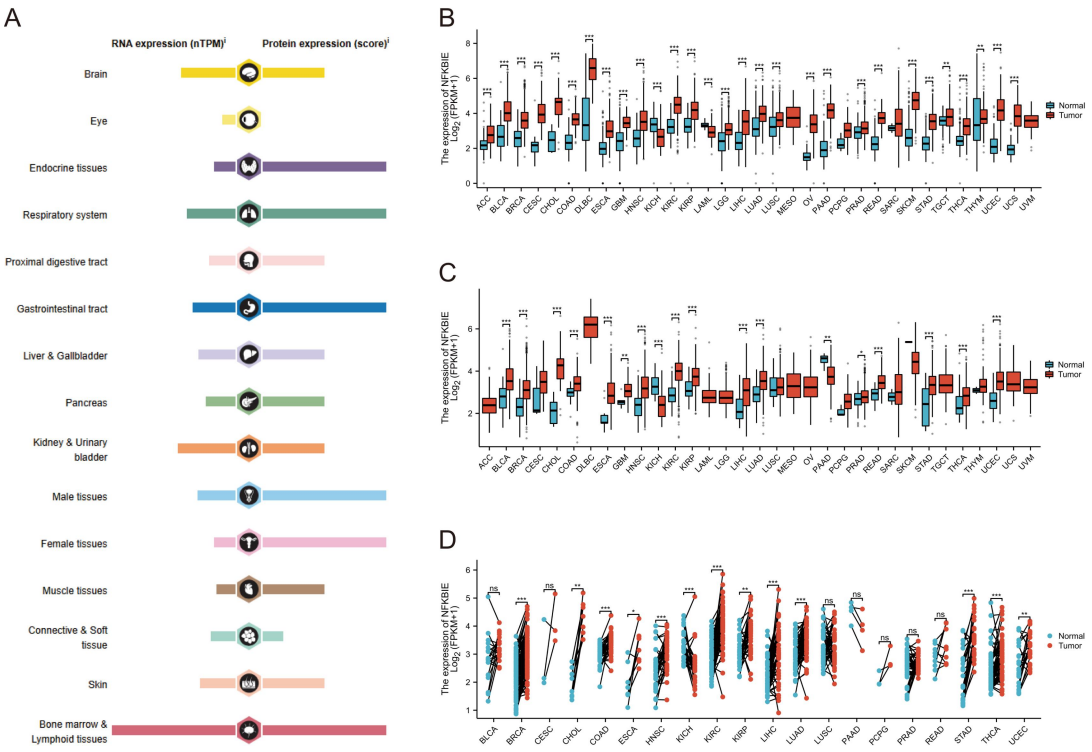
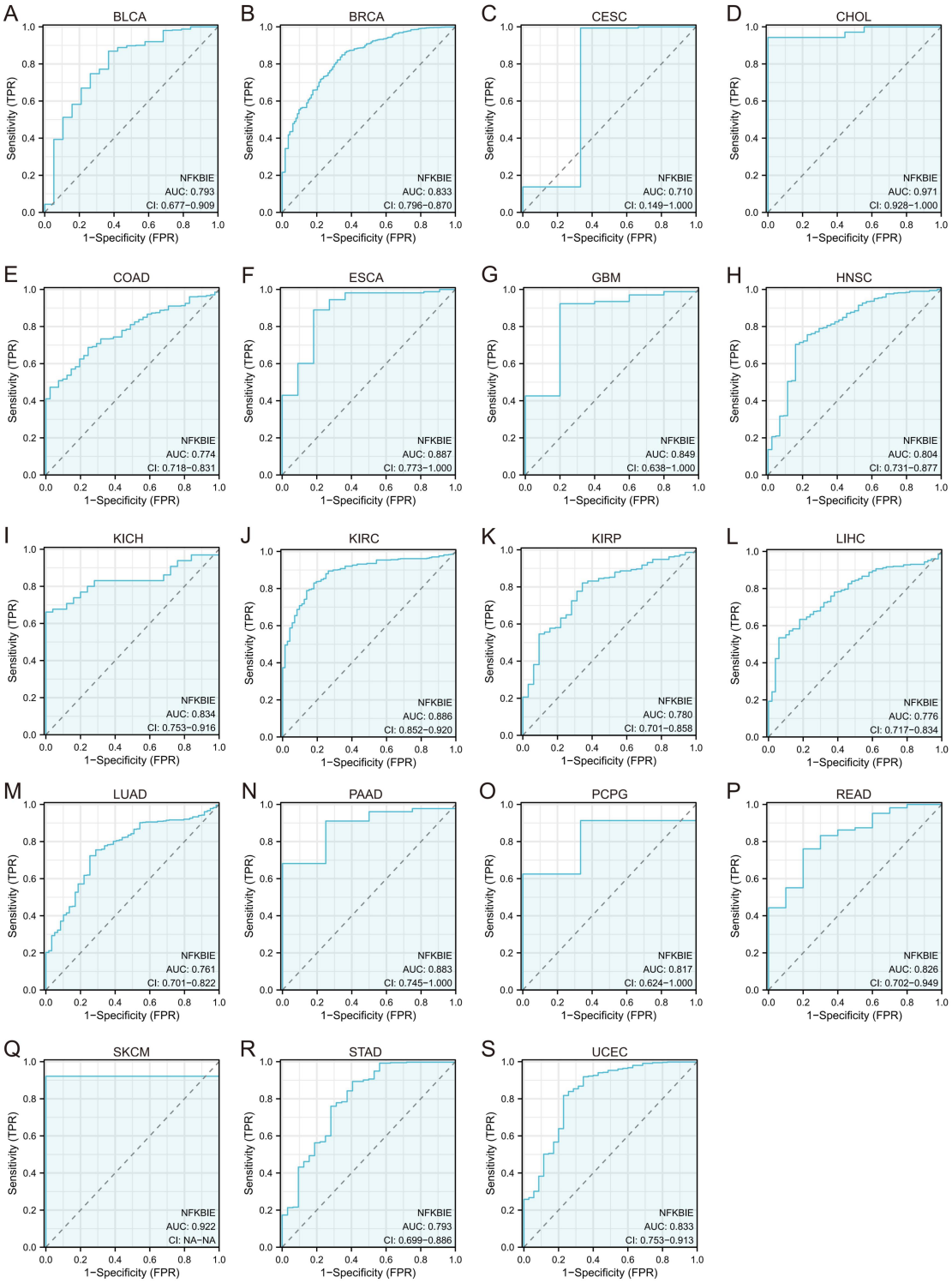
Supplementary Figure 1



