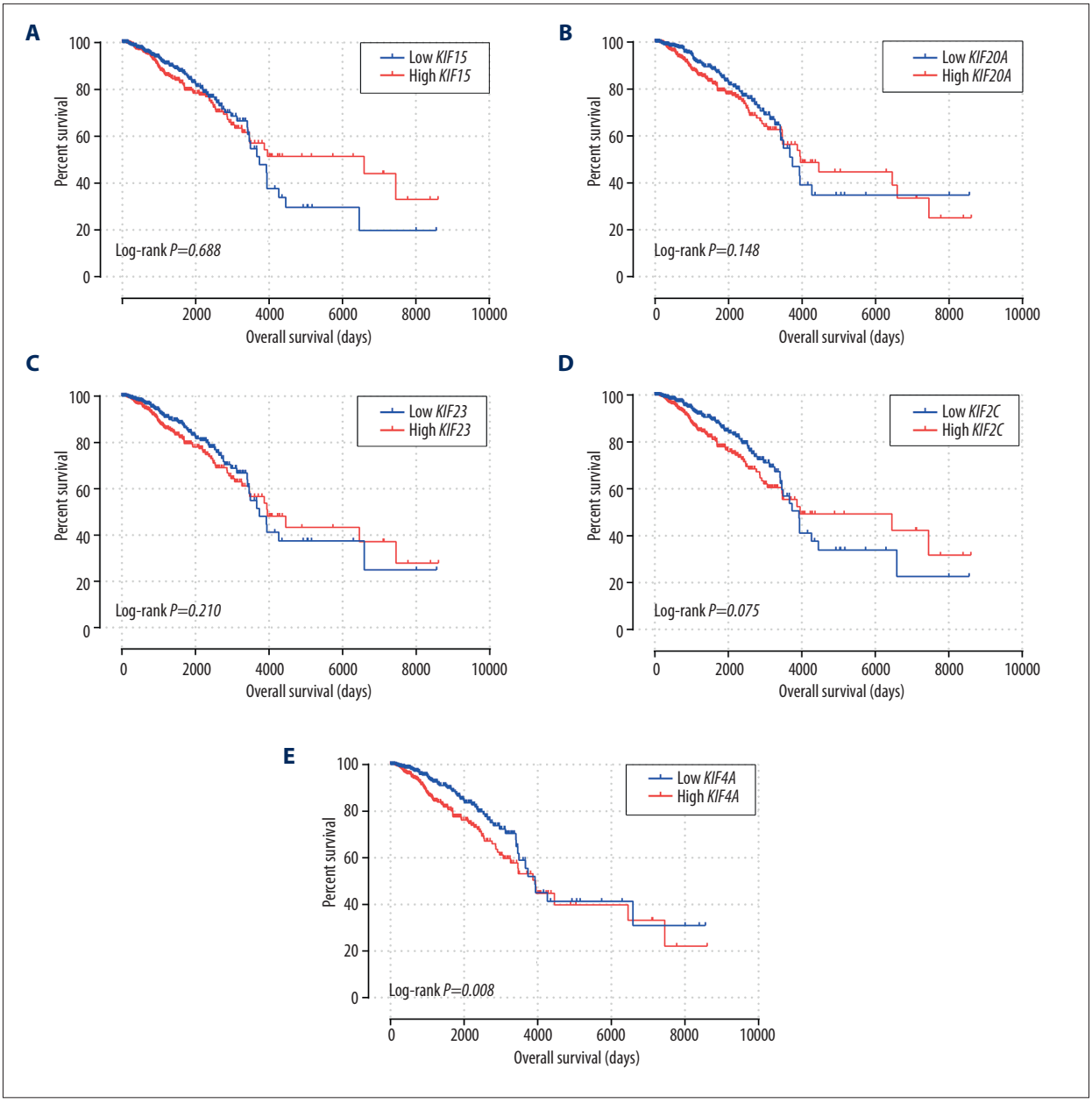


Figure 5. Kaplan-Meier survival curves for KIF genes in BC of TCGA cohort. OS stratified by *KIF15* (**A**), *KIF20A* (**B**), *KIF23* (**C**), *KIF2C* (**D**), and *KIF4A* (**E**). KIF – kinesin; TCGA – the Cancer Genome Atlas; BC – breast cancer; OS – overall survival.

Discussion

Kinesin motor activity is spatially as well as temporally controlled within mitosis to ensure that it occurs precisely with stable inward and outward forces. Nevertheless, over-expression of a few mitotic kinesins might produce further outward forces. This provokes a sequence of undesirable events, such as overshooting before anaphase, sister chromatid segregation before anaphase, increased spindle separation, and ultimately monopolar or bipolar spindle formation [28. Such things might cause imbalanced distribution of DNA, aneuploidy and a plethora of cancer phenotypes, together with metastatic and invasive behavior. Kinesin function might cause failed cytokinesis, imperfect spindle assembly and mitotic arrest, which stimulates apoptosis and killing of cancer cells [15]. Our evaluation of genetic function enrichment suggested that the KIF gene family is involved in biological processes of the cell cycle such as mitotic cytokinesis, mitotic spindle assembly, and positive regulation of cytokinesis. Our analysis established that *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* were co-expressed at the protein and gene levels.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

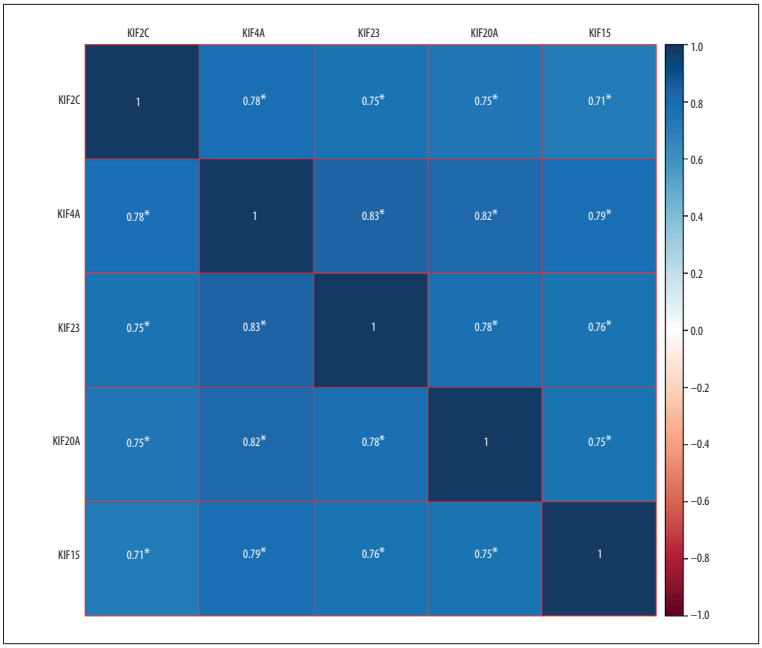


Figure 6. Co-expression heat map of *KIF2C*, *KIF4A*, *KIF23*, *KIF20A* and *KIF15* in TCGA BC patients. KIF – kinesin; TCGA – the Cancer Genome Atlas; BC – breast cancer.

Table 4. Grouping according to 2 selected genes.

