Supplementary Materials: The Epithelial and Stromal Immune Microenvironment in Gastric Cancer: A Comprehensive Analysis Reveals Prognostic Factors with Digital Cytometry

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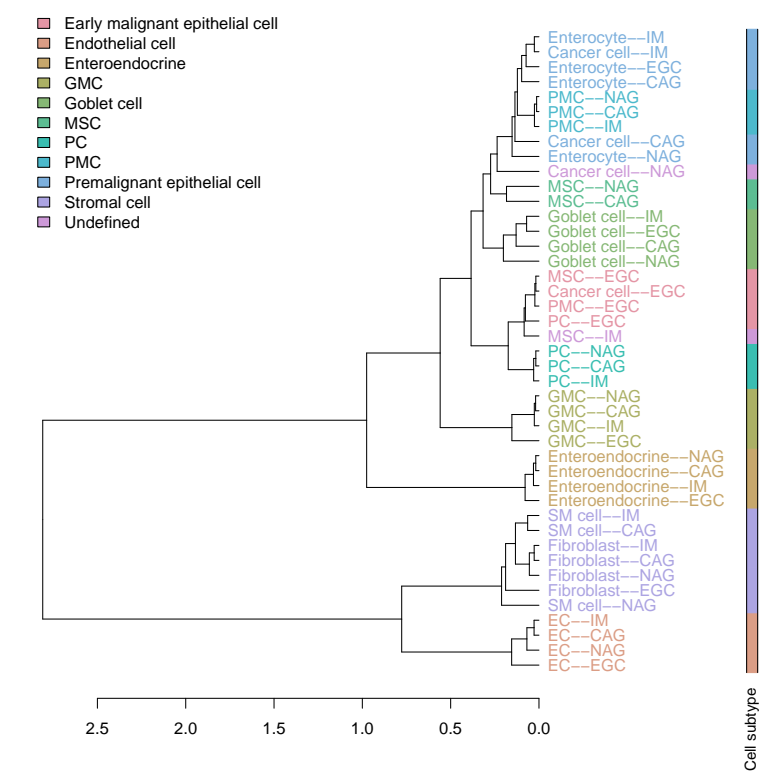

