Centrosomal protein 55 activates NF-kB signalling and promotes pancreatic cancer cells aggressiveness

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Figure S1. Elevated CEP55 mRNA expression in pancreatic cancer.

(a) Gene set enrichment analysis (GSEA) plot indicating a significant correlation between *CEP55* mRNA expression level and those genes up-regulated in pancreatic cancer gene signatures (GRUETZMANN_PANCREATIC_CANCER_UP; TCGA, n=178, GSE71729, n=145). (b) Quantitative real-time reverse transcription-PCR detection of *CEP55* mRNA in three normal pancreas tissues and nine pancreatic cancer tissues. (c) Expression of *CEP55* mRNA as detected by qRT-PCR analysis in SW1990 and Capan-1 cells upon transfection with CEP55 or CPE55 siRNAs. GAPDH was used as the loading control. Bars represent the mean \pm SD of three independent experiments. **P* < 0.05.



Figure S2. NF-κB activation could partially rescue the reduced proliferation of PANC cells upon depletion of CEP55.

(a) Western blotting analysis of p-IKK β and I κ B α expression in CEP55-downregulating SW1990 and Capan-1 cells upon transfection with siI κ B α . (b) MTT assay of CEP55-downregulating SW1990 and Capan-1 cells transduced with siI κ B α . (c) Representative micrographs (left panel) and quantification (right panel) of crystal violet-stained colonies of CEP55-knockdown SW1990 and Capan-1 cells transduced with siI κ B α after 14 days of colony formation.



Figure S3. The Spearman correlation coefficient was calculated to assess the significance of association between CEP55 protein and NF- κ B downstream targets in 9 PANC specimens. **P*<0.05.



Figure S4. The activity of MMP2 and MMP9 in pancreatic cancer cells upon transfection wieh CEP55 or siCEP55.

Characteristics	Number of cases
Gender	
Male	80
Female	46
Age(years)	
>57	59
≤57	67
Clinical Stage	
Ι	47
II	34
III	34
IV	11
T classification	
T1	25
T2	47
T3	34
T4	20
N classification	
NO	81
N1	45
M classification	
M0	115
M1	11
Expression of CEP55	
Relatively high expression	70
Relatively low expression	56
Vital status	
Survive	35
Mortality	91

Table S1. Clinicopathological characteristics of PANC patient samples.

Chi-square CEP55 **Protein level** test Characteristic **P** value High Low Age (> 57 versus 32/38 27/29 0.858 \leq 57 years) Gender (M versus F) 0.198 48/22 32/24 Clinical stage (I/II 38/32 43/13 0.009 versus III/IV) T classification (T1/T2 versus 34/36 38/18 0.032 T3/T4) N classification (N0 versus N1) 39/31 42/14 0.027 0.022 M classification (M0 versus M1) 60/10 55/1

Table S2. Correlation between CEP55 Expression and Clinicopathologic

Charateristics of Pancreatic Cancer Patient.

Gene	Real-time PCR primer
CEP55	5'- TTGGAACAAC AGATGCAGGC-3' and
	5'- GAGTGCAGCAGTGGGACTTT -3'
МҮС	5'-TCAAGAGGCGAACACACAAC-3' and
	5'-GGCCTTTTCATTGTTTTCCA-3'
IL6	5'-TCTCCACAAGCGCCTTCG-3' and
	5'-CTCAGGGCTGAGATGCCG-3'
BCLXL	5'-TCCTTGTCTACGCTTTCCACG-3' and
	5'-GGTCGCATTGTGGCCTTT-3'
VEGFC	5'-GTGTCCAGTGTAGATGAACTC-3' and
	5'-ATCTGTAGACGGACACACATG-3'
MMP9	5'-TTGGTCCACCTGGTTCAACT-3' and
	5'-ACGACGTCTTCCAGTACCGA-3'
TWIST	5'-GGAGTCCGCAGTCTTACGAG-3' and
	5'-TCTGGAGGACCTGGTAGAGG-3'
CCND1	5'-AACTACCTGGACCGCTTCCT-3' and
	5'-CCACTTGAGCTTGTTCACCA-3'
GAPDH	5'-ATTCCACCCATGGCAAATTC-3' and
	5'-TGGGATTTCCATTGATGACAAG-3'

Table S3. Primers used for detecting genes expression.