Group	Combination	Group	Combination
1	Low KIF15 + low KIF20A	16	Low KIF20A + low KIF2C
2	Low KIF15 + high KIF20A	17	Low KIF20A + high KIF2C
	High KIF15 + low KIF20A		High KIF20A + low KIF2C
3	High KIF15 + high KIF20A	18	High KIF20A + high KIF2C
4	Low KIF15 + low KIF23	19	Low KIF20A + low KIF4A
5	Low KIF15 + high KIF23	20	Low KIF20A + high KIF4A
	High KIF15 + low KIF23		High KIF20A + low KIF4A
6	High KIF15 + high KIF23	21	High KIF20A + high KIF4A
7	Low KIF15 + low KIF2C	22	Low KIF23 + low KIF2C
8	Low KIF15 + high KIF2C	23	Low KIF23 + high KIF2C
	High KIF15 + low KIF2C		High KIF23 + low KIF2C
9	High KIF15 + high KIF2C	24	High KIF23 + high KIF2C
10	Low KIF15 + low KIF4A	25	Low KIF23 + low KIF4A
11	Low KIF15 + high KIF4A	26	Low KIF23 + high KIF4A
	High KIF15 + low KIF4A		High KIF23 + low KIF4A
12	High KIF15 + high KIF4A	27	High KIF23 + high KIF4A
13	Low KIF20A + low KIF23	28	Low KIF2C + low KIF4A
14	Low KIF20A + high KIF23	29	Low KIF2C + high KIF4A
	High KIF20A + low KIF23		High KIF2C + low KIF4A
15	High KIF20A + high KIF23	30	High KIF2C + high KIF4A

KIF – kinesin.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 5. Grouping according to 3 selected genes.

Group	Combination	Group	Combination
a	Low KIF15 + low KIF20A + low KIF23	A	Low KIF15 + low KIF2C + low KIF4A
b	Low KIF15 + high KIF20A + low KIF23	B	High KIF15 + low KIF2C + low KIF4A
	High KIF15 + low KIF20A + low KIF23		Low KIF15 + high KIF2C + low KIF4A
	Low KIF15 + low KIF20A + high KIF23		Low KIF15 + low KIF2C + high KIF4A
	High KIF15 + high KIF20A + low KIF23		High KIF15 + high KIF2C + low KIF4A
	Low KIF15 + high KIF20A + high KIF23		High KIF15 + low KIF2C + high KIF4A
	High KIF15 + low KIF20A + high KIF23		Low KIF15 + high KIF2C + high KIF4A
c	High KIF15 + high KIF20A + high KIF23	C	High KIF15 + high KIF2C + high KIF4A
d	Low KIF15 + low KIF20A + low KIF2C	D	Low KIF20A + low KIF23 + low KIF2C
e	High KIF15 + high KIF20A + low KIF2C	E	High KIF20A + low KIF23 + low KIF2C
	Low KIF15 + high KIF20A + low KIF2C		Low KIF20A + high KIF23 + low KIF2C
	High KIF15 + low KIF20A + low KIF2C		Low KIF20A + low KIF23 + high KIF2C
	Low KIF15 + high KIF20A + high KIF2C		High KIF20A + high KIF23 + low KIF2C
	High KIF15 + low KIF20A + high KIF2C		High KIF20A + low KIF23 + high KIF2C
	Low KIF15 + low KIF20A + high KIF2C		Low KIF20A + high KIF23 + high KIF2C
f	High KIF15 + high KIF20A + high KIF2C	F	High KIF20A + high KIF23 + high KIF2C
g	Low KIF15 + low KIF20A + low KIF4A	G	Low KIF20A + low KIF23 + low KIF4A
h	Low KIF15 + high KIF20A + low KIF4A	H	High KIF20A + low KIF23 + low KIF4A
	High KIF15 + low KIF20A + low KIF4A		Low KIF20A + high KIF23 + low KIF4A
	Low KIF15 + high KIF20A + high KIF4A		Low KIF20A + low KIF23 + high KIF4A
	High KIF15 + low KIF20A + high KIF4A		High KIF20A + high KIF23 + low KIF4A
	High KIF15 + high KIF20A + low KIF4A		High KIF20A + low KIF23 + high KIF4A
	Low KIF15 + low KIF20A + high KIF4A		Low KIF20A + high KIF23 + high KIF4A
i	High KIF15 + high KIF20A + high KIF4A	I	High KIF20A + high KIF23 + high KIF4A
j	Low KIF15 + low KIF23 + low KIF2C	J	Low KIF20A + low KIF2C + low KIF4A
k	High KIF15 + low KIF23 + low KIF2C	K	High KIF20A + low KIF2C + low KIF4A
	Low KIF15 + high KIF23 + low KIF2C		Low KIF20A + high KIF2C + low KIF4A
	Low KIF15 + low KIF23 + high KIF2C		Low KIF20A + low KIF2C + high KIF4A
	High KIF15 + high KIF23 + low KIF2C		High KIF20A + high KIF2C + low KIF4A
	High KIF15 + low KIF23 + high KIF2C		High KIF20A + low KIF2C + high KIF4A
	Low KIF15 + high KIF23 + high KIF2C		Low KIF20A + high KIF2C + high KIF4A
l	High KIF15 + high KIF23 + high KIF2C	L	High KIF20A + high KIF2C + high KIF4A
m	Low KIF15 + low KIF23 + low KIF4A	M	Low KIF23 + low KIF2C + low KIF4A
n	High KIF15 + low KIF23 + low KIF4A	N	Low KIF23 + low KIF2C + high KIF4A
	Low KIF15 + high KIF23 + low KIF4A		High KIF23 + low KIF2C + low KIF4A
	Low KIF15 + low KIF23 + high KIF4A		Low KIF23 + high KIF2C + low KIF4A
	High KIF15 + high KIF23 + low KIF4A		High KIF23 + high KIF2C + low KIF4A
	High KIF15 + high KIF23 + high KIF4A		High KIF23 + low KIF2C + high KIF4A
	Low KIF15 + high KIF23 + high KIF4A		Low KIF23 + high KIF2C + high KIF4A
o	High KIF15 + low KIF23 + high KIF4A	O	High KIF23 + high KIF2C + high KIF4A

KIF – kinesin.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 6. Grouping according to 4 selected genes.