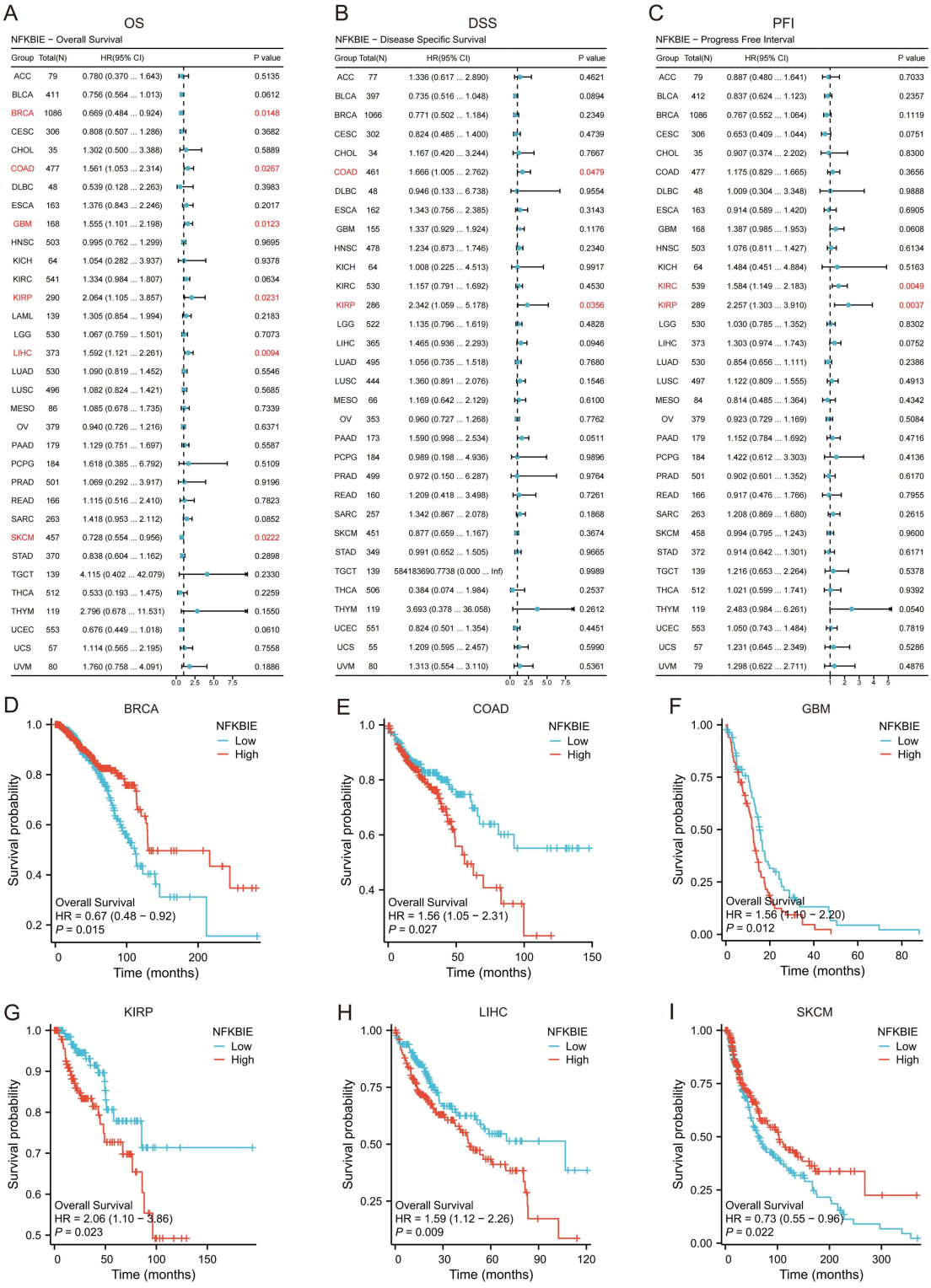
Supplementary Figure 1: Expression analysis of NFKBIE in human tissues and pan-cancer tissues. **(A)** Summary of NFKBIE mRNA and protein expressions in human organs and tissues. **(B)** Expression of NFKBIE between 33 types of cancers and normal tissues in unpaired sample analysis. **(C)** Expression of NFKBIE between 33 types of cancers and adjacent tissues in unpaired sample analysis; **(D)** Paired sample analysis of NFKBIE expression between 20 types of cancers and adjacent tissues. **P*<0.05, ***P*<0.01, ****P*<0.001, NS: *P*≥0.05, *P*<0.05 is statistically significant.

