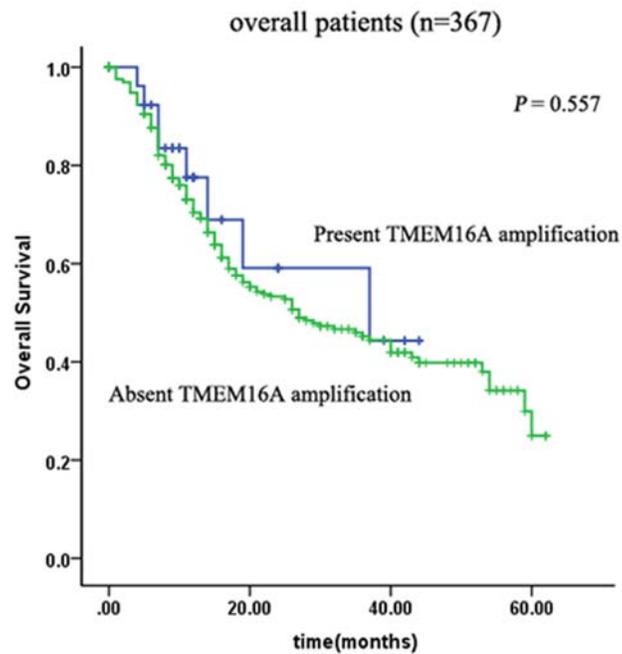
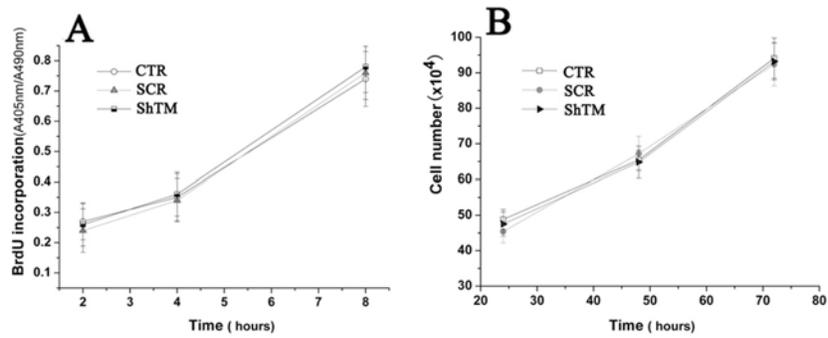


