

Supplementary Materials for: *Neutrophil extracellular traps as biomarkers for predicting prognosis and chemotherapy response in colorectal cancer*

Chen Wang^{#1,2,3,4,5}. Junxiong Chen^{#1,2,3,5}. Ziyin Song^{1,2,3,5}. Kexin Lv^{1,2,3,5}. Haiyan Dong^{1,2,3,4,5}. Liumei Liang^{1,2,3,5}. Jingdan Zhang^{1,2,3,5}. Huanliang Liu^{1,2,3,5}. Jianxia Li^{*1,2,3,5,6}. Xiangling Yang^{*1,2,3,5}.

[#]Chen Wang and Junxiong Chen were co-first authors.

^{*}Xiangling Yang and Jianxia Li contributed equally to this work as co-corresponding authors.

Email: yangxl28@mail.sysu.edu.cn; lijx55@mail2.sysu.edu.cn

Affiliations:

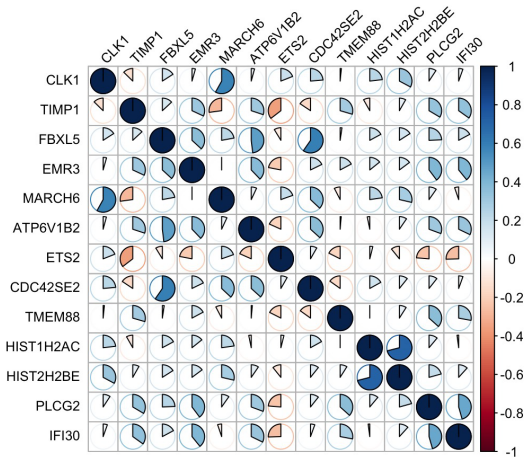
- 1 Department of Clinical Laboratory, The Sixth Affiliated Hospital, Sun Yat-sen University, Guangzhou, Guangdong 510655, China
- 2 Guangdong Provincial Key Laboratory of Colorectal and Pelvic Floor Diseases, The Sixth Affiliated Hospital, Sun Yat-sen University, Guangzhou, Guangdong 510655, China
- 3 Guangdong Institute of Gastroenterology, The Sixth Affiliated Hospital, Sun Yat-sen University, Guangzhou, Guangdong 510655, China
- 4 Department of General Surgery, The Sixth Affiliated Hospital, Sun Yat-sen University, Guangzhou, Guangdong 510655, China
- 5 Biomedical Innovation Center, The Sixth Affiliated Hospital, Sun Yat-sen University, Guangzhou, Guangdong 510655, China
- 6 Medical Oncology, The Sixth Affiliated Hospital, Sun Yat-sen University, Guangzhou, Guangdong 510655, China

Figure S1. Pairwise Correlation Analysis of 13 Prognostic NRGs.

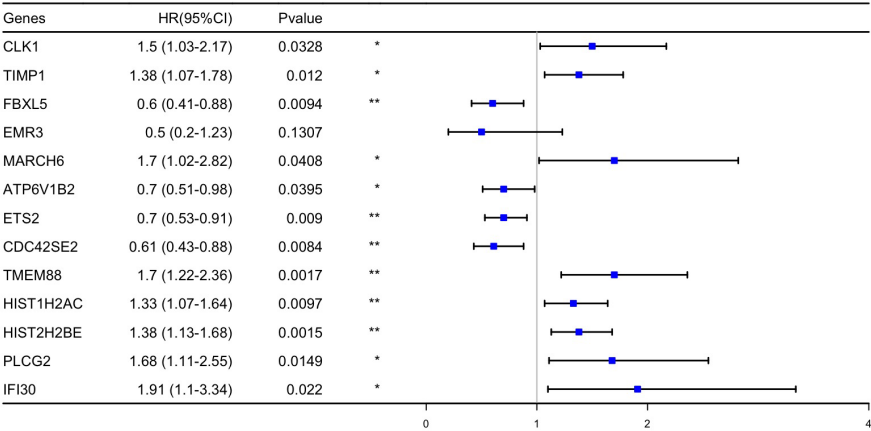
Figure S2. Cox Regression Analyses. **A** Forest plot of univariate Cox regression for NETs risk scores and clinical parameters (HR with 95% CI). **B** Forest plot of multivariate Cox regression adjusting for age, TNM stage, and gender.

Table S1. Kaplan-Meier Analysis of the NETs Risk Score Model by Tumor Stage in GSE39582.