Supplementary Figure 2



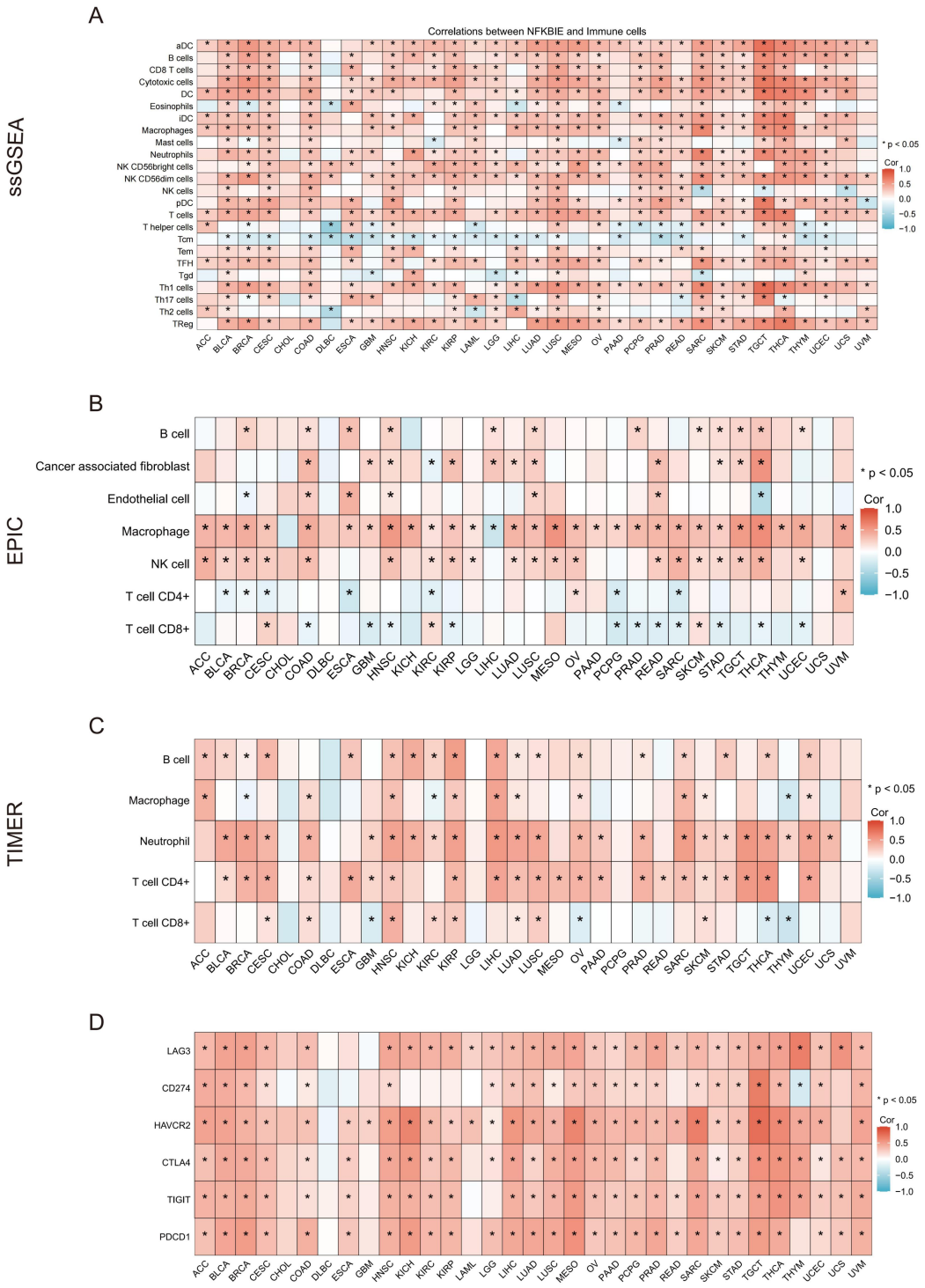
Supplementary Figure 2: ROC curves of NFKBIE in 19 types of cancers. Cancers with AUC>0.7: **(A)** BLCA, **(B)** BRCA, **(C)** CESC, **(D)** CHOL, **(E)** COAD, **(F)** ESCA, **(G)** GBM, **(H)** HNSC, **(I)** KICH, **(J)** KIRC, **(K)** KIRP, **(L)** LIHC, **(M)** LUAD, **(N)** PAAD, **(O)** PCPG, **(P)** READ, **(Q)** SKCM, **(R)** STAD, **(S)** UCEC.

