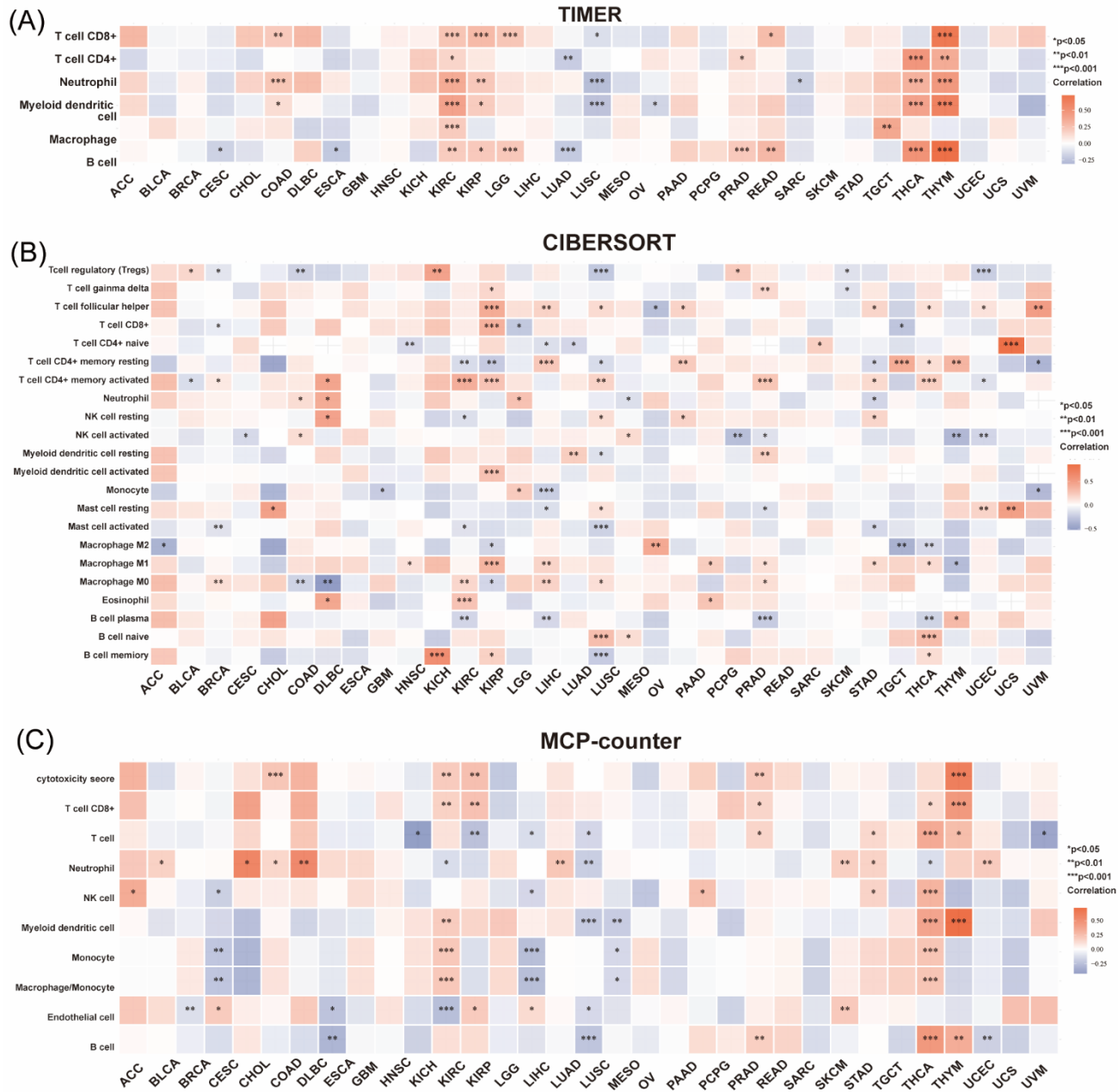