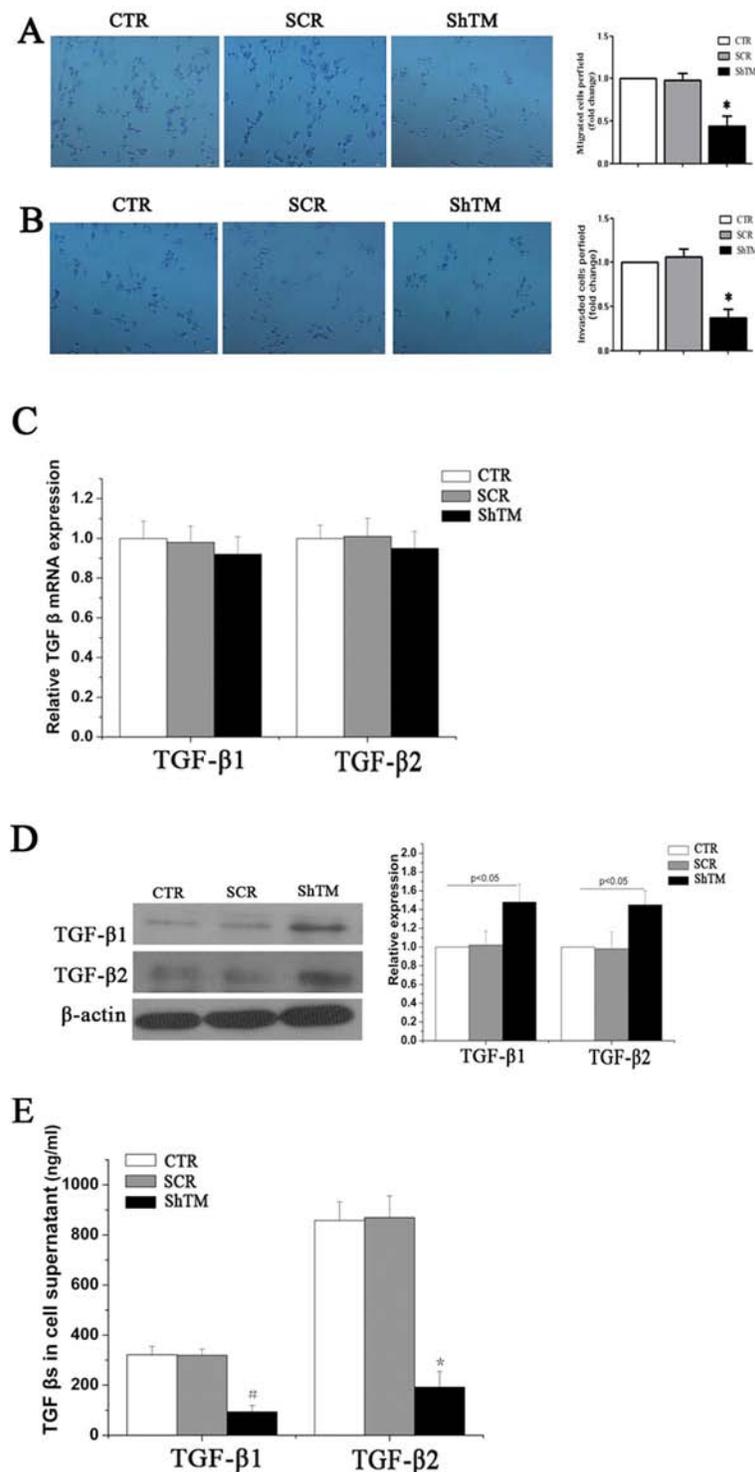
SUPPLEMENTARY FIGURES AND TABLES



Supplementary Figure S1: Kaplan–Meier estimated of overall survival according to TMEM16A amplification in patients with gastric cancer (log-rank test). Amplification of TMEM16A was not significantly correlated with overall survival ($p = 0.557$).

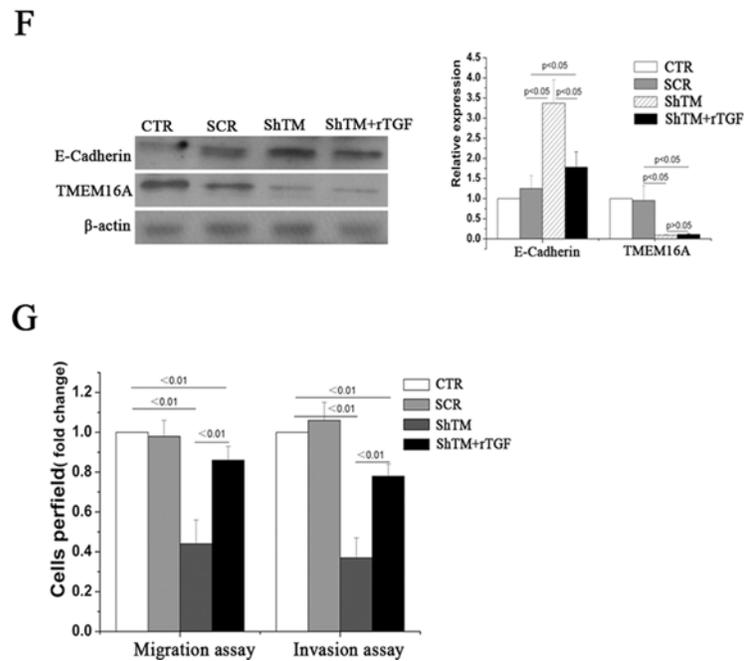


Supplementary Figure S2: Knockdown of TMEM16A does not affect BGC-823 cell proliferation. A. BrdU incorporation assay at 2 hours, 4 hours and 8 hours and B. cell count experiments at 24 hours, 48 hours and 72 hours from CTR group, SCR group and ShTM group ($n = 6, p > 0.05$).



