

## Supplementary table

**Supplementary Table S1.** Univariate and multivariate analyses of ENO2 mRNA level and patient disease-free survival (DFS).

Variable	Univariate analysis			Multivariate analysis <sup>c</sup>		
	HR <sup>a</sup>	95%CI <sup>b</sup>	P	HR	95% CI	P
Disease-free survival						
Age (years)						
≤60 (n = 238)	1.468	0.930-2.318	0.099			
>60 (n = 198)						
Gender						
Female (n = 149)	1.144	0.699-1.873	0.591			
Male (n = 287)						
T stage						
T1 or T2 (n = 292)	6.556	3.923-10.956	0.000*			
T3 or T4 (n = 144)						
N stage						
N0 (n = 192)	7.098	3.212-15.687	0.000*	3.319	1.490-7.392	0.003*
N1 (n = 13)						
M stage						
M0 (n = 354)	11.937	7.501-18.996	0.000*	3.457	1.797-6.650	0.000*
M1 (n = 53)						
G grade						
G1 or G2 (n = 210)	5.899	3.109-11.195	0.000*	2.133	1.000-4.549	0.050*
G3 or G4 (n = 219)						
TNM stage						
I+II (n = 279)	11.458	6.173-21.270	0.000*	4.501	1.743-11.362	0.002*
III+IV (n = 115)						
ENO2						
Low(n = 225)	2.175	1.348-3.510	0.001*			
High (n = 211)						

a Hazard ratio, estimated from Cox proportional hazard regression model.

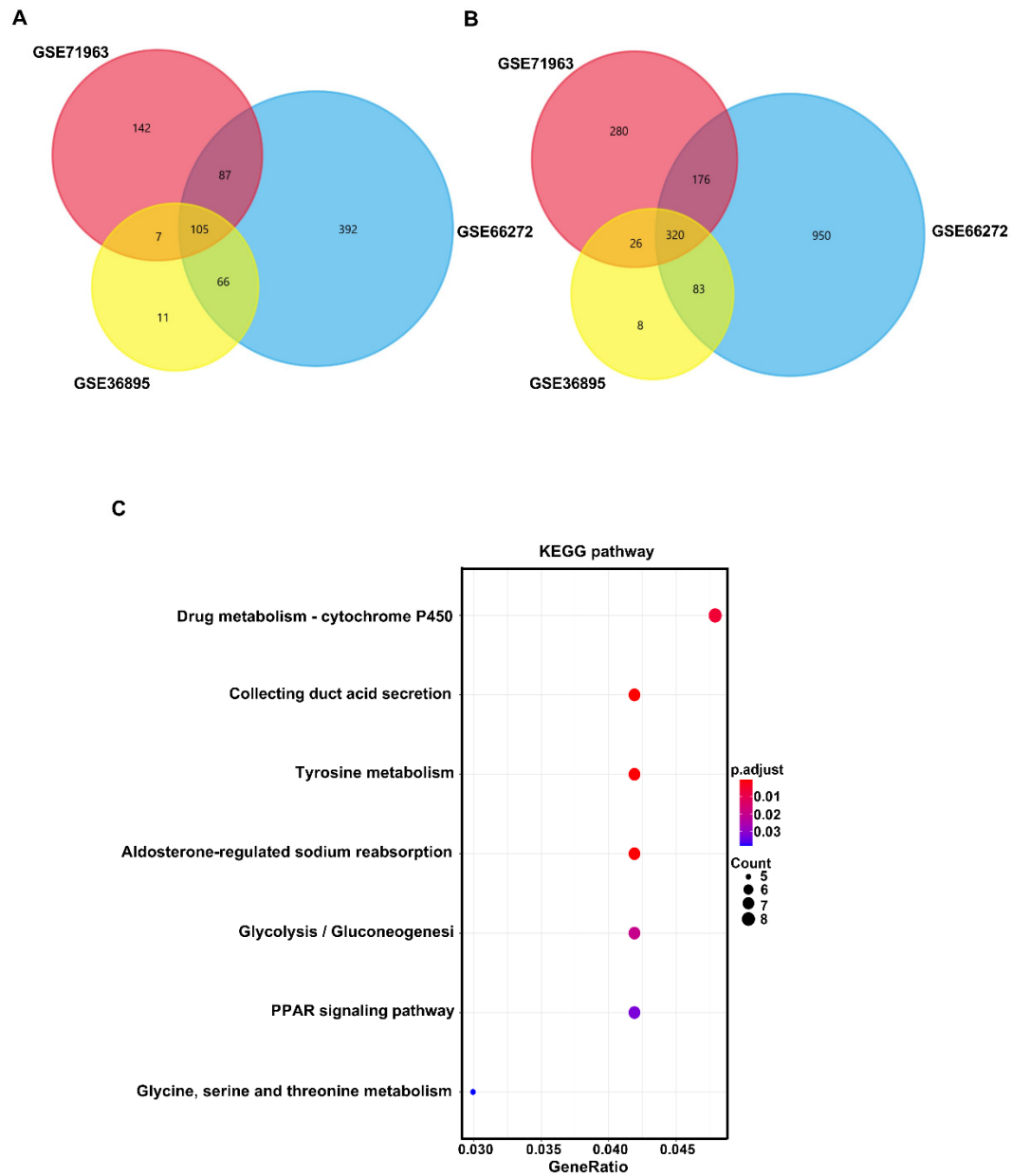
b Confidence interval of the estimated HR.