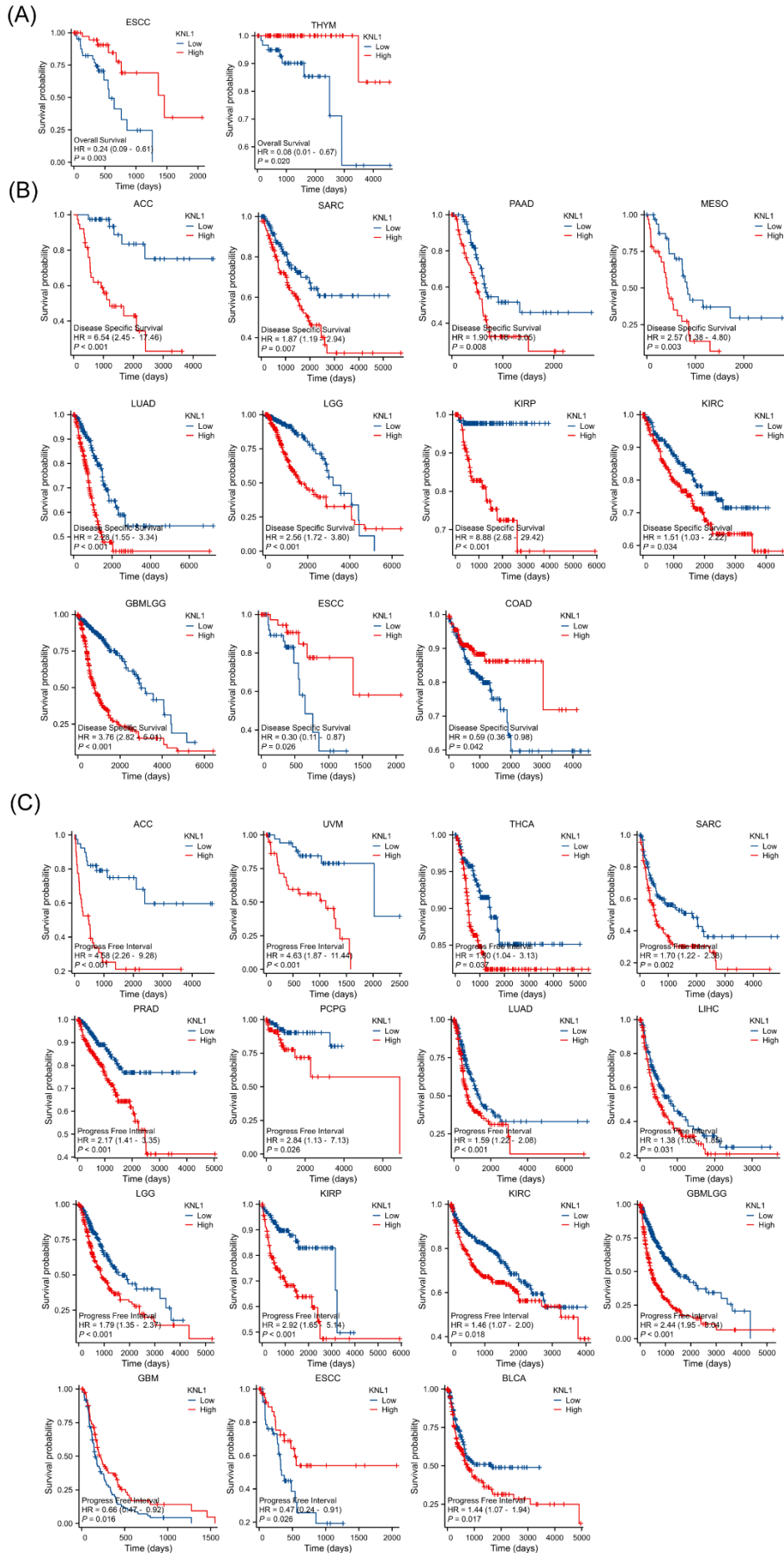