Group	Combination	Group	Combination
I	Low KIF15 + low KIF20A + low KIF23 + low KIF2C	X	Low KIF15 + low KIF23 + low KIF2C + low KIF4A
II	High KIF15 + high KIF20A + low KIF23 + low KIF2C	XI	High KIF15 + low KIF23 + low KIF2C + low KIF4A
	Low KIF15 + high KIF20A + high KIF23 + high KIF2C		High KIF15 + low KIF23 + high KIF2C + high KIF4A
	High KIF15 + low KIF20A + high KIF23 + high KIF2C		High KIF15 + high KIF23 + low KIF2C + high KIF4A
	High KIF15 + high KIF20A + low KIF23 + high KIF2C		High KIF15 + high KIF23 + high KIF2C + low KIF4A
	High KIF15 + high KIF20A + high KIF23 + low KIF2C		Low KIF15 + high KIF23 + low KIF2C + high KIF4A
	Low KIF15 + high KIF20A + low KIF23 + high KIF2C		Low KIF15 + high KIF23 + high KIF2C + low KIF4A
	Low KIF15 + high KIF20A + high KIF23 + low KIF2C		High KIF15 + low KIF23 + high KIF2C + low KIF4A
	High KIF15 + low KIF20A + high KIF23 + low KIF2C		High KIF15 + low KIF23 + low KIF2C + high KIF4A
	High KIF15 + low KIF20A + low KIF23 + high KIF2C		High KIF15 + high KIF23 + low KIF2C + low KIF4A
	High KIF15 + low KIF20A + low KIF23 + low KIF2C		Low KIF15 + high KIF23 + low KIF2C + low KIF4A
	Low KIF15 + high KIF20A + low KIF23 + low KIF2C		Low KIF15 + low KIF23 + high KIF2C + low KIF4A
	Low KIF15 + low KIF20A + high KIF23 + low KIF2C		Low KIF15 + low KIF23 + low KIF2C + high KIF4A
	Low KIF15 + low KIF20A + low KIF23 + high KIF2C		Low KIF15 + low KIF23 + high KIF2C + high KIF4A
	Low KIF15 + low KIF20A + high KIF23 + high KIF2C		Low KIF15 + high KIF23 + high KIF2C + high KIF4A
III	High KIF15 + high KIF20A + high KIF23 + high KIF2C	XII	High KIF15 + high KIF23 + high KIF2C + high KIF4A
IV	Low KIF15 + low KIF20A + low KIF23 + low KIF4A	XIII	Low KIF20A + low KIF23 + low KIF2C + low KIF4A
V	High KIF15 + high KIF20A + low KIF23 + low KIF4A	XIV	High KIF20A + low KIF23 + low KIF2C + low KIF4A
	Low KIF15 + high KIF20A + high KIF23 + high KIF4A		High KIF20A + low KIF23 + high KIF2C + high KIF4A
	High KIF15 + low KIF20A + high KIF23 + high KIF4A		High KIF20A + high KIF23 + low KIF2C + high KIF4A
	High KIF15 + high KIF20A + low KIF23 + high KIF4A		High KIF20A + high KIF23 + high KIF2C + low KIF4A
	High KIF15 + high KIF20A + high KIF23 + low KIF4A		Low KIF20A + high KIF23 + low KIF2C + high KIF4A
	Low KIF15 + high KIF20A + low KIF23 + high KIF4A		Low KIF20A + high KIF23 + high KIF2C + low KIF4A
	Low KIF15 + high KIF20A + high KIF23 + low KIF4A		High KIF20A + low KIF23 + high KIF2C + low KIF4A
	High KIF15 + low KIF20A + high KIF23 + low KIF4A		High KIF20A + low KIF23 + low KIF2C + high KIF4A
	High KIF15 + low KIF20A + low KIF23 + high KIF4A		High KIF20A + high KIF23 + low KIF2C + low KIF4A
	High KIF15 + low KIF20A + low KIF23 + low KIF4A		Low KIF20A + high KIF23 + low KIF2C + low KIF4A
	Low KIF15 + high KIF20A + low KIF23 + low KIF4A		Low KIF20A + low KIF23 + high KIF2C + low KIF4A
	Low KIF15 + low KIF20A + high KIF23 + low KIF4A		Low KIF20A + low KIF23 + low KIF2C + high KIF4A
	Low KIF15 + low KIF20A + low KIF23 + high KIF4A		Low KIF20A + low KIF23 + high KIF2C + high KIF4A
	Low KIF15 + low KIF20A + high KIF23 + high KIF4A		Low KIF20A + high KIF23 + high KIF2C + high KIF4A
VI	High KIF15 + high KIF20A + high KIF23 + high KIF4A	XV	High KIF20A + high KIF23 + high KIF2C + high KIF4A
VII	Low KIF15 + low KIF20A + low KIF2C + low KIF4A		

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 6 continued. Grouping according to 4 selected genes.

Group	Combination	Group	Combination
VIII	High KIF15 + high KIF20A + low KIF2C + low KIF4A		
	Low KIF15 + high KIF20A + high KIF2C + high KIF4A		
	High KIF15 + low KIF20A + high KIF2C + high KIF4A		
	High KIF15 + high KIF20A + low KIF2C + high KIF4A		
	High KIF15 + high KIF20A + high KIF2C + low KIF4A		
	Low KIF15 + high KIF20A + low KIF2C + high KIF4A		
	Low KIF15 + high KIF20A + high KIF2C + low KIF4A		
	High KIF15 + low KIF20A + high KIF2C + low KIF4A		
	High KIF15 + low KIF20A + low KIF2C + high KIF4A		
	High KIF15 + low KIF20A + low KIF2C + low KIF4A		
	Low KIF15 + high KIF20A + low KIF2C + low KIF4A		
	Low KIF15 + low KIF20A + high KIF2C + low KIF4A		
	Low KIF15 + low KIF20A + low KIF2C + high KIF4A		
	Low KIF15 + low KIF20A + high KIF2C + high KIF4A		
IX	High KIF15 + high KIF20A + high KIF2C + high KIF4A		

KIF – kinesin.

We found 5 KIF genes of diagnostic and prognostic value. Extensive studies have reported these genes as potential diagnostic markers in multiple cancers. Among the 5 genes, *KIF15* is likewise overexpressed in lung adenocarcinoma and might play a significant role in modifying the cell cycle [29]. Likewise, *KIF15* promotes proliferation of pancreatic cancer cells via the MEK/ERK pathway [30]. *KIF15* is overexpressed in BC cells and might have potential as a novel therapeutic target and a prognostic factor in endocrine-therapy-resistant BC [31]. We found that expression of *KIF15* mRNA was significantly higher in BC than in adjacent tissues, and elevated expression of *KIF15* in patients with BC was associated with poor OS. Our results agreed with previous studies that designated the *KIF15* as an oncogene in BC.

In 2005, a study by Keisuke et al. [32] found that *KIF20A* was overexpressed in pancreatic cancer according to cDNA microarray analysis, and down-regulation of *KIF20A* significantly decreased tumor cell proliferation, confirming that *KIF20A* is carcinogenic in pancreatic cancer. Numerous studies have shown that *KIF20A* also has carcinogenic traits in various other cancers, such as nasopharyngeal carcinoma, liver cancer, melanoma, lung adenocarcinoma and glioma [33]. It has been suggested that the *KIF20A* gene is a potential diagnostic biomarker. We found that *KIF20A* has differential expression in BC and adjacent tissues, and high expression of *KIF20A* is related to poor OS in patients with BC, so it might also be a prognostic biomarker.

It has been found that *KIF23* is up-regulated in patients with hepatocellular carcinoma, and it may be a marker for OS [34]. Zou et al. [31] showed that *KIF4A*, *KIF15*, *KIF20A* and *KIF23* expression was significant in proliferating BC cells. They also showed that, among patients treated with tamoxifen, high expression of these 4 genes was highly correlated with poor recurrence-free survival. It has been suggested that over-expression of *KIF23* is a valuable independent prognostic factor in lung tumors, particularly lung adenocarcinoma, and patients with p-stage I tumor stage and high expression of *KIF23* have poorer survival than those with low expression [35]. In addition, the multivariate Cox proportional hazards model in our study, which was based on expression of *KIF23*, likewise divided patients in low- and high-expression groups, and patients with high expression had poor OS.

Nowadays, it is certain that *KIF4A* performs a significant role in cancer development and progression. Numerous studies have shown that *KIF4A* is a potential contributor to several malignant tumors, such as lung cancer [36], breast cancer [37], cervical cancer [38], hepatocellular carcinoma [39], and oral cancer [40]. Our results were consistent with previous studies. We also found that BC patients with high expression of *KIF4A* had poor OS compared with patients with low expression.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 7. Grouping according to 5 selected genes.