c Multivariate models were adjusted for N, M classification, G stage and TNM stage.

**Supplementary Table S2.** Correlation between ENO2 mRNA expression and clinicopathological parameters of ccRCC patients.

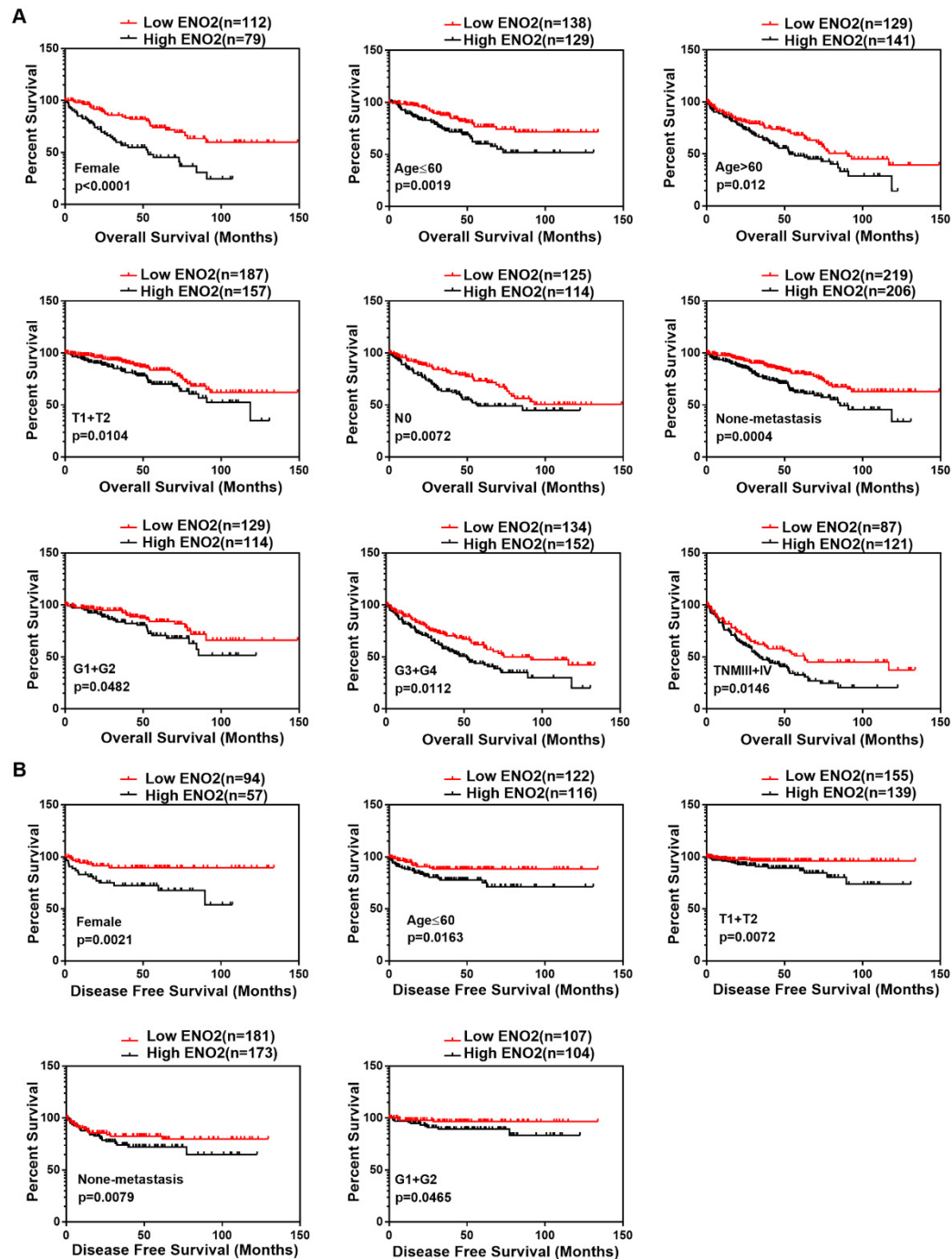
Parameter		Number	ENO2 mRNA expression		P value
			Low	High	
Age(years)	<=60	267	138	129	0.365
	>60	270	129	141	
gender	female	191	112	79	0.002*
	male	346	155	191	
T stage	T1+T2	344	187	157	0.004*
	T3+T4	193	80	113	
N stage	N0	239	125	114	0.068
	N1	17	5	12	
M stage	M0	425	219	206	0.166
	M1	79	34	45	
TNM stage	I+II	326	181	145	0.001*
	III+IV	208	85	123	
Disease	DiseaseFree	310	173	137	0.006*
Free Status	Recurred/Progressed	128	53	75	

## Supplementary figure



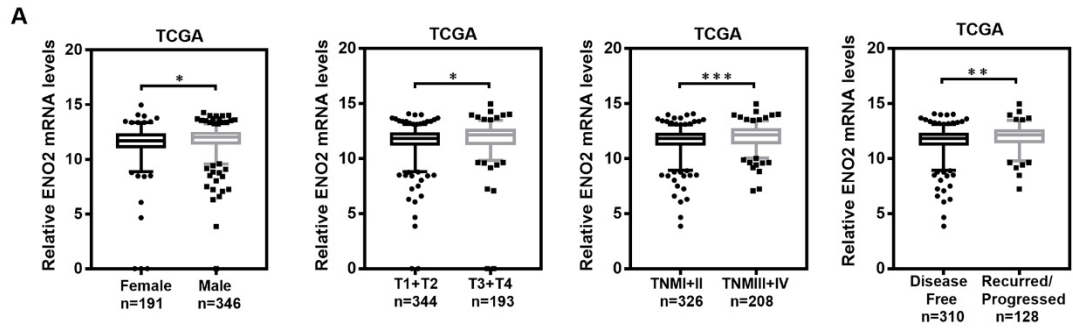
### Supplemental Figure S1. Identifying differentially expressed genes

Identify the DEGs of three ccRCC gene expression profile data GSE36895, GSE66272, GSE71963,  $p < 0.05$  and  $|FC| > 2$  are the screening criteria used. (A) 105 up-regulated DEGs in ccRCC. (B) 420 down-regulated DEGs in ccRCC. (C) Analysis of the down-regulated KEGG pathway,  $p \text{ value} < 0.05$  is considered a significant difference.



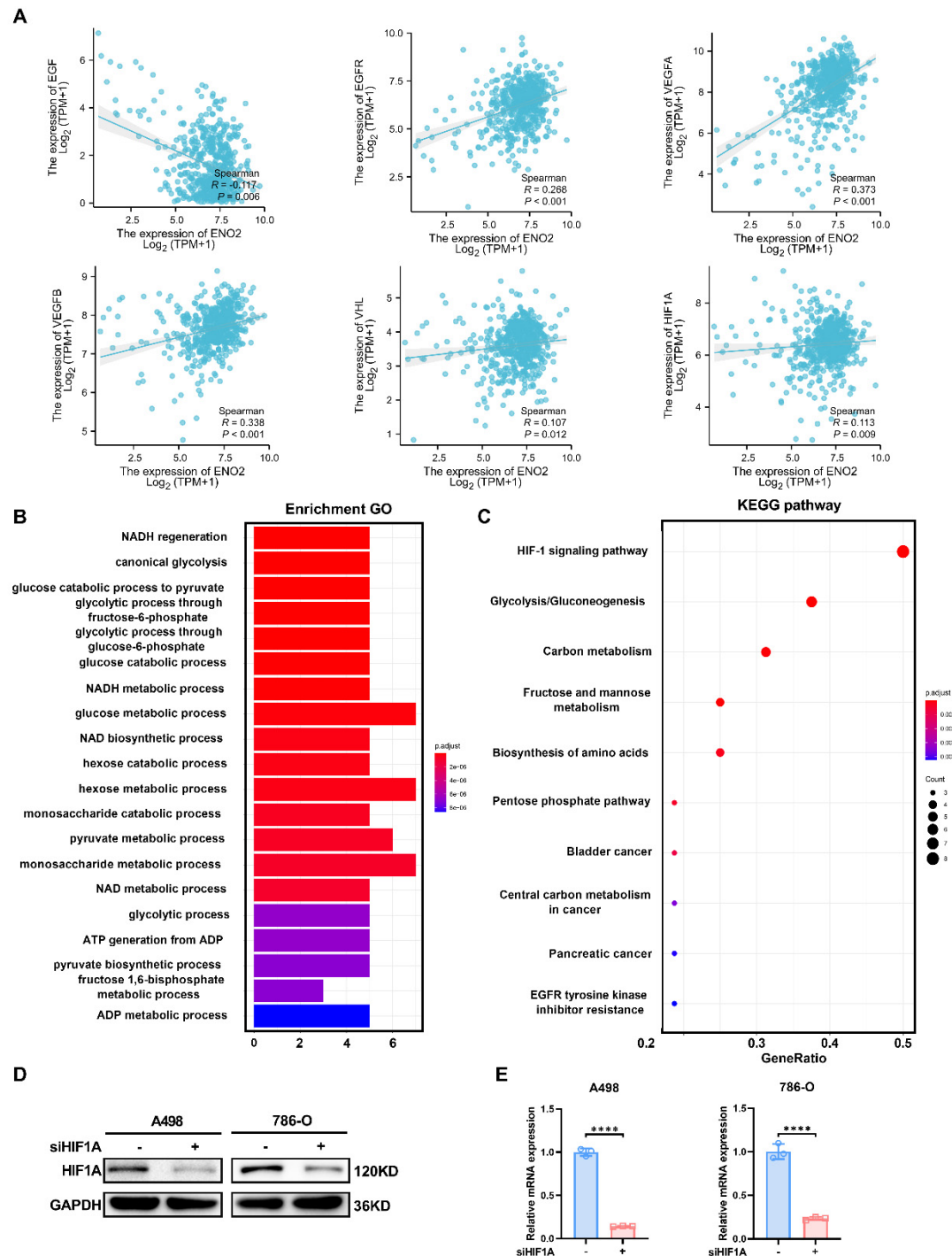
**Supplemental Figure S2. ENO2 is overexpressed and indicates poor survival in clear cell renal cell carcinoma**

(A) An analysis of Kaplan-Meier curves stratified by subgroups to determine overall survival. The p value is obtained through Log-rank (Mantel-Cox) test. (B) An analysis of Kaplan-Meier curves stratified by subgroups to determine disease-free survival. The p value is obtained through Log-rank (Mantel-Cox) test.