Supp Fig 1. The protein expression of KNL1 was verified using the CPTAC database.

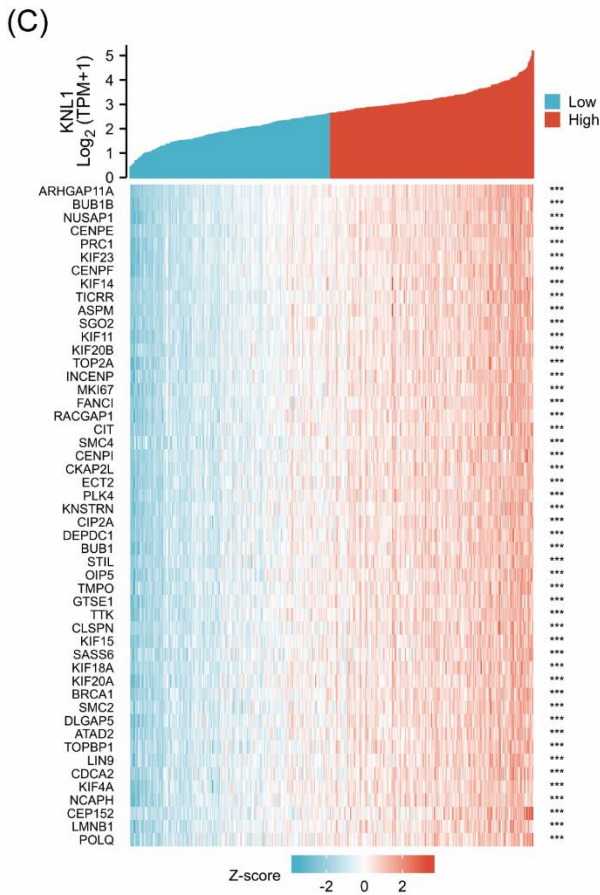
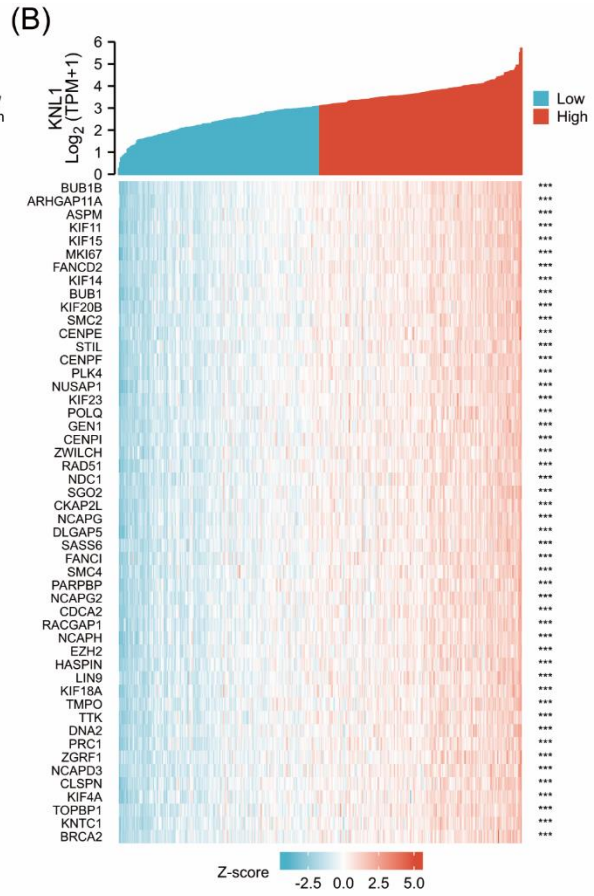
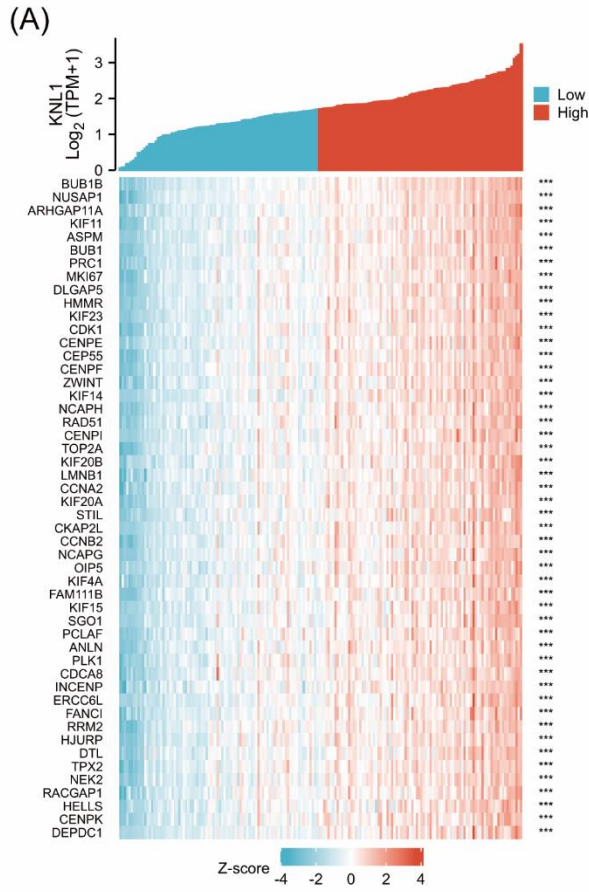