Group	Combination
①	Low KIF15 + low KIF20A + low KIF23 + low KIF2C + low KIF4A
②	High KIF15 + high KIF20A + low KIF23 + low KIF2C + low KIF4A
	High KIF15 + low KIF20A + low KIF23 + low KIF2C + low KIF4A
	Low KIF15 + high KIF20A + low KIF23 + low KIF2C + low KIF4A
	Low KIF15 + low KIF20A + high KIF23 + low KIF2C + low KIF4A
	Low KIF15 + low KIF20A + low KIF23 + high KIF2C + low KIF4A
	Low KIF15 + low KIF20A + low KIF23 + low KIF2C + high KIF4A
	High KIF15 + low KIF20A + high KIF23 + low KIF2C + low KIF4A
	High KIF15 + low KIF20A + low KIF23 + high KIF2C + low KIF4A
	High KIF15 + low KIF20A + low KIF23 + low KIF2C + high KIF4A
	Low KIF15 + high KIF20A + high KIF23 + low KIF2C + low KIF4A
	Low KIF15 + high KIF20A + low KIF23 + high KIF2C + low KIF4A
	Low KIF15 + high KIF20A + low KIF23 + low KIF2C + high KIF4A
	Low KIF15 + low KIF20A + high KIF23 + high KIF2C + low KIF4A
	Low KIF15 + low KIF20A + high KIF23 + low KIF2C + high KIF4A
	Low KIF15 + low KIF20A + low KIF23 + high KIF2C + high KIF4A
	Low KIF15 + high KIF20A + low KIF23 + high KIF2C + high KIF4A
	Low KIF15 + high KIF20A + high KIF23 + low KIF2C + high KIF4A
	Low KIF15 + high KIF20A + high KIF23 + high KIF2C + low KIF4A
	High KIF15 + low KIF20A + low KIF23 + high KIF2C + high KIF4A
	High KIF15 + low KIF20A + high KIF23 + low KIF2C + high KIF4A
	High KIF15 + low KIF20A + high KIF23 + high KIF2C + low KIF4A
	High KIF15 + high KIF20A + low KIF23 + low KIF2C + high KIF4A
	High KIF15 + high KIF20A + low KIF23 + high KIF2C + low KIF4A
	High KIF15 + high KIF20A + high KIF23 + low KIF2C + low KIF4A
	Low KIF15 + high KIF20A + high KIF23 + high KIF2C + high KIF4A
	High KIF15 + low KIF20A + high KIF23 + high KIF2C + high KIF4A
	High KIF15 + high KIF20A + low KIF23 + high KIF2C + high KIF4A
	High KIF15 + high KIF20A + high KIF23 + low KIF2C + high KIF4A
	High KIF15 + high KIF20A + high KIF23 + high KIF2C + low KIF4A
	Low KIF15 + low KIF20A + high KIF23 + high KIF2C + high KIF4A
③	High KIF15 + high KIF20A + high KIF23 + high KIF2C + high KIF4A

KIF – kinesin.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 8. Joint analysis of the prognostic value of combination of KIF15, KIF20A, KIF23, KIF2C, and KIF4A expression of BC.

Group	Patients	MST (days)	Crude p	Crude HR	Adjusted p	Adjusted HR(95% CI) *
1	432	3736	0.367	1	0.065	1
2	192	3941	0.185	1.354 (0.865–2.121)	0.16	1.419 (0.87–2.313)
3	431	6593	0.291	1.219 (0.844–1.759)	**0.021**	1.578 (1.073–2.322)
4	444	3669	0.439	1	**0.049**	1
5	168	4456	0.229	1.328 (0.836–2.11)	0.223	1.363 (0.828–2.245)
6	443	3959	0.356	1.184 (0.827–1.695)	**0.014**	1.615 (1.1–2.372)
7	428	3736	0.398	1	**0.008**	1
8	200	6456	0.279	1.296 (0.811–2.071)	**0.021**	1.825 (1.097–3.037)
9	427	3959	0.23	1.244 (0.871–1.778)	**0.003**	1.776 (1.211–2.604)
10	436	3669	0.237	1	**0.013**	1
11	184	6456	0.396	1.223 (0.768–1.946)	0.196	1.39 (0.843–2.29)
12	435	3873	0.09	1.369 (0.952–1.967)	**0.003**	1.787 (1.215–2.63)
13	450	3736	0.292	1	**0.038**	1
14	156	3461	0.265	1.317 (0.812–2.137)	0.116	1.508 (0.904–2.517)
15	449	3959	0.14	1.308 (0.916–1.87)	**0.012**	1.625 (1.112–2.374)
16	441	3736	0.221	1	**0.008**	1
17	174	4456	0.546	1.169 (0.704–1.94)	0.652	1.133 (0.659–1.949)
18	440	3959	0.083	1.366 (0.96–1.944)	**0.003**	1.76 (1.21–2.56)
19	454	3736	0.051	1	**0.011**	1
20	148	6593	0.772	0.919 (0.518–1.631)	0.735	1.114 (0.598–2.074)
21	453	3941	**0.032**	1.463 (1.034–2.07)	**0.004**	1.715 (1.188–2.475)
22	445	3736	0.207	1	**0.006**	1
23	166	3941	0.197	1.379 (0.846–2.249)	0.394	1.258 (0.742–2.131)
24	444	3959	0.101	1.344 (0.944–1.914)	**0.002**	1.844 (1.262–2.696)
25	453	3736	0.070	1	**0.004**	1
26	150	NUM	0.805	0.93 (0.525–1.648)	0.932	1.026 (0.573–1.837)
27	452	3941	**0.040**	1.436 (1.017–2.027)	**0.002**	1.787 (1.236–2.585)
28	457	3736	**0.033**	1	**0.001**	1
29	142	4456	0.751	0.909 (0.505–1.636)	0.457	0.785 (0.414–1.486)
30	456	3873	**0.020**	1.503 (1.066–2.12)	**0.001**	1.93 (1.34–2.78)
a	402	3736	0.343	1	**0.042**	1
b	258	3941	0.157	1.354 (0.89–2.059)	0.097	1.466 (0.933–2.302)
c	395	3959	0.298	1.225 (0.836–1.794)	**0.013**	1.674 (1.113–2.516)
d	390	3736	0.37	1	**0.04**	1
e	283	3941	0.232	1.296 (0.847–1.983)	0.129	1.426 (0.902–2.254)
f	382	3959	0.211	1.274 (0.872–1.861)	**0.011**	1.675 (1.123–2.497)
g	399	3736	0.335	1	**0.028**	1
h	262	4456	0.45	1.181 (0.767–1.818)	0.225	1.335 (0.837–2.131)
i	394	3959	0.139	1.328 (0.912–1.934)	**0.008**	1.713 (1.152–2.548)
j	399	3669	0.34	1	**0.018**	1
k	267	4456	0.168	1.348 (0.882–2.06)	0.088	1.487 (0.942–2.346)

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

Table 8 continued. Joint analysis of the prognostic value of combination of KIF15, KIF20A, KIF23, KIF2C, and KIF4A expression of BC.