Figure S1. Cluster tree of non-immune cell types of patients with NAG, CAG, IM and EGC (Top 50 genes).
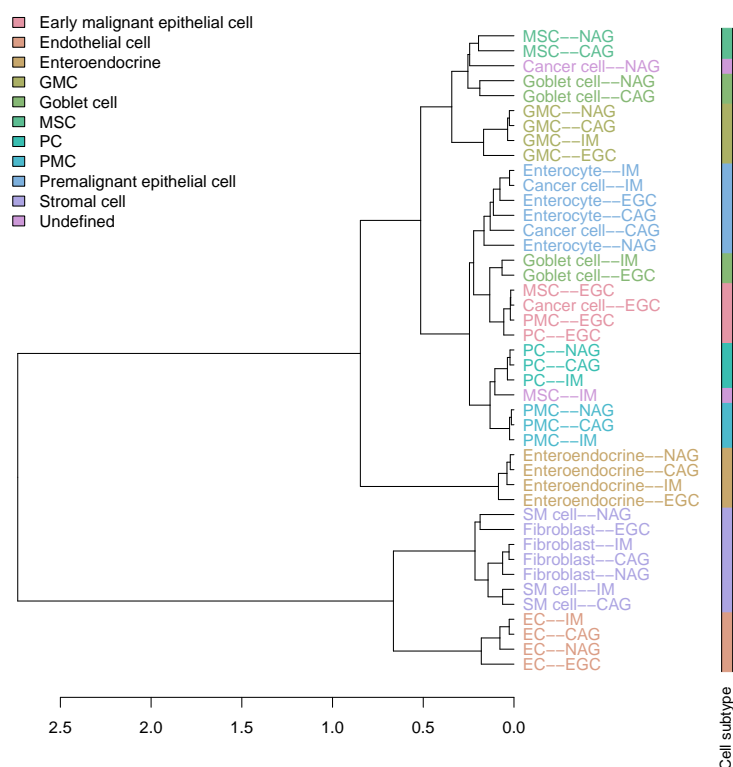
Figure S2. Cluster tree of non-immune cell types of patients with NAG, CAG, IM and EGC (Top 150 genes).