Supplementary Figure 3



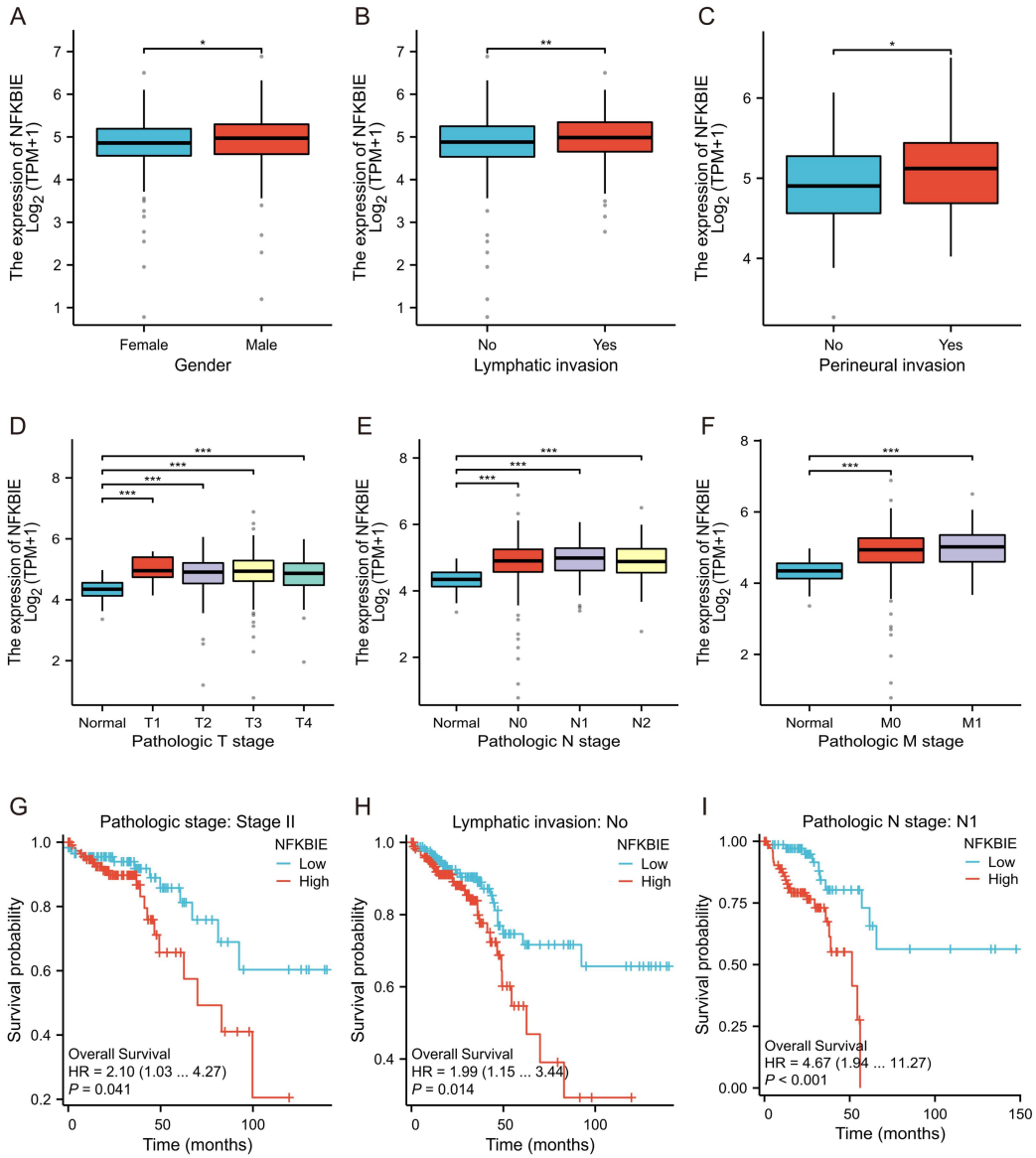
Supplementary Figure 3: Prognostic analysis of NFKBIE in pan-cancer. **(A-C)** Forest plots of the correlations between NFKBIE expression and OS, DSS, and PFI in pan-cancer. **(D-I)** Kaplan-Meier survival analysis results showed that patients with higher NFKBIE expression levels in COAD, GBM, KIRP, and LIHC had poorer OS, while the opposite results were observed in BPCA and SKCM patients. $P<0.05$ is statistically significant.