Group	Patients	MST (days)	Crude p	Crude HR	Adjusted p	Adjusted HR(95% CI) *
l	389	3959	0.262	1.24 (0.851–1.805)	**0.005**	1.787 (1.193–2.675)
m	403	3669	0.44	1	**0.019**	1
n	251	6456	0.519	1.154 (0.747–1.782)	0.316	1.267 (0.798–2.011)
o	401	3873	0.2	1.275 (0.879–1.849)	**0.006**	1.763 (1.181–2.631)
A	398	3669	0.334	1	**0.018**	1
B	263	6456	0.558	1.14 (0.735–1.767)	0.315	1.273 (0.795–2.04)
C	394	3959	0.141	1.321 (0.912–1.913)	**0.005**	1.749 (1.18–2.593)
D	411	3736	0.424	1	**0.026**	1
E	248	3941	0.312	1.252 (0.809–1.937)	0.316	1.269 (0.797–2.023)
F	396	6456	0.225	1.258 (0.869–1.821)	**0.007**	1.721 (1.158–2.559)
G	419	3736	0.2	1	**0.023**	1
H	227	6593	0.885	1.035 (0.649–1.651)	0.46	1.204 (0.735–1.973)
I	409	3941	0.096	1.358 (0.947–1.949)	**0.007**	1.691 (1.151–2.484)
J	414	3736	0.081	1	**0.002**	1
K	232	4456	0.964	0.989 (0.613–1.597)	0.97	0.99 (0.591–1.658)
L	409	3873	**0.046**	1.44 (1.006–2.061)	**0.002**	1.823 (1.248–2.664)
M	419	3736	0.218	1	**0.002**	1
N	229	4456	0.891	1.033 (0.646–1.653)	0.965	0.989 (0.6–1.629)
O	407	3959	0.103	1.346 (0.942–1.925)	**0.002**	1.865 (1.268–2.742)
I	375	3736	0.391	1	**0.044**	1
II	324	3941	0.179	1.324 (0.879–1.995)	0.103	1.44 (0.929–2.231)
III	356	3959	0.353	1.203 (0.815–1.777)	**0.013**	1.693 (1.115–2.569)
IV	381	3736	0.477	1	**0.038**	1
V	304	4456	0.338	1.224 (0.81–1.852)	0.189	1.347 (0.864–2.099)
VI	370	3959	0.254	1.252 (0.851–1.843)	**0.01**	1.715 (1.135–2.593)
VII	375	3736	0.381	1	**0.037**	1
VIII	315	4456	0.376	1.208 (0.795–1.837)	0.186	1.355 (0.864–2.124)
IX	365	3959	0.169	1.309 (0.892–1.921)	**0.01**	1.701 (1.135–2.55)
X	381	3669	0.544	1	**0.026**	1
XI	306	4456	0.441	1.178 (0.776–1.789)	0.257	1.294 (0.829–2.02)
XII	368	3959	0.285	1.231 (0.841–1.801)	**0.007**	1.752 (1.163–2.638)
XIII	394	3736	0.41	1	**0.015**	1
XIV	287	3941	0.553	1.138 (0.742–1.745)	0.479	1.179 (0.747–1.859)
XV	374	3959	0.182	1.29 (0.887–1.876)	**0.006**	1.768 (1.182–2.645)
①	364	3736	0.455	1	**0.045**	1
②	348	3941	0.24	1.275 (0.85–1.912)	0.128	1.4 (0.908–2.158)
③	343	3959	0.316	1.224 (0.824–1.816)	**0.013**	1.708 (1.119–2.606)

* Adjusted for age (stratified by 65 years) and tumor stage. KIF – kinesin; OS – overall survival; MST – median survival time; HR – hazard ratio; CI: confidence interval.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

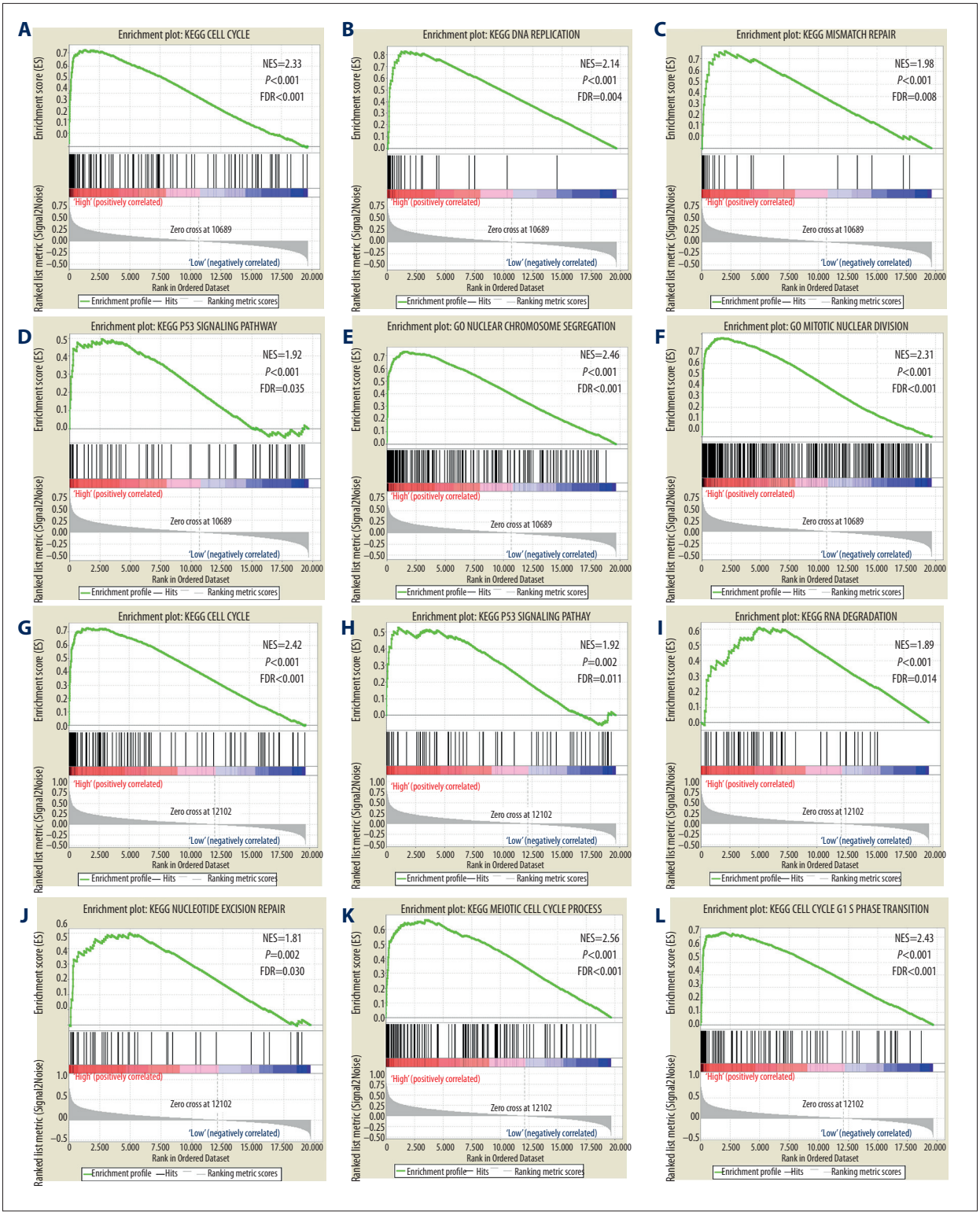


Figure 7. A–F shows GSEA of *KIF2C* in TCGA patients. (**A–D**) GSEA results of c2 reference gene sets for high *KIF2C* expression groups, (**E–F**) GSEA results of c5 reference gene sets for high *KIF2C* expression groups; **G–L** shows GSEA of *KIF4A* in TCGA patients. (**G–J**) GSEA results of c2 reference gene sets for high *KIF4A* expression groups; (**K–L**) GSEA results of c5 reference gene sets for high *KIF4A* expression groups. KIF – kinesin; GSEA – gene set enrichment analysis; TCGA – the Cancer Genome Atlas; BC – breast cancer.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