Table S2. Functions and Clinical Significance of 13 Genes in the NETs Risk Score Model.



A



B

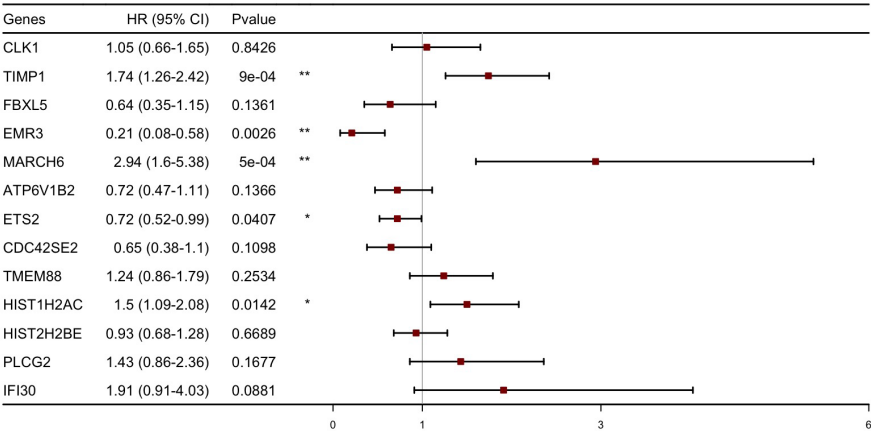


Table S1 Kaplan-Meier Analysis of the NETs Risk Score Model by Tumor Stage

Group	No. of Patients	Death Events	Median OS (days)	<i>p</i>
Early stage (I+II)	310	86	High-risk: 60.0	0.59
Advanced stage (III+IV)	269	108	High-risk: 34.5	0.0028

Table S2 Functions and Clinical Significance of 13 Genes in the NETs Risk Score Model

Gene Name	Function	Clinical Significance	References
CLK1	Phosphorylates ribosomal ribonucleoproteins and splicing factors to regulate pre-mRNA splicing, promoting tumor cell migration and invasion.	High expression in cancer patients correlates with poor prognosis.	(17-19)
TIMP1	Inhibits matrix metalloproteinases to regulate tissue remodeling, wound healing, and inflammation.	High expression in CRC patients is associated with poor prognosis.	(20)
FBXL5	Regulates cellular iron metabolism, indirectly affecting DNA repair and redox balance.	Elevated expression in colon cancer patients correlates with unfavorable outcomes.	(21, 22)
EMR3	A membrane protein involved in immune cell signaling and adhesion.	Gene knockout studies revealed that it promotes glioblastoma invasion.	(23, 24)
MARCH6	An E3 ubiquitin ligase mediating intracellular protein ubiquitination.	Upregulated in thyroid cancer tissues, it enhances thyroid cancer progression via signal transduction pathways.	(25, 26)
ATP6V1B2	Encodes a subunit of V-ATPase, maintaining intracellular acidity and membrane transport.	The expression level correlates with improved overall survival in CRC patients.	(27, 28)
ETS2	A transcription factor critical for cell growth and differentiation.	Upregulated in CRC tissues, it enhances the migratory and invasive capacities of colorectal cancer cells.	(29, 30)
CDC42SE2	Encodes a CDC42 effector protein regulating cytoskeletal rearrangement.	High expression in CRC tissues correlates with disease severity and lower patient survival rates.	(31, 32)
TMEM88	A transmembrane protein regulating the Wnt/ β -catenin pathway.	Abnormal expression of this gene is closely associated with cancer progression and prognosis across multiple cancer types.	(33, 34)
HIST1H2AC	Encodes histone H2A, maintains chromatin structure and gene regulation.	It modulates the expression of oncogenes and tumor suppressor genes through epigenetic mechanisms, thereby promoting tumorigenesis and cancer progression.	(35, 36)
HIST2H2BE	Encodes histone H2B, involved in DNA repair and cell cycle control.	By altering gene expression and chromatin structure, it promotes tumorigenesis and cancer development.	(35, 37)
PLCG2	Encodes PLC γ 2, a key mediator of immune cell signaling.	High expression in CRC is strongly associated with poor clinical outcomes in patients.	(38, 39)
IFI30	Encodes GILT, critical for antigen presentation.	High expression in breast cancer patients correlates with shortened relapse-free survival periods.	(40, 41)