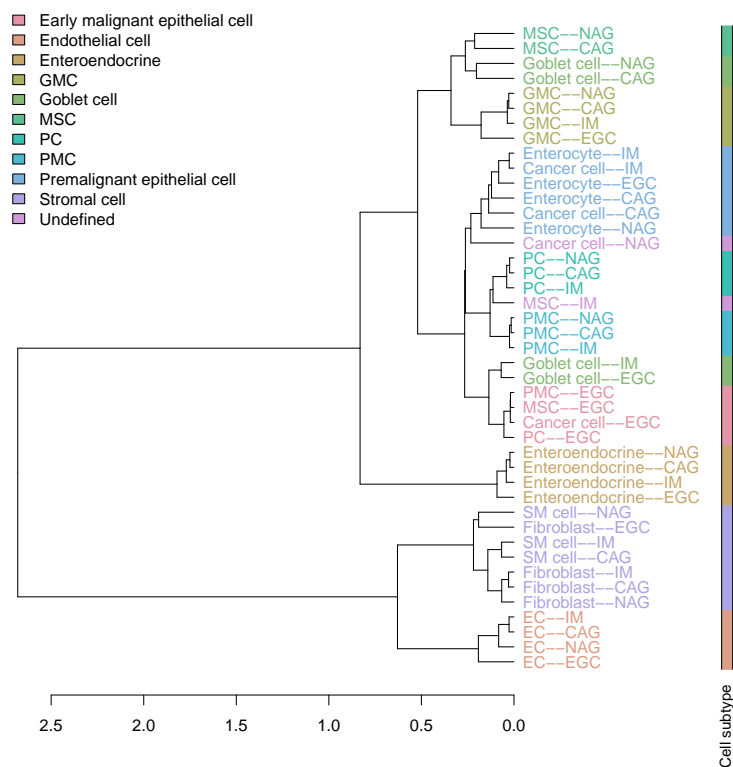

Figure S3. Cluster tree of non-immune cell types of patients with NAG, CAG, IM and EGC (Top 200 genes).
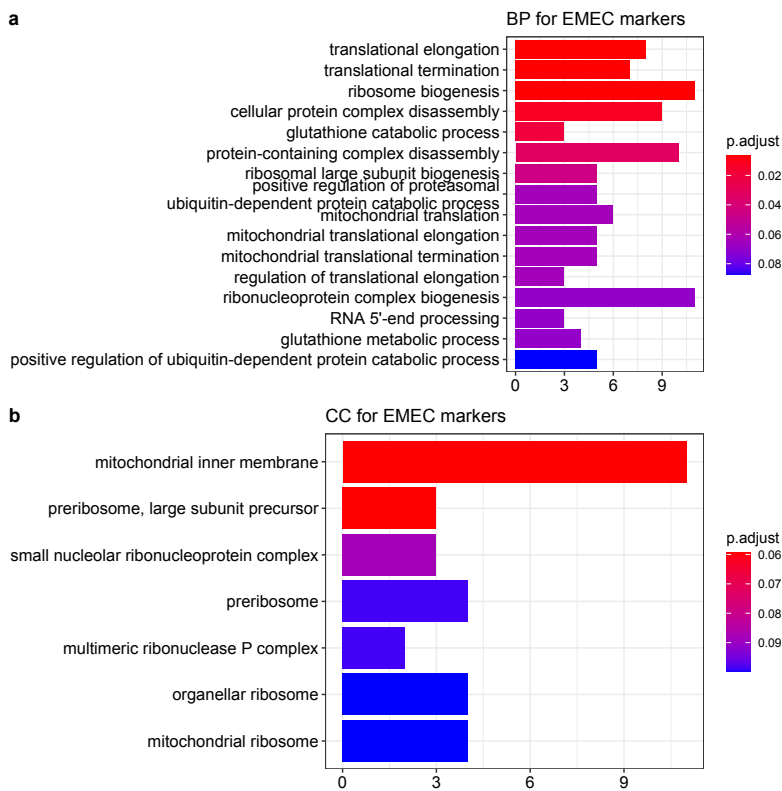
a

b

Figure S4. Top GO terms enriched for the EMEC gene signatures in the BP (**a**) and CC (**b**) ontologies.
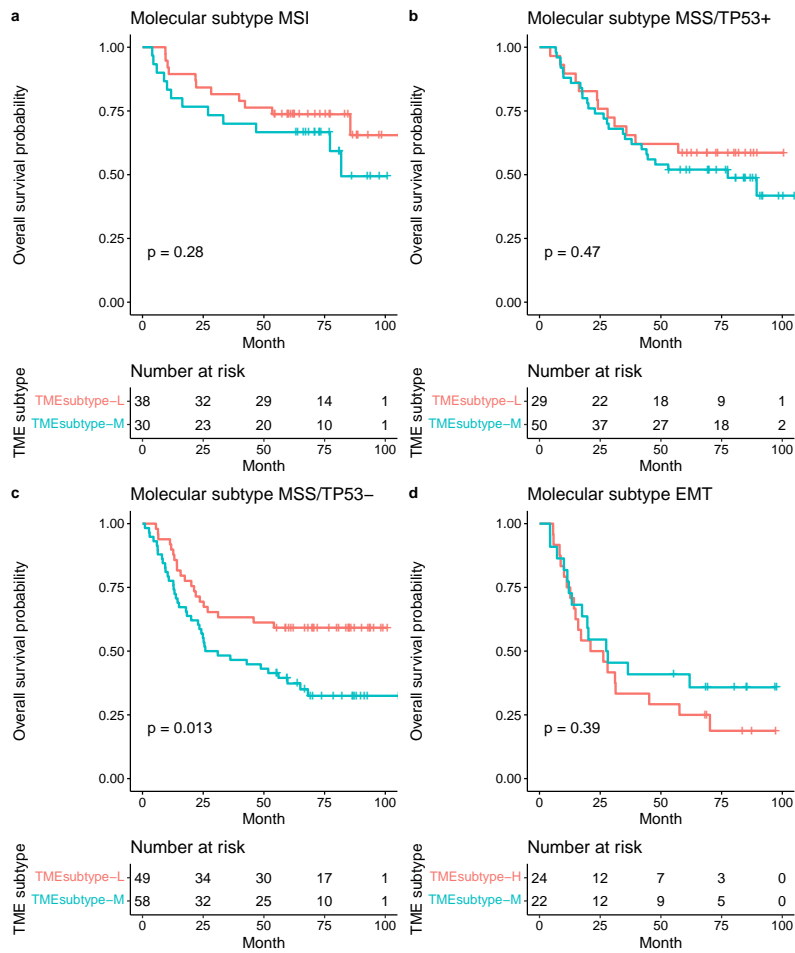
Figure S5. Complementary prognostic value of the TME subtypes to the ACRG molecular subtypes.
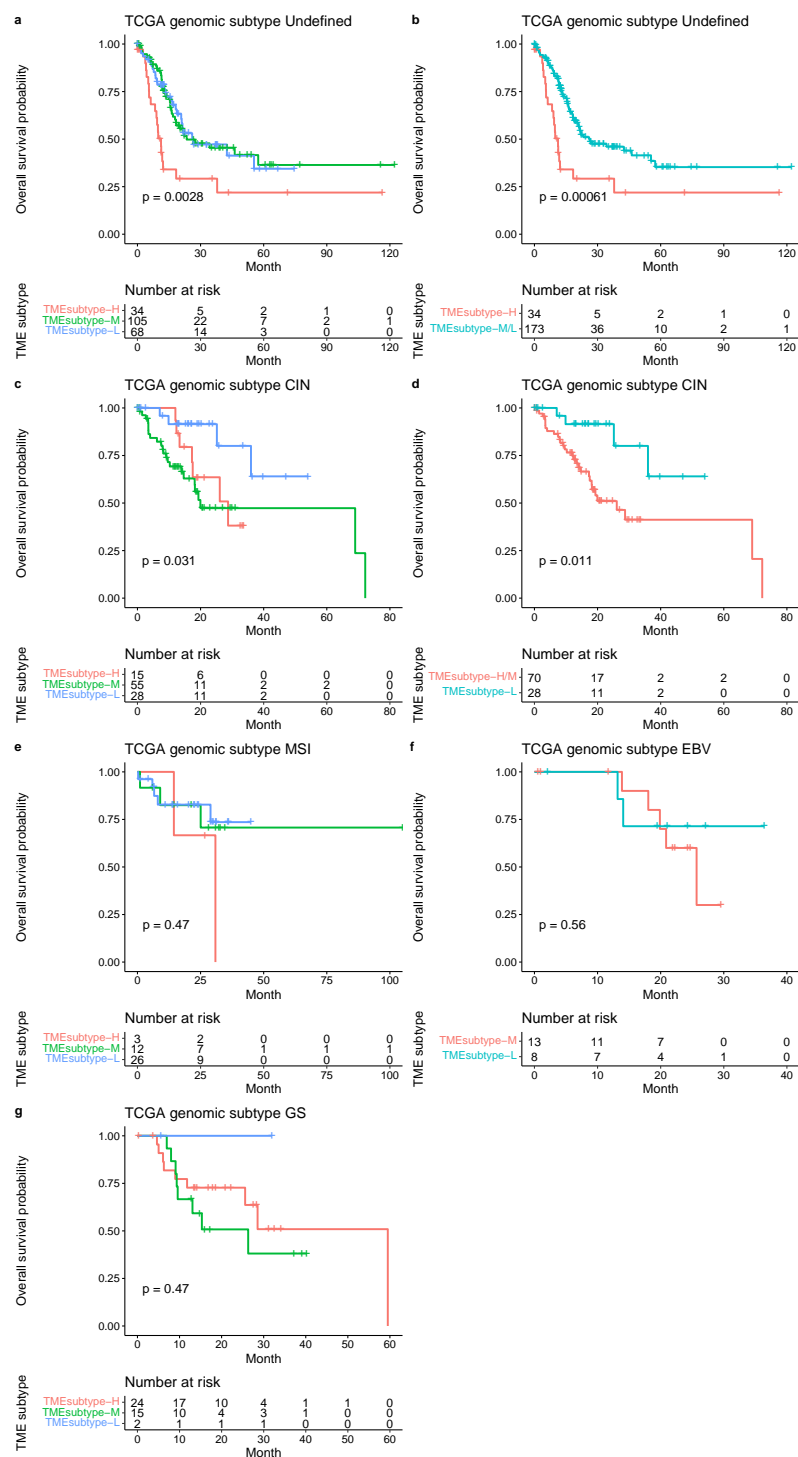
Figure S6. Complementary prognostic value of the TME subtypes to the TCGA genomic subtypes.
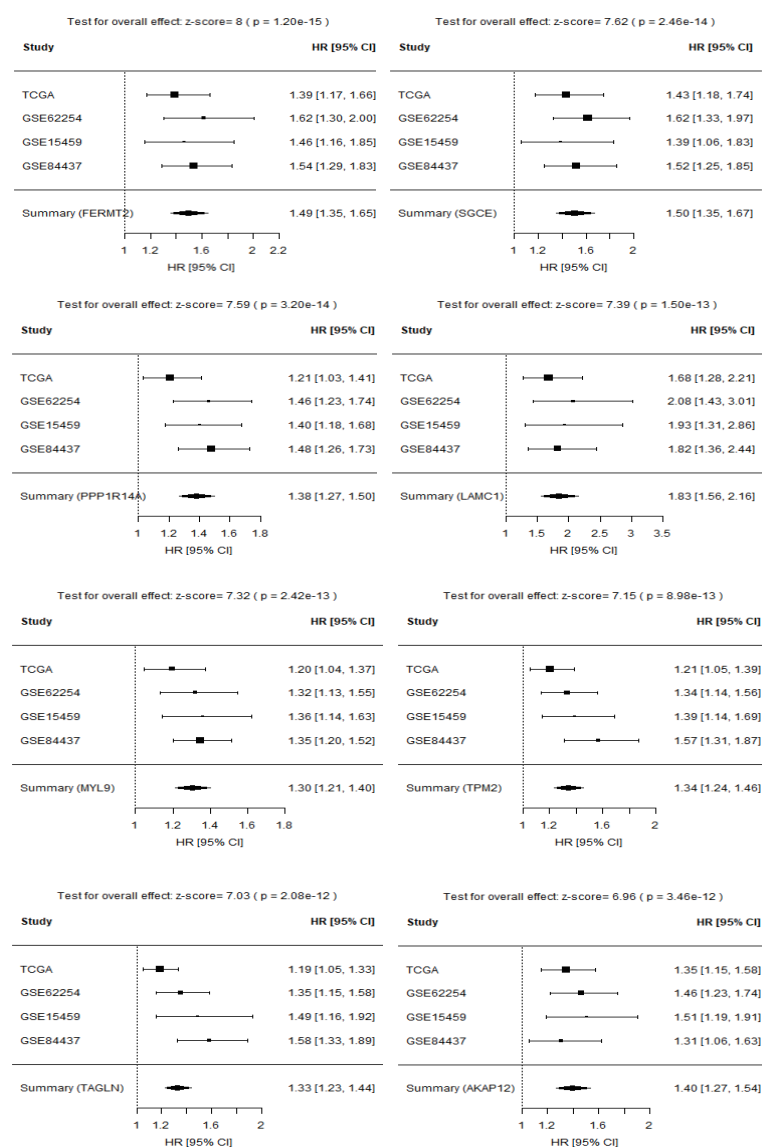
Figure S7. Forest plot of estimated hazard ratios of prognostic gene signatures in stromal cell subtype.

Figure S8. Forest plot of estimated hazard ratios of prognostic gene signatures in EMEC population.

Table S1. Clinicopathologic and treatment information for patients in the discovery and validation cohorts. A dash indicates that data are not available.

	ACRG	GSE84437	GSE15459	TCGA-STAD
No. of patients	300	433	192	415
Median age in yrs (range)	64 (24–86)	62 (27–86)	67 (23–92)	67 (30–90)
Male (%)	199 (66)	296 (68)	125 (65)	268 (65)
Stage				
I	30 (10)	-	31 (16)	57 (15)
II	96 (32)	-	29 (15)	123 (32)
III	95 (32)	-	72 (38)	169 (43)
IV	77 (26)	-	60 (31)	41 (11)
Lauren (%)				
Intestinal	146 (49)	-	99 (52)	176 (72)
Diffuse/mixed	151 (51)	-	93 (48)	69 (28)
Chemotherapy (%)	144 (48)	-	-	-
Radiationtherapy	-	-	-	72 (19)
Median follow-up (mo.)	80	116	74	22

Table S2. Grouping of the non-immune cell types for bulk gene expression samples deconvolution.

Non-immune cell types	Full name	Stages	Grouping for Bulk deconvolution
Cancer cell	Cancer cell	EGC	EMEC
MSC	Metaplastic stem-like cell	EGC	EMEC
PC	Proliferative cell	EGC	EMEC
PMC	Pit mucous cell	EGC	EMEC
EC	Endothelial cell	CAG	Endothelial cell
EC	Endothelial cell	EGC	Endothelial cell
EC	Endothelial cell	IM	Endothelial cell
EC	Endothelial cell	NAG	Endothelial cell
Enteroendocrine	Enteroendocrine cell	CAG	Enteroendocrine
Enteroendocrine	Enteroendocrine cell	EGC	Enteroendocrine
Enteroendocrine	Enteroendocrine cell	IM	Enteroendocrine
Enteroendocrine	Enteroendocrine cell	NAG	Enteroendocrine
GMC	Antral basal gland mucous cell	CAG	GMC
GMC	Antral basal gland mucous cell	EGC	GMC
GMC	Antral basal gland mucous cell	IM	GMC
GMC	Antral basal gland mucous cell	NAG	GMC
Goblet cell	Goblet cell	CAG	Goblet cell
Goblet cell	Goblet cell	EGC	Goblet cell
Goblet cell	Goblet cell	IM	Goblet cell
Goblet cell	Goblet cell	NAG	Goblet cell
MSC	Metaplastic stem-like cell	CAG	MSC
MSC	Metaplastic stem-like cell	NAG	MSC
PC	Proliferative cell	CAG	PC
PC	Proliferative cell	IM	PC
PC	Proliferative cell	NAG	PC
PMC	Pit mucous cell	CAG	PMC
PMC	Pit mucous cell	IM	PMC
PMC	Pit mucous cell	NAG	PMC
Cancer cell	Cancer cell	CAG	PMEC
Cancer cell	Cancer cell	IM	PMEC
Enterocyte	Enterocyte	CAG	PMEC
Enterocyte	Enterocyte	EGC	PMEC
Enterocyte	Enterocyte	IM	PMEC
Enterocyte	Enterocyte	NAG	PMEC
Fibroblast	Fibroblast	CAG	Stromal cell
Fibroblast	Fibroblast	EGC	Stromal cell
Fibroblast	Fibroblast	IM	Stromal cell
Fibroblast	Fibroblast	NAG	Stromal cell
SM cell	Smooth muscle cell	CAG	Stromal cell
SM cell	Smooth muscle cell	IM	Stromal cell
SM cell	Smooth muscle cell	NAG	Stromal cell
Cancer cell	Cancer cell	NAG	Undefined
MSC	Metaplastic stem-like cell	IM	Undefined

Table S3. Comparision of deconvolution accuracy on artificial bulk data by combining different signature matrices. The table shows average root mean square error (RMSD), mean absolute deviation (mAD), and Pearson correlation (R) across all samples and cell types.

Top M genes	RMSD	mAD	R
Top 50	0.0837	0.0492	0.8907
Top 100	0.0837	0.0492	0.8907
Top 150	0.0836	0.0488	0.8923
Top 200	0.0840	0.0488	0.8917
Top 250	0.0847	0.0495	0.8899

Table S4. Grouping of the immune cell types for bulk gene expression samples deconvolution.

Immune cell types	Full name	Grouping for Bulk deconvolution
Monocytes C	Classical monocytes	Monocytes
Monocytes NC+I	Non-classical/intermediate monocytes	Monocytes
NK	Natural killer cells	NK
T gd non-Vd2	$\gamma\delta$ non-Vδ2 T cells	T Innate
T gd Vd2	$\gamma\delta$ Vδ2 T cells	T Innate
Neutrophils LD	Low-density neutrophils	Granulocytes
Basophils LD	Low-density basophils	Granulocytes
mDCs	Myeloid dendritic cells	DCs
pDCs	Plasmacytoid dendritic cells	DCs
T CD8 Naive	Naïve CD8 T cells	T adaptive
T CD8 Memory	Memory CD8 T cells	T adaptive
B Naive	Naive B cells	B adaptive
B Memory	Memory B cells	B adaptive
Plasmablasts	Plasmablasts	B adaptive
T CD4 Naive	Naïve CD4 T cells	T adaptive
T CD4 Memory	Memory CD4 T cells	T adaptive
MAIT	Mucosal associated invariant T cells	T Innate