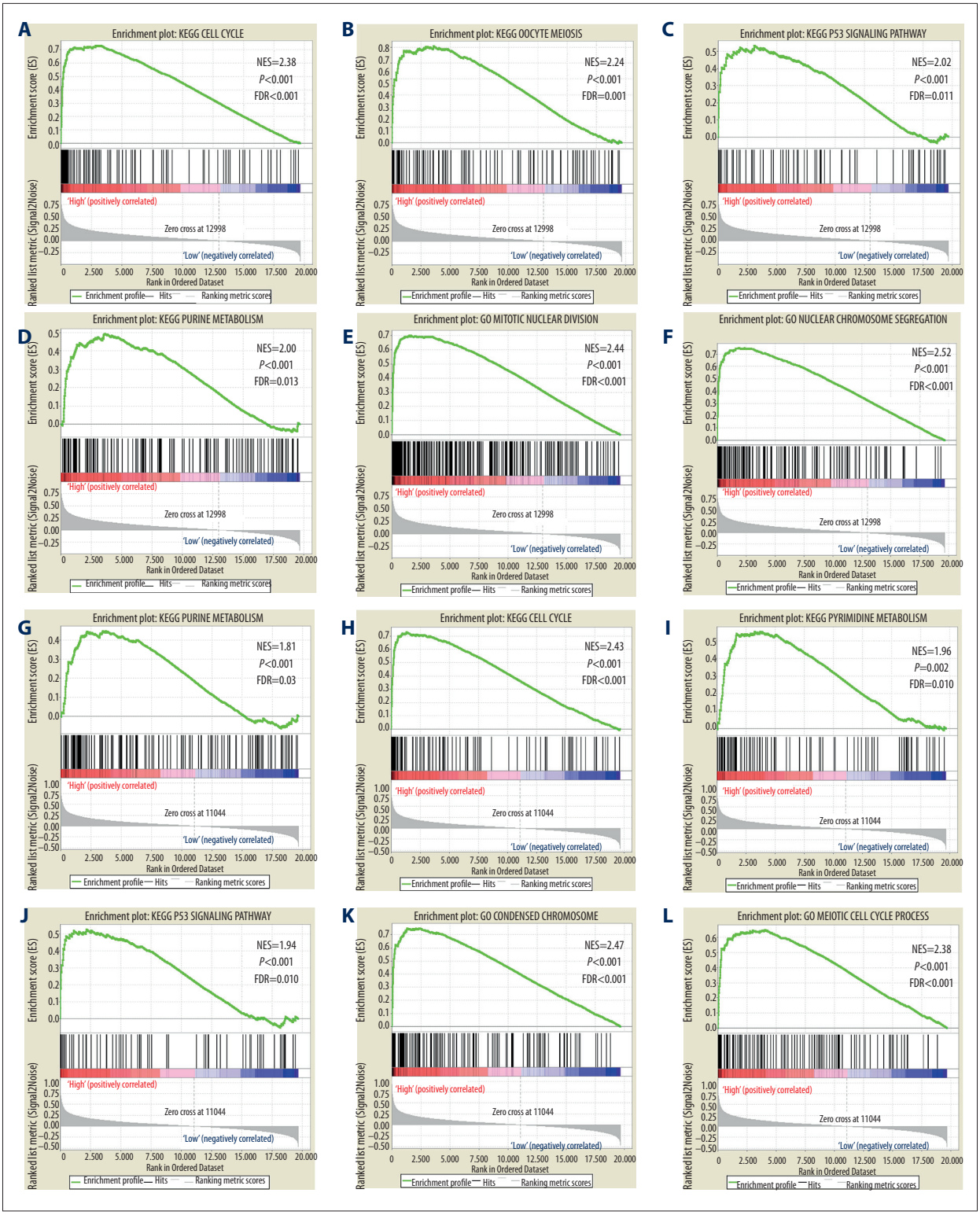


Figure 8. **A–F** shows GSEA of *KIF15* in TCGA patients. (**A–D**) GSEA results of c2 reference gene sets for high *KIF15* expression groups. (**E–F**) GSEA results of c5 reference gene sets for high *KIF15* expression groups. **G–L** shows GSEA of *KIF20A* in TCGA patients. (**G–J**) GSEA results of c2 reference gene sets for high *KIF20A* expression groups. (**K–L**) GSEA results of c5 reference gene sets for high *KIF20A* expression groups. KIF – kinesin; GSEA – gene set enrichment analysis; TCGA – the Cancer Genome Atlas; BC – breast cancer.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

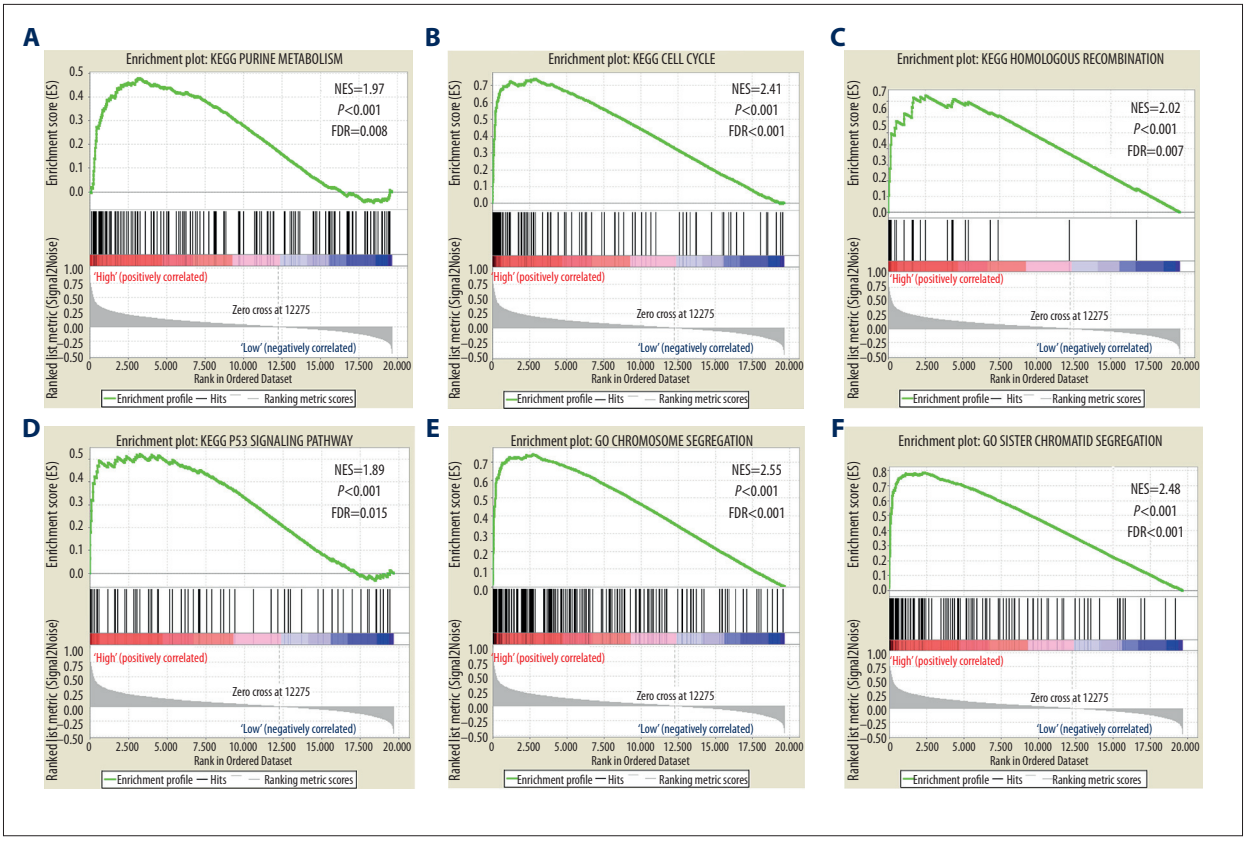


Figure 9. GSEA results of *KIF23* in TCGA BC patients. (**A–D**) GSEA results of c2 reference gene sets for high *KIF23* expression groups. (**E–F**) GSEA results of c5 reference gene sets for high *KIF23* expression groups. KIF – kinesin; GSEA – gene set enrichment analysis; TCGA – the Cancer Genome Atlas; BC – breast cancer.

Shimo et al. have demonstrated that *KIF2C* is overexpressed in BC cells, and plays a major part in cytokinesis within these cells [41]. Additionally, they have discovered that down-regulation of *KIF2C* through treatment with siRNA suppresses development of BC cells [41]. We investigated the high expression of *KIF4A*, *KIF15*, *KIF20A*, *KIF23* and *KIF2C* in patients in the TCGA database, which has been linked to poor OS. Our joint genetic analysis suggests that BC patients with high expression of 2–5 of these genes have poorer OS compared with patients with low gene expression. These findings suggest *KIF4A*, *KIF15*, *KIF20A*, *KIF23* and *KIF2C* as potential prognostic biomarkers and therapeutic targets in BC.