Supplementary Figure 4

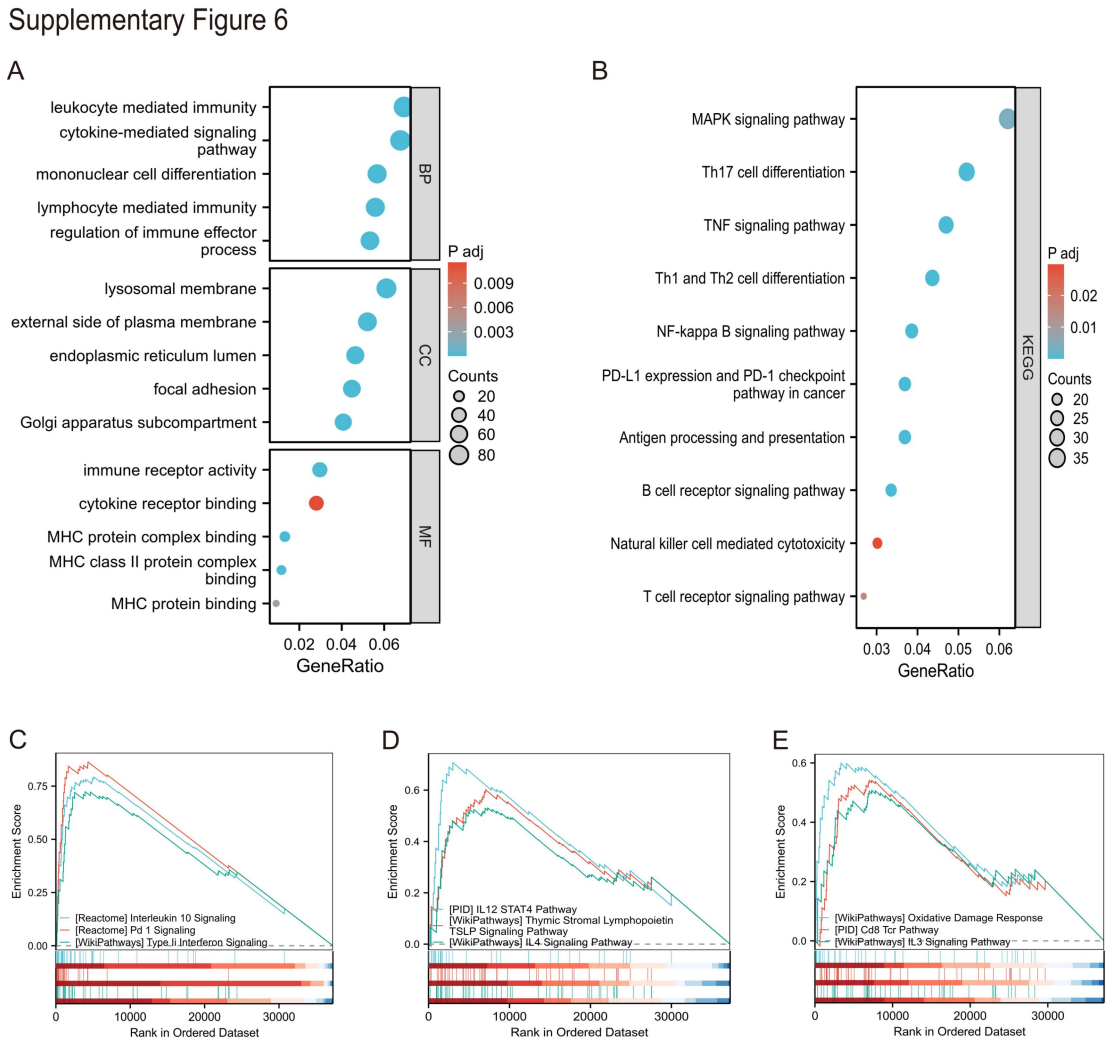


Supplementary Figure 4: Correlation of NFKBIE expression with immune infiltrating cells and immune checkpoints in pan-cancer: **(A)** Immune infiltrating cells under the ssGSEA algorithm. **(B)** Immune infiltrating cells under the EPIC algorithm. **(C)** Immune infiltrating cells under the TIMER algorithm. **(D)** Correlation of NFKBIE expression with 6 common immune checkpoints in the TIMER2.0 database. **P*<0.05, ***P*<0.01, *P*<0.05 is statistically significant.