Supp Fig 2. The immune score of immune cells were calculated with TIMER, CIBERSORT, and MCP-counter algorithms. ns, $p \geq 0.05$; * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

Supp Fig 3. The association between KNL1 expression and tumor grade (A), stage (B) were explored in 30 tumors from TISIDB database. Higher KNL1 expression was positively related with higher tumor stage and grade in 6 and 7 cancers, respectively. (C) ROC curves for the other 17 cancers, indicated that KNL1 had a great prognostic predictive ability.

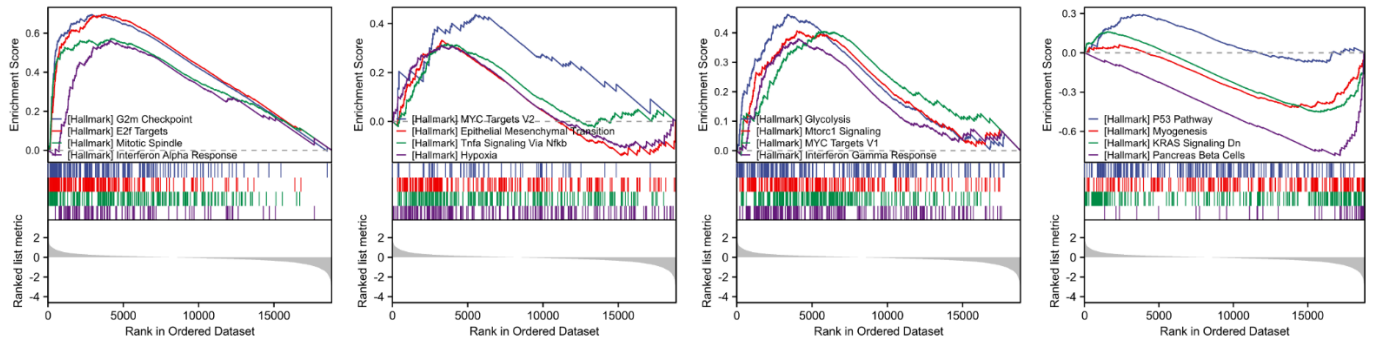


Supp Fig 4. (A) The KNL1-high group exhibited better OS in ESCC and THYM. (B) K-M survival analysis showed that DSS was significantly associated with KNL1 expression levels in 11 cancers. The KNL1-high group exhibited better DSS in ESCC and COAD. (C) PFI was significantly associated with KNL1 expression levels in 15 cancers. The KNL1-high group exhibited better DSS in ESCC and GBM.