GSEA showed that *KIF4A*, *KIF15*, *KIF20A*, *KIF23* and *KIF2C* were significantly associated with the cell cycle, p53 and mismatch repair, that were associated with their biological functioning. It is well known that KIF genes play critical roles in DNA replication and cell cycle progression [15]. The results require additional experimental validation.

The present study had a few limitations. First, all the information was obtained from open databases, and the medical parameters were not complete. Therefore, we were not able to perform a far-reaching survival analysis of *KIF* genes, considering each latent prognostic variable of BC in the multivariate Cox proportional hazards regression model. Second, because of the varied origin of BC patients, together with the number of elements affecting BC prognosis, we were not able to construct a comprehensive hazard score model, which depended on the KIF genes articulation level for visualization forecast. Third, with the help of the correlation with the past research work, the constraint of our present investigation suggested that it just researched the relationship existing between the mRNA expression of the KIF genes and BC prognosis. Nonetheless, the connection between KIF protein level and BC requires additional investigation.

In spite of the above limitations, we established and validated the prognostic and diagnostic values of expression of KIF genes in BC patients, and similarly examined the potential mechanism linked with *KIF4A*, *KIF15*, *KIF20A*, *KIF23* and *KIF2C* within BC prognosis by GSEA. When these outcomes are confirmed, the prognostic and diagnostic standards of KIF genetics on the extent of protein, such genes might hold a substantial clinical implication value in diagnosis of BC, as well as targeted therapy. Nevertheless, future verification with a larger study

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

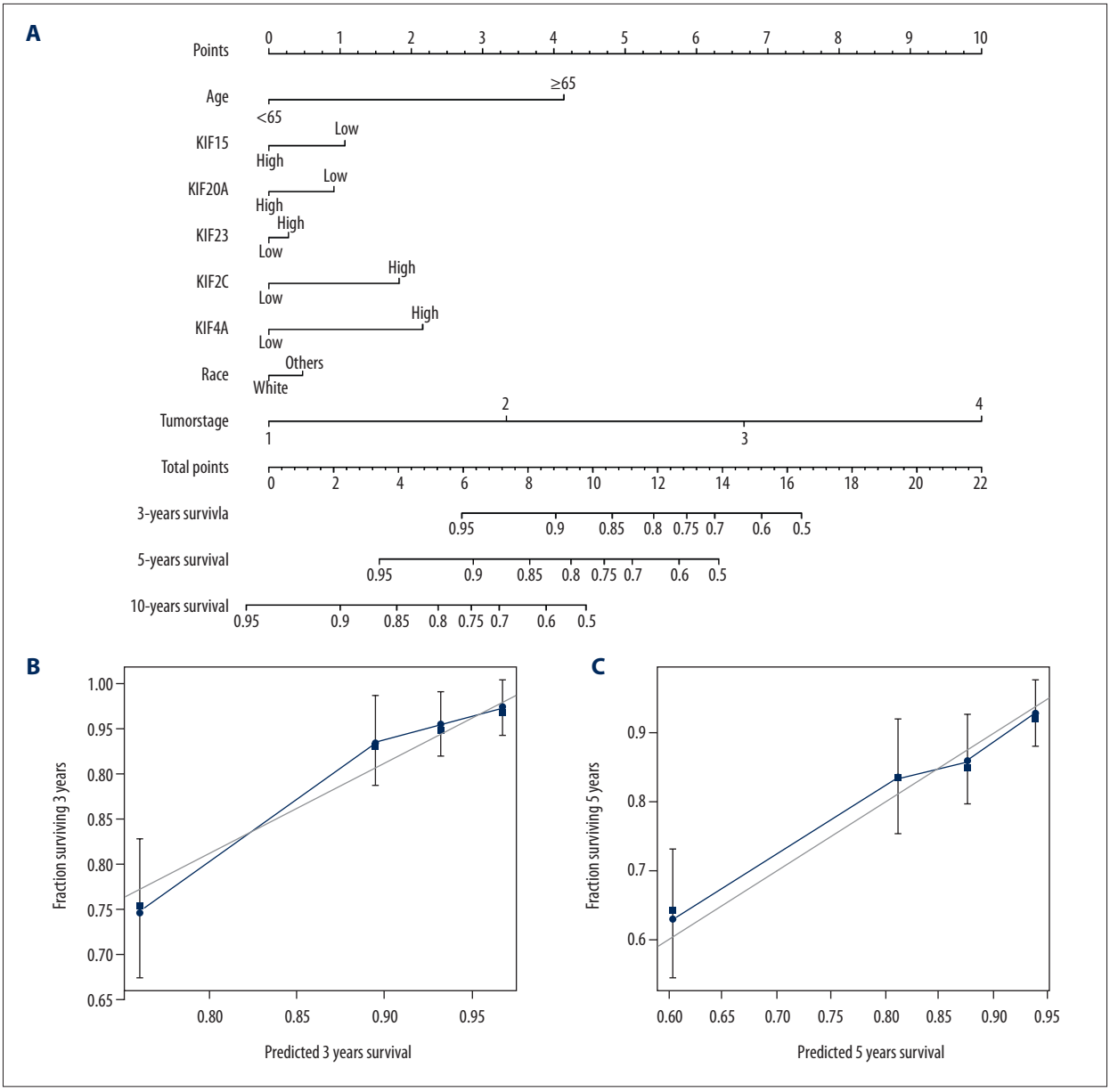


Figure 10. (**A–C**) Relationship between risk score and clinical information. Nomogram for predicting the 1-, 3-, and 5-year event (death) with risk score and clinical information. OS – overall survival.

population is required to confirm that the KIF genes could be involved in diagnosis and prognostic monitoring of BC.

Conclusions

We revealed that 13 KIF genes were differentially expressed in BC tumor tissues, and may serve as latent diagnostic biomarkers in patients with BC. *KIF15*, *KIF20A*, *KIF23*, *KIF2C* and *KIF4A* have the potential to serve as prognostic biomarkers in patients with BC. Multivariate survival analysis, nomograms, and joint survival analysis showed high expression of these genes correlated with poor prognosis of BC. GO, KEGG and GSEA suggested that these genes affect the prognosis of BC by influencing the cell cycle. Our results need to be confirmed in further research.

Acknowledgements

We acknowledge the support of the National Key Clinical Specialty Programs (General Surgery & Oncology), in addition to the Key Laboratory of Early Prevention & Treatment for Regional High-Incidence Tumor (Guangxi Medical University), Ministry of Education, China. We also thank the contributors of

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]

the TCGA database (*https://cancergenome.nih.gov/*) for sharing the BC data in open access.

Conflicts of interest

None.