Supplementary Figure 5

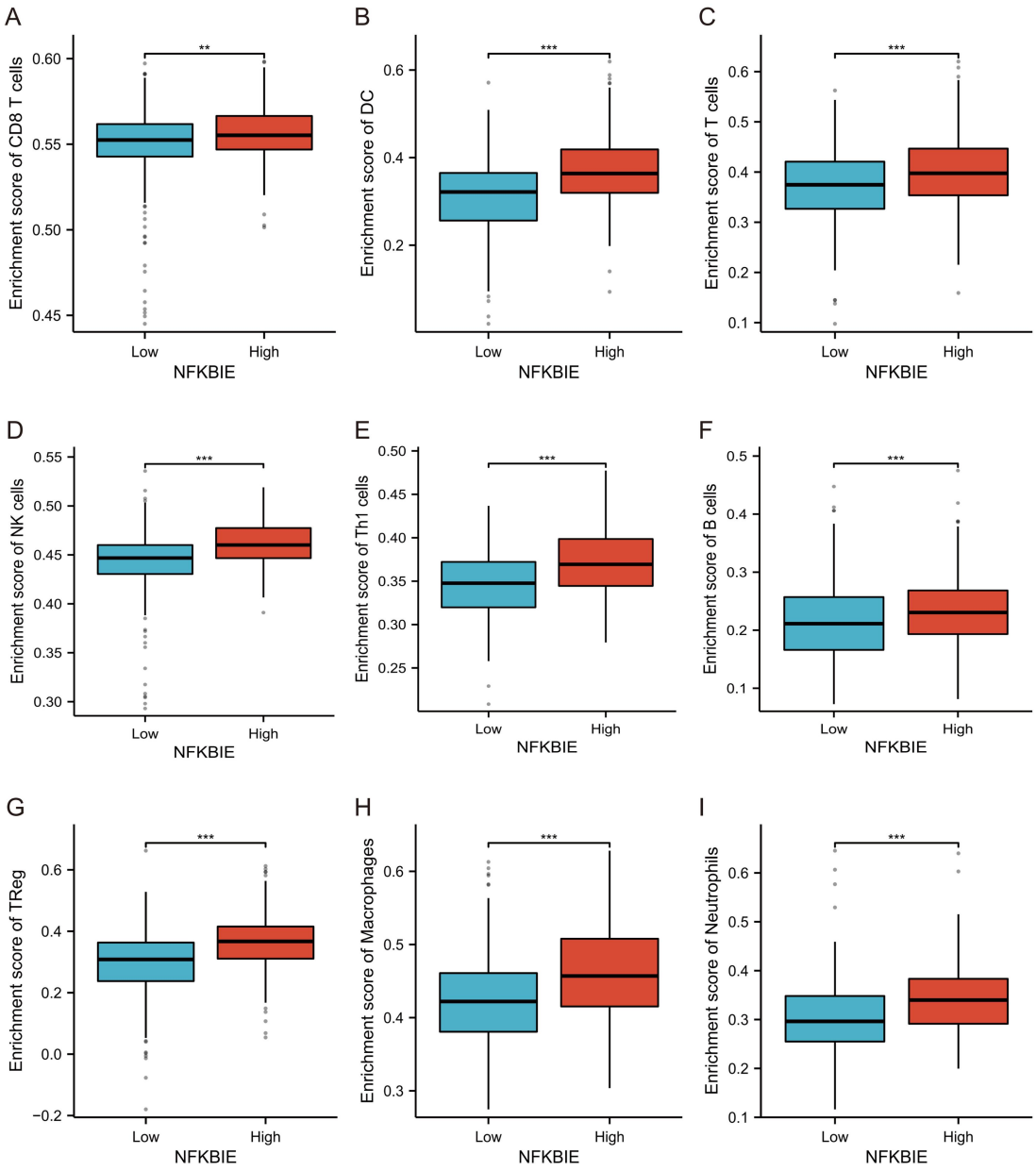


Supplementary Figure 5: Relationship between NFKBIE expression and clinicopathological parameters. (**A-C**) Relationship between NFKBIE expression and gender, lymphovascular invasion, and perineural invasion. (**D-F**) Relationship between NFKBIE expression and T, N, and M stages. (**G-I**) The overall survival of CRC subgroups with higher NFKBIE expression is poorer, (**G**) Pathological stage II group; (**H**) Lymphovascular invasion N0 subgroup; (**I**) N stage N1 subgroup. **P*<0.05, ***P*<0.01, ****P*<0.001, *P*<0.05 is statistically significant.



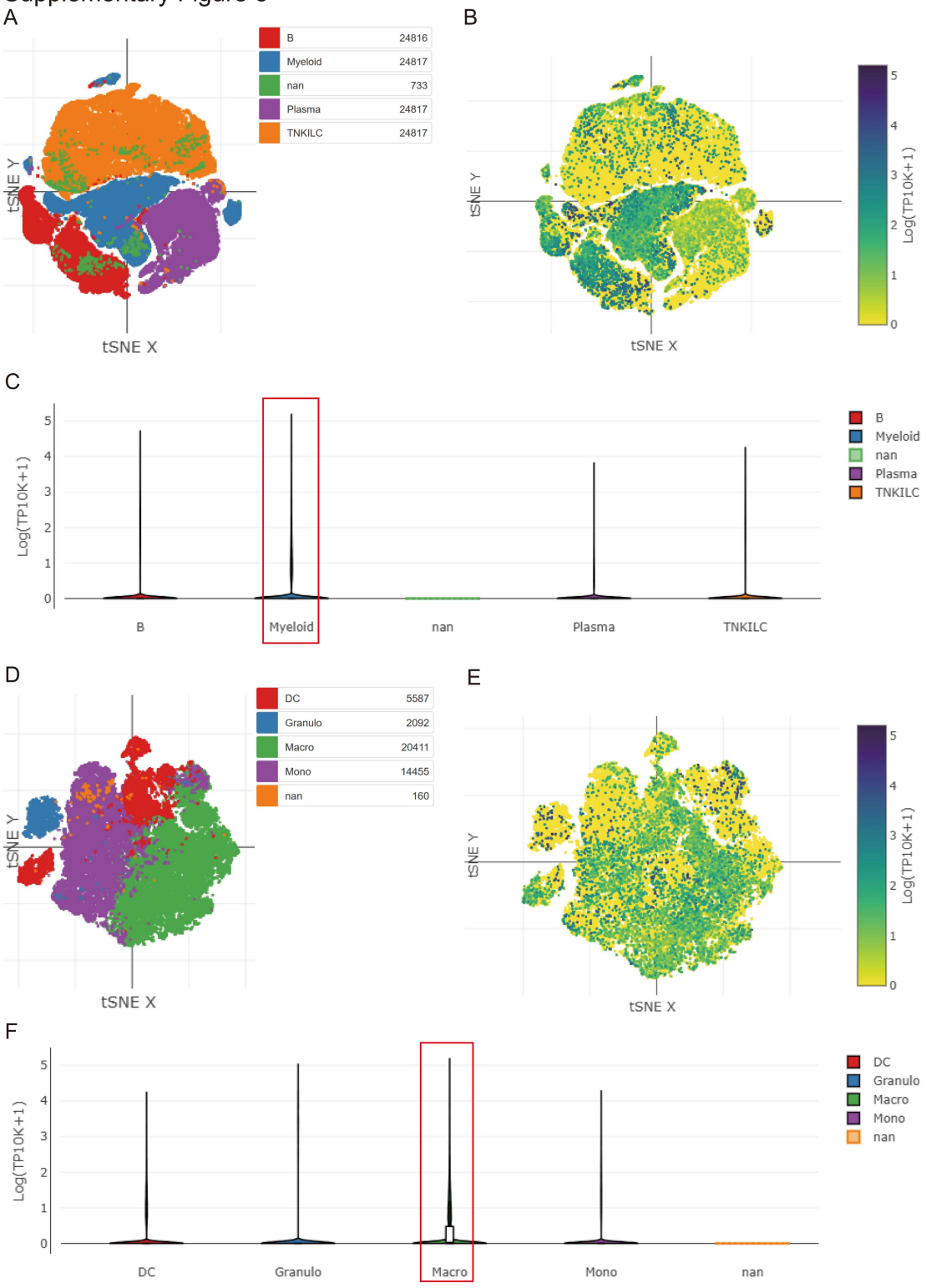
Supplementary Figure 6: Functional enrichment analysis of NFKBIE. **(A)** GO enrichment analysis results of NFKBIE. **(B)** KEGG enrichment analysis results of NFKBIE. **(C-E)** GSEA enrichment analysis results of NFKBIE.

Supplementary Figure 7



Supplementary Figure 7: Relationship between high and low expression groups of NFKBIE and immune cells in CRC. Compared with the low expression group of NFKBIE, the concentrations of (**A**) CD8 + T cells, (**B**) DC cells, (**C**) T cells, (**D**) NK cells, (**E**) Th1 cells, (**F**) B cells, (**G**) Treg cells, (**H**) Macrophages, and (I) Neutrophils were higher in the high expression group of NFKBIE. ***P*<0.01, ****P*<0.001, *P*<0.05 is statistically significant.

Supplementary Figure 8



Supplementary Figure 8: Single-cell analysis of NFKBIE in CRC. **(A)** Distribution of immune cells in CRC. **(B)** Distribution of NFKBIE in immune cells of CRC. **(C)** Expression of NFKBIE in immune cells of CRC. **(D)** Distribution of myeloid cells in CRC. **(E)** Distribution of NFKBIE in myeloid cells of CRC. **(F)** Expression of NFKBIE in myeloid cells of CRC.

Supplementary Table1. Correlation between NFKBIE expression and clinicopathological features (chi-square test)

n	**322**	**322**	
Pathologic T stage, n (%)			0.890
T1	9 (1.4%)	11 (1.7%)	
T2	57 (8.9%)	54 (8.4%)	
T3	214 (33.4%)	222 (34.6%)	
T4	39 (6.1%)	35 (5.5%)	
Pathologic N stage, n (%)			0.217
N0	192 (30%)	176 (27.5%)	
N1	67 (10.5%)	86 (13.4%)	
N2	60 (9.4%)	59 (9.2%)	
Pathologic M stage, n (%)			0.195
M0	233 (41.3%)	242 (42.9%)	
M1	37 (6.6%)	52 (9.2%)	
Pathologic stage, n (%)			0.428
Stage I	55 (8.8%)	56 (9%)	
Stage II	125 (20.1%)	113 (18.1%)	
Stage III	91 (14.6%)	93 (14.9%)	
Stage IV	38 (6.1%)	52 (8.3%)	
Gender, n (%)			**0.033**
Female	164 (25.5%)	137 (21.3%)	
Male	158 (24.5%)	185 (28.7%)	
Age, n (%)			**0.004**
<= 65	120 (18.6%)	156 (24.2%)	
> 65	202 (31.4%)	166 (25.8%)	
CEA level, n (%)			0.074
<= 5	117 (28.2%)	144 (34.7%)	
> 5	83 (20%)	71 (17.1%)	
Lymphatic invasion, n (%)			**0.019**
No	184 (31.6%)	166 (28.5%)	
Yes	99 (17%)	133 (22.9%)	
Residual tumor, n (%)			0.359
R0	226 (44.3%)	242 (47.5%)	
R1	4 (0.8%)	2 (0.4%)	
R2	14 (2.7%)	22 (4.3%)	

Note. $P<0.05$, and the results were statistically significant.

Supplementary Table2. Correlation between NFKBIE expression and clinicopathologic features (logistic regression)

Characteristics	Total (N)	*OR* (95% *CI*)	*P* value
Pathologic T stage (T3&T4 vs. T1&T2)	641	1.031 (0.703 – 1.514)	0.874
Pathologic N stage (N1&N2 vs. N0)	640	1.246 (0.910 – 1.705)	0.170
Pathologic M stage (M1 vs. M0)	564	1.353 (0.856 – 2.140)	0.196
Pathologic stage (Stage III&Stage IV vs. Stage I&Stage II)	623	1.197 (0.872 – 1.643)	0.265
CEA level (> 5 vs. <= 5)	415	0.695 (0.466 – 1.037)	0.075
Lymphatic invasion (Yes vs. No)	582	1.489 (1.066 – 2.080)	**0.020**
Age (> 65 vs. <= 65)	644	0.632 (0.462 – 0.866)	**0.004**
Gender (Male vs. Female)	644	1.402 (1.027 – 1.912)	**0.033**
Residual tumor (R1&R2 vs. R0)	510	1.245 (0.658 – 2.355)	0.500

Note. $P<0.05$, and the results were statistically significant.

Supplementary Table3. Univariate and multifactorial analysis of clinicopathologic parameters in CRC patients

Characteristics	Total(N)	Univariate analysis		Multivariate analysis	
		Hazard ratio (95% *CI*)	*P* value	Hazard ratio (95% *CI*)	*P* value
Pathologic T	640				
T1&T2	131	Reference		Reference	
T4&T3	509	2.468 (1.327 - 4.589)	**0.004**	2.956 (0.664 - 13.157)	0.155
Pathologic N	639				
N0	367	Reference		Reference	
N2&N1	272	2.627 (1.831 - 3.769)	**< 0.001**	0.117 (0.023 - 0.596)	**0.010**
Pathologic M	563				
M0	474	Reference		Reference	
M1	89	3.989 (2.684 - 5.929)	**< 0.001**	1.681 (0.653 - 4.327)	0.282
Pathologic stage	622				
Stage I&Stage II	348	Reference		Reference	
Stage III&Stage	274	2.988 (2.042 - 4.372)	**< 0.001**	15.618 (2.326 - 104.874)	**0.005**
Gender	643				
Female	301	Reference			
Male	342	1.054 (0.744 - 1.491)	0.769		
Age	643				
<= 65	276	Reference		Reference	
> 65	367	1.939 (1.320 - 2.849)	**< 0.001**	3.206 (1.491 - 6.895)	**0.003**
Lymphatic	581				
No	349	Reference		Reference	
Yes	232	2.144 (1.476 - 3.114)	**< 0.001**	2.616 (1.205 - 5.682)	**0.015**
CEA level	414				
<= 5	260	Reference		Reference	
> 5	154	2.620 (1.611 - 4.261)	**< 0.001**	1.400 (0.690 - 2.839)	0.351
Residual tumor	509				
R0	467	Reference		Reference	
R1&R2	42	4.609 (2.804 - 7.577)	**< 0.001**	2.457 (1.090 - 5.535)	**0.030**
NFKBIE	643				
Low	321	Reference		Reference	
High	322	1.431 (1.008 - 2.030)	**0.045**	2.115 (1.070 - 4.184)	**0.031**

Note. $P<0.05$, and the results were statistically significant.