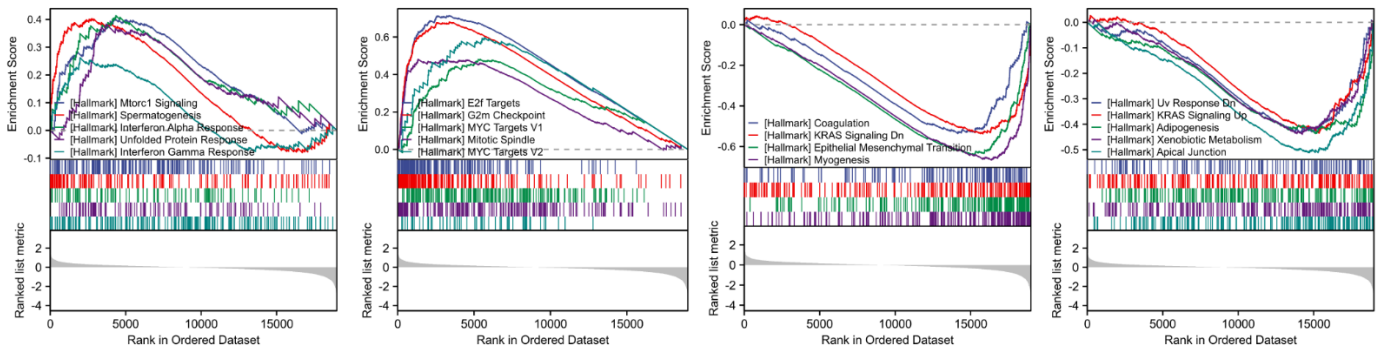


Supp Fig 5. The heatmap of the top 50 most related genes of KNL1 in PAAD (A), STAD (B), and BCLA (C).

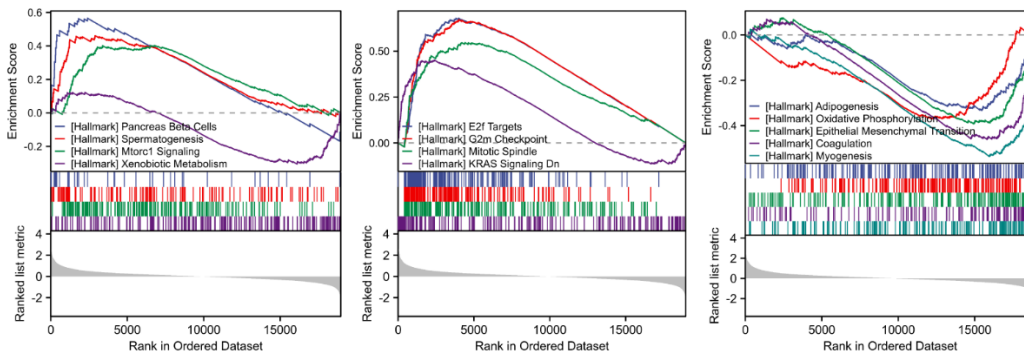
(A)



(B)

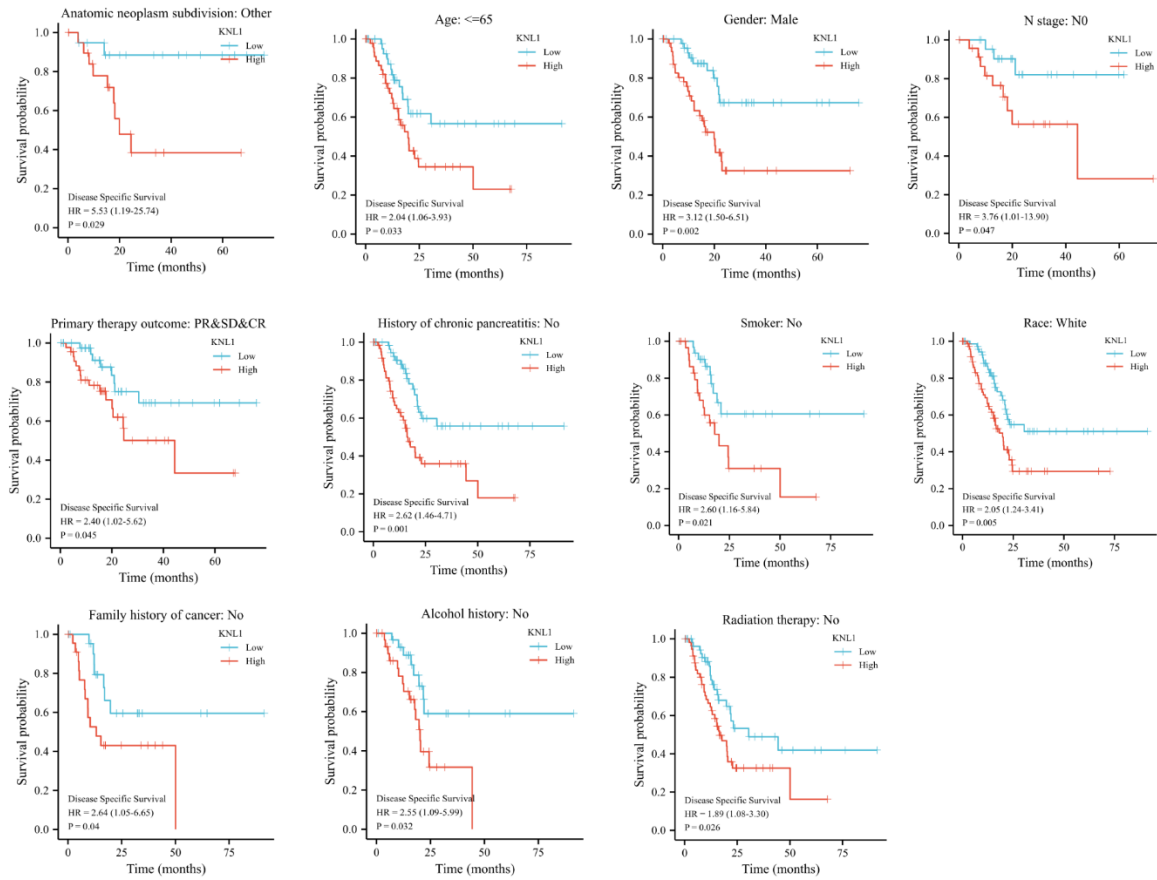


(C)