References:

1. Zhong ZB, Shan M, Wang J et al: Decreased Wnt5a expression is a poor prognostic factor in triple-negative breast cancer. Med Sci Monit, 2016; 22: 1–7
2. Ferlay J, Soerjomataram I, Dikshit R et al: Cancer incidence and mortality worldwide: Sources, methods and major patterns in GLOBOCAN 2012. Int J Cancer, 2015; 136(5): E359–86
3. Torre LA, Bray F, Siegel RL et al: Global cancer statistics, 2012. Cancer J Clin, 2015; 65(2): 87–108
4. Guo XH, Li JT, Zhang HW et al: Relationship between ADAMTS8, ADAMTS18, and ADAMTS20 (a disintegrin and metalloproteinase with thrombospondin motifs) expressions and tumor molecular classification, clinical pathological parameters, and prognosis in breast invasive ductal carcinoma. Med Sci Monit, 2018; 24: 3726–35
5. Harbeck N, Gnant M: Breast cancer. Lancet, 2017; 389(10074): 1134–50
6. Ginsburg O, Bray F, Coleman MP et al: The global burden of women's cancers: A grand challenge in global health. Lancet, 2017; 389(10071): 847–60
7. Yu M, Zhang X, Li H et al: MicroRNA-588 is downregulated and may have prognostic and functional roles in human breast cancer. Med Sci Monit, 2017; 23: 5690–96
8. Wu L, Shi W, Long J et al: A transcriptome-wide association study of 229,000 women identifies new candidate susceptibility genes for breast cancer. Nat Genet, 2018; 50(7): 968–78
9. Nathanson KN, Wooster R, Weber BL: Breast cancer genetics: What we know and what we need. Nat Med, 2001; 7(5): 552–56
10. Yi H, Wang K, Jin H et al: Overexpression of Rho-associated coiled-coil containing protein kinase 2 is correlated with clinical progression and poor prognosis in breast cancer. Med Sci Monit, 2018; 24: 4776–81
11. Zhang N, Zuo L, Zheng H et al: Increased expression of CD81 in breast cancer tissue is associated with reduced patient prognosis and increased cell migration and proliferation in MDA-MB-231 and MDA-MB-435S human breast cancer cell lines *in vitro*. Med Sci Monit, 2018; 24: 5739–47
12. Kwan TT, Bardia A, Spring LM et al: A digital RNA signature of circulating tumor cells predicting early therapeutic response in localized and metastatic breast cancer. Cancer Discov, 2018; 8L 1286–99
13. Rodriguez Bautista R, Ortega Gomez A, Hidalgo Miranda A et al: Long noncoding RNAs: implications in targeted diagnoses, prognosis, and improved therapeutic strategies in human non- and triple-negative breast cancer. Clin Epigenetics, 2018; 10: 88
14. Liao XW, Zhu GZ, Huang R et al: Identification of potential prognostic microRNA biomarkers for predicting survival in patients with hepatocellular carcinoma. Cancer Manag Res, 2018; 10: 787–803
15. Lucanus AJ, Yip GW: Kinesin superfamily: Roles in breast cancer, patient prognosis and therapeutics. Oncogene, 2018; 37(7): 833–38
16. Goldstein LS, Philp AV: The road less traveled: Emerging principles of kinesin motor utilization. Ann Rev Cell Dev Biol, 1999; 15: 141–83
17. Miki H, Setou M, Hirokawa N: All kinesin superfamily protein, KIF, genes in the mouse and human genome and transcripts. Mol Biol Cell, 2002; 13: 184a
18. Liu XR, Gong H, Huang K: Oncogenic role of kinesin proteins and targeting kinesin therapy. Cancer Sci, 2013; 104(6): 651–56
19. Gao HW, Chen XR, Cai QL et al: Increased KIF4A expression is a potential prognostic factor in prostate cancer. Oncol Lett, 2018; 15(5): 7941–47
20. Eskova A, Knapp B, Matelska D et al: An RNAi screen identifies KIF15 as a novel regulator of the endocytic trafficking of integrin. J Cell Sci, 2014; 127(11): 2433–47
21. Groth-Pedersen L, Aits S, Corcelle-Termeau E et al: Identification of cytoskeleton-associated proteins essential for lysosomal stability and survival of human cancer cells. PLoS One, 2012; 7(10): e45381
22. Warde-Farley D, Donaldson SL, Comes O et al: The GeneMANIA prediction server: Biological network integration for gene prioritization and predicting gene function. Nucleic Acids Res, 2010; 38: W214–20
23. von Mering C, Huynen M, Jaeggi D et al: STRING: A database of predicted functional associations between proteins. Nucleic Acids Res, 2003; 31(1): 258–61
24. Maere S, Heymans K, Kuiper M: BiNGO: A Cytoscape plugin to assess overrepresentation of Gene Ontology categories in Biological Networks. Bioinformatics, 2005; 21(16): 3448–49
25. Subramanian A, Tamayo P, Mootha VK et al: Gene set enrichment analysis: A knowledge-based approach for interpreting genome-wide expression profiles. Proc Natl Acad Sci USA, 2005; 102(43): 15545–50
26. Mootha VK, Lindgren CM, Eriksson KF et al: PGC-1 alpha-responsive genes involved in oxidative phosphorylation are coordinately downregulated in human diabetes. Nat Genet, 2003; 34(3): 267–73
27. Liberzon A, Birger C, Thorvaldsdottir H et al: The Molecular Signatures Database (MSigDB) hallmark gene set collection. Cell Syst, 2015; 1(6): 417–25
28. Wordeman L: How kinesin motor proteins drive mitotic spindle function: Lessons from molecular assays. Semin Cell Dev Biol, 2010; 21(3): 260–68
29. Bidkhori G, Narimani Z, Hosseini Ashtiani S et al: Reconstruction of an integrated genome-scale co-expression network reveals key modules involved in lung adenocarcinoma. PLoS One, 2013; 8(7): e67552
30. Wang J, Guo XJ, Xie CC, Jiang JX: KIF15 promotes pancreatic cancer proliferation via the MEK-ERK signalling pathway. Br J Cancer, 2017; 117(2): 245–55
31. Zou JX, Duan ZJ, Wang JJ et al: Kinesin family deregulation coordinated by bromodomain protein ANCCA and histone methyltransferase MLL for breast cancer cell growth, survival, and tamoxifen resistance. Mol Cancer Res, 2014; 12(4): 539–49
32. Taniuchi K, Nakagawa H, Nakamura T et al: Down-regulation of RAB6KIFL/KIF20A, a kinesin involved with membrane trafficking of discs large homologue 5, can attenuate growth of pancreatic cancer cell. Cancer Res, 2005; 65(1): 105–12
33. Zhao X, Zhou LL, Li X et al: Overexpression of KIF20A confers malignant phenotype of lung adenocarcinoma by promoting cell proliferation and inhibiting apoptosis. Cancer Med, 2018; 7(9): 4678–89
34. Sun XT, Jin ZT, Song X et al: Evaluation of KIF23 variant 1 expression and relevance as a novel prognostic factor in patients with hepatocellular carcinoma. BMC Cancer, 2015; 15: 961
35. Kato T, Wada H, Patel P et al: Overexpression of KIF23 predicts clinical outcome in primary lung cancer patients. Lung Cancer, 2016; 92: 53–61
36. Taniwaki M, Takano A, Ishikawa N et al: Activation of KIF4A as a prognostic biomarker and therapeutic target for lung cancer. Clin Cancer Res, 2007; 13(22): 6624–31
37. Wang H, Lu CQ, Li Q et al: The role of Kif4A in doxorubicin-induced apoptosis in breast cancer cells. Mol Cells, 2014; 37(11): 812–18
38. Narayan G, Bourdon V, Chaganti S et al: Gene dosage alterations revealed by cDNA microarray analysis in cervical cancer: Identification of candidate amplified and overexpressed genes. Gene Chromosome Canc, 2007; 46(4): 373–84
39. Hou GJ, Dong CP, Dong ZH et al: Upregulate KIF4A enhances proliferation, invasion of hepatocellular carcinoma and indicates poor prognosis across human cancer types. Sci Rep, 2017; 7(1): 4148
40. Minakawa Y, Kasamatsu A, Koike H et al: Kinesin family member 4A: A potential predictor for progression of human oral cancer. PLoS One, 2013; 8(12): e85951
41. Shimo A, Tanikawa C, Nishidate T et al: Involvement of kinesin family member 2C/mitotic centromere-associated kinesin overexpression in mammary carcinogenesis. Cancer Sci, 2008; 99(1): 62–70

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS]