

Supplementary Materials

Low *RCAN1.2* mRNA Expression Is Associated with Poor Prognosis of Patients with Esophageal Squamous Cell Carcinoma

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Supplementary Tables

Table S1 Primer sequences used for real-time PCR

	Forward primer	Reverse primer
<i>RCAN1.1</i>	3'-TGGAGCTTCATTGACTGCGA-3'	3'-ACGTCCCTAAAGAGGGACTCA-3'
<i>RCAN1.2</i>	3'-GGCGACGTGACTCAGTGTT-3'	3'-GGTGATGTCCTTGTCAACGTC-3'
<i>RCAN1.4</i>	3'-TTTAGCTCCCTGATTGCCTGT-3'	3'-AAAGGTGATGTCCTTGTCAACG-3'
<i>GAPDH</i>	3'-GCCACATCGCTCAGACACC-3'	3'-GATGGCAACAATATCCACTTACC-3'

Table S2 The mRNA expression level of three *RCAN1* transcripts in ESCC tumor tissues compared with matched normal tissues in GSE164158 and GSE149609 datasets.

<i>RCAN1</i> transcripts	Dataset	Fold Change	P value	P adj	Regulated
<i>RCAN1.1</i>	GSE164158	0.69	0.293398	0.583475	Down regulation
<i>RCAN1.1</i>	GSE149609	2.63	0.080205	0.325335	Up regulation
<i>RCAN1.2</i>	GSE164158	0.59	0.248591	0.535982	Down regulation
<i>RCAN1.2</i>	GSE149609	0.39	0.464981	0.777611	Down regulation
<i>RCAN1.4</i>	GSE164158	0.79	0.667443	0.85669	Down regulation
<i>RCAN1.4</i>	GSE149609	1.88	0.132468	0.428839	Up regulation

Table S3 Clinical characteristics of 100 ESCC patients according to *RCAN1.1* mRNA expression level

	<i>RCAN1.1</i> Low (n=50)	<i>RCAN1.1</i> High (n=50)	P value
Age/years			0.015*
>60	30	41	
≤60	20	9	
Gender			0.534
Female	17	20	
Male	33	30	
Smoke			0.545
Yes	20	23	
No	30	27	
Drink			0.224
Yes	24	18	
No	26	32	
Family history of cancer			0.383
Yes	17	13	
No	33	37	
cardiovascular and cerebrovascular diseases			0.841
Yes	26	27	
No	24	23	
Stage			0.198
I-II	31	37	
III-IV	19	23	
LNM			0.539
No	29	32	
Yes	21	18	
Tumor diameter			0.517
≥5cm	14	17	
<5cm	36	33	

Differentiation			0.373
Well	16	12	
Poor-Moderate	34	38	

Footnote: * $P<0.05$

Table S4 Clinical characteristics of 100 ESCC patients according to *RCAN1.4* mRNA expression level

	<i>RCAN1.4</i> Low (n=50)	<i>RCAN1.4</i> High (n=50)	<i>P</i> value
Age/years			0.015*
>60	30	41	
≤60	20	9	
Gender			0.147
Female	15	22	
Male	35	28	
Smoke			0.313
Yes	24	19	
No	26	31	
Drink			0.015*
Yes	27	15	
No	23	35	
Family history of cancer			1.000
Yes	15	15	
No	35	35	
cardiovascular and cerebrovascular diseases			0.316
Yes	29	24	
No	21	26	
Stage			0.198
I-II	31	37	
III-IV	19	13	
LNM			0.539
No	29	32	
Yes	21	18	
Tumor diameter			0.517
≥5cm	17	14	
<5cm	33	36	

Differentiation			0.026*
Well	9	19	
Poor-Moderate	41	31	

Footnote: * $P<0.05$

Table S5 Univariate and multivariate Cox analysis of factors for the overall survival of 87 esophageal cancer patients after PSM analysis.

Variables	Univariate analysis		Multivariate analysis	
	HR	P value	HR	P value
Gender (Female vs. Male)	0.7561	0.36		
Age (≤ 60 vs > 60)	0.5155	0.0621	0.6119	0.17609
Smoke (Yes vs. No)	1.5096	0.152		
Drink (Yes vs. No)	1.01032	0.972		
Family history of cancer (Yes vs. No)	0.7397	0.365		
Cardiovascular and cerebrovascular diseases (Yes vs. No)	0.7565	0.332		
<i>RCAN1.1</i> (High vs. Low)	0.8611	0.603		
<i>RCAN1.2</i> (High vs. Low)	0.5331	0.0398*	0.5446	0.05737
<i>RCAN1.4</i> (High vs. Low)	0.8874	0.677		
LNM (Yes vs. No)	3.0092	0.000194***	3.9346	0.00272**
Stage (I-II vs III-IV)	0.4506	0.00575**	1.6836	0.24633
Tumor diameter (≥ 5 cm vs < 5 cm)	0.6509	0.175		
Differentiation (Well vs Poor- moderate)	1.2765	0.41		

Footnote: * $P < 0.05$, ** $P < 0.001$, *** $P < 0.001$