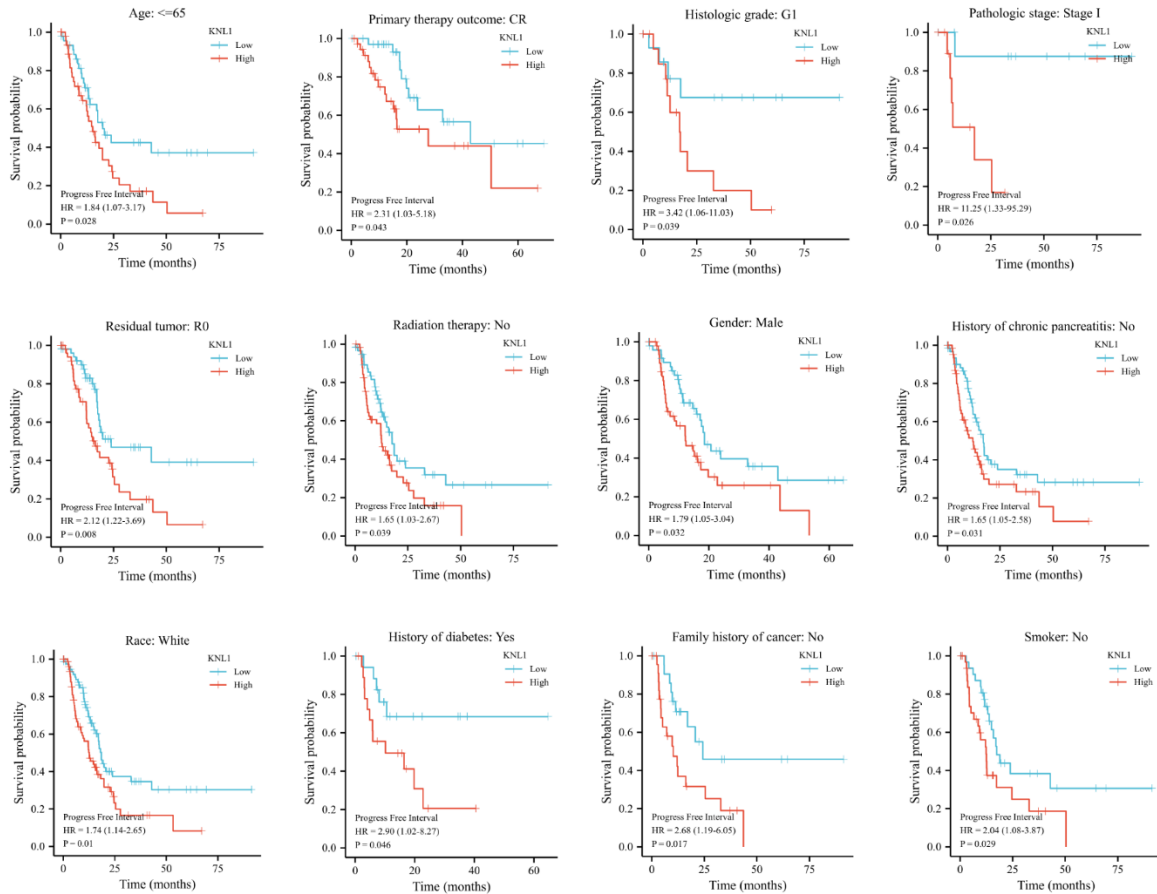


Supp Fig 6. GSEA analysis in PAAD (A), STAD (B), and BLCA (C). G2m checkpoint, E2f targets, mitotic spindle, epithelial mesenchymal transition, myogenesis and KRAS signaling pathways were significantly enriched in KNL1-related DEGs.

(A)



(B)



Supp Fig 7. Survival analysis of different clinicopathological subgroups in PAAD indicated that high expression levels of KNL1 were associated with poor DSS (A) and PFI (B) in clinical subgroups

Supplementary table 1. The amplification's expression level of KNL1 of qPCR in SV-HUC-1, 5637, T24, UMUC3 and J82 cell lines after being processed with the $\Delta\Delta\text{CT}$ technique and adjusted against GAPDH mRNA.

Group	Sample	GAPDH	KNL1
SV-HUC-1	sample1	16.47	23.86
SV-HUC-1	sample2	16.35	23.82
SV-HUC-1	sample3	16.29	23.79
5637	sample1	20.5	25.52
5637	sample2	20.45	25.59
5637	sample3	20.3	25.63
T24	sample1	16.4	22.46
T24	sample2	16.36	22.57
T24	sample3	16.24	22.52
UMUC3	sample1	17.43	24.1
UMUC3	sample2	17.32	23.89
UMUC3	sample3	17.18	24.03
J82	sample1	18.2	24.75
J82	sample2	18.15	24.71
J82	sample3	18.09	24.67

Supplementary table 2. H-score obtained after IHC staining of cancer and paracancerous tissue sections from 8 case-confirmed PAAD patients