Supplementary Figure S3: Knockdown of TMEM16A prevents the invasion and migration of BGC-823 cells by TGF-βs signalling. **A.** The transwell migration assay and **B.** transwell invasion assay revealed TMEM16A knockdown reduced migration and invasion ($n = 6$, $*p < 0.05$ vs CTR group). **C.** Knockdown of TMEM16A did not affect mRNA expression of TGF-β1 and TGF-β2 in BGC-823 cells ($n = 6$, $p > 0.05$). **D.** Protein expression of TGF-β1 and TGF-β2 in BGC-823 cells increased when knockdown of TMEM16A as shown by western blot analysis ($n = 6$, $p < 0.05$ vs CTR group). **E.** Supernatant concentrations of TGF-β1 and TGF-β2 dramatically reduced when knockdown of TMEM16A as shown by ELISA ($n = 6$, $^{\#}p < 0.01$ vs CTR group, $*p < 0.05$ vs CTR group).

(Continued)



Supplementary Figure S3 (Continued): E. Supernatant concentrations of TGF- β 1 and TGF- β 2 dramatically reduced when knockdown of TMEM16A as shown by ELISA ($n = 6$, # $p < 0.01$ vs CTR group, * $p < 0.05$ vs CTR group). F. Adding recombinant purified TGF- β increased E-cadherin expression in BGC-823 cells with TMEM16A knockdown ($n = 6$). G. Supplement of recombinant purified TGF- β increased the migration and invasion of cells with TMEM16A knockdown ($n = 6$). CTR: control BGC-823, SCR: scrambled BGC-823, ShTM: shTMEM16A BGC-823.

Supplementary Table S1. The correlation between TMEM16A expression and amplification in gastric cancer tissues

Variables		All cases	TMEM16A expression		P value	Phi
			High(%)	Low(%)		
TMEM16A amplification	Absent(%)	340	230(67.6%)	110(32.4%)	0.021	0.120
	Present(%)	27	24(88.9%)	3(11.1%)		

Supplementary Table S2. Correlation of TMEM16A amplification and TMEM16A expression with patients' features in gastric cancer

Variables	All cases	TMEM16A amplification (overall patients <i>n</i> = 367)			TMEM16A expression (overall patients <i>n</i> = 367)		
		Absent	Present	<i>P</i> value ^a	Low	High	<i>P</i> value ^a
Gender							
Male	256	235	21	0.346	74	182	0.235
Female	111	105	6		39	72	
Age at surgery							
≥57 ^b	191	174	17	0.238	51	140	0.077
<57	176	166	10		62	114	
Tumor location							
Upper half	193	179	14	0.484	57	136	0.695
Lower half	158	145	13		52	106	
Whole	16	16	0		4	12	
Tumor size							
≥5cm ^c	221	205	16	0.916	66	155	0.636
<5cm	146	135	11		47	99	
Histological type							
Intestinal	288	267	21	0.927	84	204	0.198
Diffuse	79	73	6		29	50	
TNM							
I + II	139	121	18	0.001	61	78	0.000
III + IV	228	219	9		52	176	
Lymphnode metastasis							
Present	249	236	13	0.023	57	192	0.000
Absent	118	104	14		56	62	

^achi-square test^bmedian age^cmedian tumor size

Supplementary Table S3. Primers used for real-time PCR

Gene and primer		Sequences(5' to 3')
NM_000660.4	TGF- β 1_F	CCCACAACGAAATCTATGACAA
	TGF- β 1_R	AAGATAACCACTCTGGCGAGTC
NM_001135599.2	TGF- β 2_F	ATTGCCCTCCTACAGACTTGAG
	TGF- β 2_R	CAGCACAGAAGTTGGCATTGTA
NM_002046.3	GAPDH_F	GCACCGTCAAGGCTGAGAAC
	GAPDH_R	TGGTGAAGACGCCAGTGGA