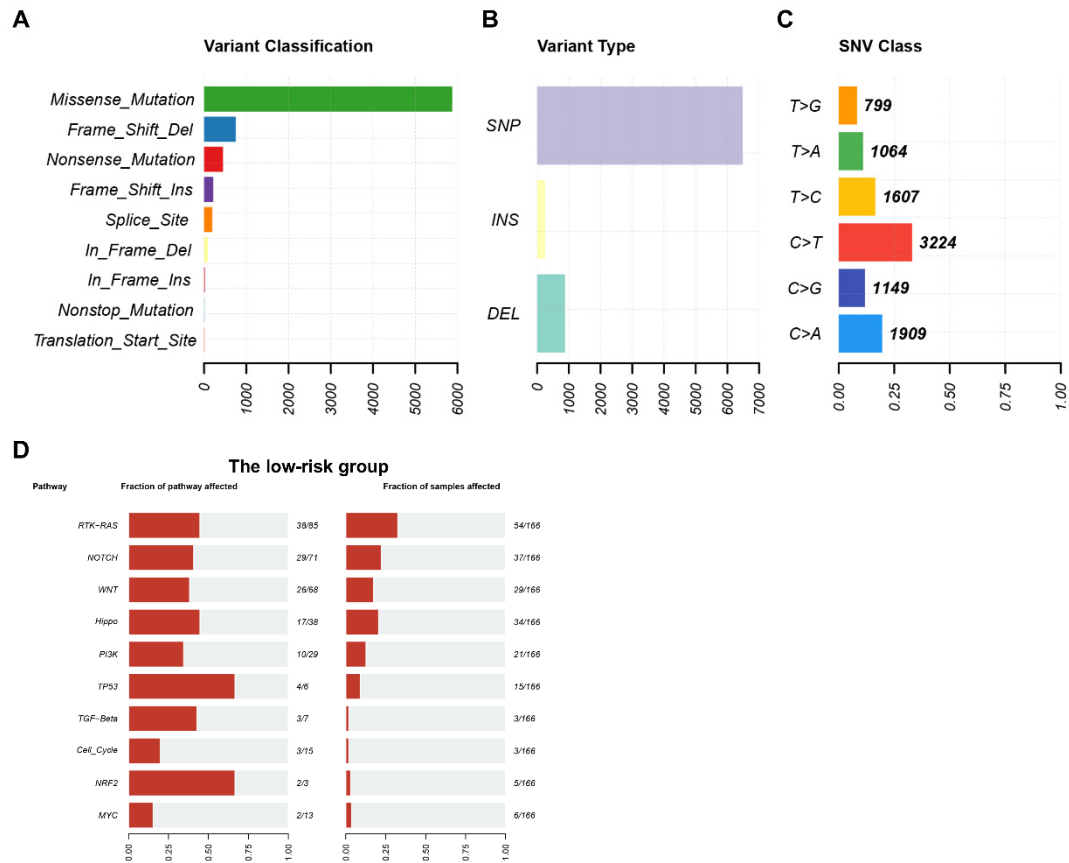


**Supplemental Figure S3. ENO2 expression correlates with clinicopathological parameters of clear cell renal cell carcinoma**

(A) The correlations between ENO2 expression and different clinicopathological parameters based on the data from the TCGA-KIRC database; t-test, \*\*\*\*  $P < 0.0001$ , \*\*\*  $P < 0.001$ , \*\*  $P < 0.01$ , and \*  $P < 0.05$  (Independent-Samples t-test for statistics).



**Supplemental Figure S4. ENO2 is significantly related to important genes in ccRCC** (A) The correlation graph between ENO2 and EGF, EGFR, VEGFA, VEGFB, VHL, HIF1A expression based on the data from the TCGA-KIRC database. (B) GO annotations for PPI network. (C) KEGG pathway analysis for PPI network. (D) Western blot analysis of HIF1A knockdown. (E) qRT-PCR results of HIF1A knockdown.



**Supplemental Figure S5. CcRCC patients with high ENO2 expression have higher levels of tumor mutation burden (TMB)**

(A-B) Gene mutation categories in the low-expression subgroup of ENO2. (C) Classification of SNVs in the low-expression subgroup of ENO2. (D) Top 10 signaling pathways enriched with mutated genes in the low-expression subgroup of ENO2.