ID Index	215938	215938	215637	215637	215384	215384	214163	214163	213735	213735	200947	200947	192287	192287	183039	183039
	9	9	2	2	0	0	9	9	0	0	7	7	1	1	2	2
Site	P	T	P	T	P	T	T	P	T	P	P	T	T	P	T	P
No. Total non-background pixels	745457	943243	804381	710603	111843	389143	859873	166149	750853	962636	108848	779277	155121	475452	237818	535585
No. Background pixels	455402	803456	281361	949417	911221	987871	109570	370686	102038	193298	570612	545174	351561	663013	265461	121997
No. Negative pixels	57508.00	19782.00	151109.00	268600.00	248178.00	48672.00	42648.00	80222.00	55572.00	682323.00	235191.00	67145.00	176944.00	80250.00	139110.00	761011.00
No. Weak-Positive pixels	530563	602185	396098	256019	670842	137613	504394	114315	490605	729047	738949	521118	735695	247432	883023	235733
	1.00	7.00	7.00	9.00	1.00	7.00	3.00	87.00	8.00	0.00	7.00	7.00	7.00	9.00	.00	3.00

No. Moderate-Positive pixels	203579	332902	376860	326329	403853	201438	323010	508110	246408	159401	312985	247769	788308	201805	107269	212185
No. Strong-Positive pixels	4.00	9.00	3.00	3.00	3.00	0.00	3.00	8.00	2.00	8.00	6.00	0.00	6.00	7.00	4.00	3.00
Mean Density of all pixels	12.97	13.99	16.07	19.95	14.58	19.83	14.98	13.43	13.74	12.25	13.78	13.46	16.17	15.88	19.22	15.60
Mean Density of Negative pixels	14.69	16.27	17.35	13.80	17.43	9.51	8.80	12.37	10.42	18.18	17.15	11.68	17.06	12.07	16.29	14.02
Mean Density of Weak-Positive pixels	10.29	10.78	11.28	10.76	10.86	10.63	10.44	11.20	10.57	10.14	10.98	10.70	12.00	10.67	10.75	11.00
Mean Density	20.12	20.06	20.80	22.82	20.26	23.14	21.21	18.83	20.14	19.56	19.98	19.63	20.21	21.42	22.48	20.81

**of
Modera
te-
Positive
pixels
Mean
Density
of
Strong-
Positive
pixels
Ratio of
Negativ
e pixels
to total
pixels
Ratio of
Strong-
Positive
pixels
to total
pixels
Ratio of
Modera
te-
Positive**

40.35	38.74	40.30	44.30	43.05	43.48	41.97	39.54	40.78	41.47	39.90	39.97	38.02	41.43	43.77	41.58
1.00	0.00	2.00	4.00	2.00	1.00	0.00	0.00	1.00	7.00	2.00	1.00	1.00	2.00	6.00	14.00
1.00	1.00	2.00	14.00	2.00	12.00	3.00	0.00	1.00	1.00	1.00	0.00	1.00	4.00	12.00	2.00
27.00	35.00	47.00	46.00	36.00	52.00	38.00	31.00	33.00	17.00	29.00	32.00	51.00	42.00	45.00	40.00

pixels to total pixels Ratio of Weak- Positive pixels to total pixels Ratio of all positive pixels to total pixels H- Score	71.00	64.00	49.00	36.00	60.00	35.00	59.00	69.00	65.00	76.00	68.00	67.00	47.00	52.00	37.00	44.00
	99.23	99.79	98.12	96.22	97.78	98.75	99.50	99.52	99.26	92.91	97.84	99.14	98.86	98.31	94.15	85.79
	128.03	136.39	149.03	170.68	137.27	173.76	143.63	130.36	134.28	110.71	128.99	131.88	150.91	148.41	163.09	129.73

P: paracancerous tissue; T: cancer tissue