Figure S1. LRFN2 expression in pan-cancers of TCGA and Xiangya BLCA cohort. (A) Relative LRFN2 expression in normal tissue and tumor in pan-cancers of TCGA. (B) Relative LRFN2 expression in normal tissue and tumor in the Xiangya BLCA cohort. *P<0.05, **P<0.01, ***P<0.001.

Figure S2. LRFN2 expression and tumor immune microenvironment in pan-cancers. (A) Heatmap of LRFN2 and multiple immunomodulators in pan-cancers. (B) 28 tumor infiltrating immune cells and LRFN2 expression in pan-cancers. (C) Relationship between LRFN2 and PD-1, PD-L1, CTLA-4 and LAG-3 in BLCA. *P<0.05, **P<0.01, ***P<0.001, ****P<0.0001.

Figure S3. Expression pattern of LRFN2 in TCGA-BLCA. (A) Heatmap of multiple immunomodulators and low and high LRFN2 expression in TCGA-BLCA. (B-C) LRFN2 expression and T cell-inflamed related genes (B) and immune checkpoint genes (C) in TCGA-BLCA.

Figure S4. Correlations among LRFN2, immune related genes, and stromal related genes. (A) Venn diagram of LRFN2 down, immune down and stromal down. (B) Venn diagram of LRFN2 DEGs, immune DEGs and stromal DEGs.

Figure S5. LRFN2 predicts BLCA molecular subtypes and treatment regimen efficacy. (A) Correlation between high and low LRFN2 expression and seven independent molecular typing systems and 12 signature of bladder cancer. (B) LRFN2 validate BLCA molecular subtypes accuracy. (C) LRFN2 expression and different therapeutic targets. (D-F) Differences in the

distribution of CNV patterns of multiple immunotherapies over progress-related genes in the TCGA cohort (D), mutation patterns of neoadjuvant chemotherapy efficacy predictors (E), differences in the expression distribution of various bladder cancer drug target molecules screened in Drugbank database (F) in high- and low-LRFN2 expression groups.

Figure S6-S8: Validation the role of LRFN2 in predicting TME infiltration characterization in GSE32894, GSE13507, and GSE70691. (A) Heatmap of correlation between multiple immunomodulators and LRFN2 low and high expression. (B) High- and low-LRFN2 expression showed different infiltrated immune cells. (C-D) LRFN2 expression and various immune checkpoint genes (C) and T cell-inflamed related genes (D). (E) LRFN2 validate BLCA molecular subtypes accuracy. (F) LRFN2 expression and immune cell effector genes. (G) LRFN2 expression and different therapeutic targets. (H) LRFN2 expression and molecular subtypes in BLCA. ns, P>0.05, *P<0.05, **P<0.01, ***P<0.001, ****P<0.0001.

Figure S9-S11: Validation the role of LRFN2 in predicting TME infiltration characterization in GSE48075, GSE48276, and GSE52219. (A) Heatmap of correlation between multiple immunomodulators and LRFN2 low and high expression. (B) High- and low-LRFN2 expression showed different infiltrated immune cells. (C) High- and low-LRFN2 expression showed differences in DEGs enriched pathways. (D) LRFN2 expression and immune cell effector genes. (E) LRFN2 expression and various immune checkpoint genes. (F) LRFN2 validate BLCA molecular subtypes accuracy. (G) LRFN2 expression and molecular subtypes in BLCA. (H) LRFN2 expression and different therapeutic targets. ns, P>0.05, *P<0.05, **P<0.01, ***P<0.001, ****P<0.0001.

Figure S12: Validation the role of LRFN2 in predicting TME infiltration characterization in imvigor210. (A) Correlation between LRFN2 expression and different therapeutic targets. (B) Correlation between LRFN2 expression and cancer immunity cycles (bottom left) and infiltrated immune cells (upper right). (C) The accuracy of LRFN2 in predicting molecular subtypes. (D) LRFN2 expression and molecular subtypes in BLCA. (E) Differences in DEGs enriched pathways in high- and low-LRFN2 expression groups. ns, P>0.05, *P<0.05, ****P<0.0001.

Figure S13-S14: Validation of the role of LRFN2 in predicting TME infiltration characterization in GSE104922 and GSE120736. (A) Heatmap of correlation between multiple immunomodulators and LRFN2 low and high expression. (B) LRFN2 expression and infiltrated immune cells. (C) LRFN2 expression and immune cell effector genes. (D) LRFN2 validate BLCA molecular subtypes accuracy. (E-F) LRFN2 expression and various immune checkpoint genes (E) and T cell-inflamed related genes (F). (G) Correlation between LRFN2 expression and molecular subtypes in BLCA. (H) LRFN2 expression and different therapeutic targets.

Figure S15-S16: Validation the role of LRFN2 in predicting TME infiltration characterization in GSE52329 and GSE69795. (A) Heatmap of correlation between multiple immunomodulators and LRFN2 low and high expression. (B) LRFN2 expression and infiltrated immune cells. (C-D) Correlation between LRFN2 and various immune checkpoint genes (C) and T cell-inflamed related genes (D). (E) LRFN2 validate BLCA molecular subtypes accuracy. (F)

LRFN2 expression and immune cell effector genes. (G) Correlation between LRFN2 expression and different therapeutic targets. (H) Correlation between LRFN2 expression and molecular subtypes in GSE52329.

Figure S17: Stimulation experiments and key signaling cascades related to Figure 2. Proteins expression level of cytokine/chemokines in T24-shNC, T24-shLRFN2-1, T24-shLRFN2-2 cell lines after stimulated with IFN-γ (A) and TNF-α (B). qRT-PCR detected CCL2, CCL3, CCL4, CCL5, CXCL9 and CXCL10 RNA express levels after stimulated with IFN-γ (C) and TNF-α (D). (E) Key molecules within the NF-κB signaling pathway in dependence of the LRFN2 expression level. ns, P>0.05, **P<0.01, ***P<0.001.

Figure S18: T24 in vitro studies. (A) Flow chart of the study. (B-C) Line chart showed the cytotoxicity of T cells (B) and remained tumor cell numbers (C) after co-culture with T24-shNC and T24-shLRFN2 cells for 12 h. (D-E) Schematic diagram (Created with BioRender.com) (D) and histogram (E) of relative migration index of activated $CD8^+$ T cells between T24-shLRFN2 and T24-shNC. (F) KEGG enrichment pathway of RNA sequence between T24-shLRFN2 and T24-shNC. *P<0.05, **P<0.01.

Figure S19: Additional multicolor fluorescence results related to Figure 3. The multicolor fluorescence merge plots of five patients with the inflamed subtype (A) and five patients with the non-inflamed subtype(B).

Figure S20: In situ experiment related to Figure 5. (A-B) qRT-PCR (A) and Western Blot (B) of LRFN2 knock down in mouse bladder cancer cell line MB49. (C) Flow chart of the study. (D) The luciferase imaging of the tumor-bearing mice at Day 18. (E) Photon flux of panel D. (F) Representative flow plots and histograms showed the infiltration of $CD8^+$ TILs and $CD4^+$ TILs within the live $CD45^+$ TILs between MB49-shLRFN2 and MB49-shNC groups. (G-H) TCF-1 (G) and PD-1 (H) expression within the live $CD45^+$ $CD8^+$ TILs between MB49-shLRFN2 and MB49-shNC groups. ns, P>0.05, *P<0.05, **P<0.01, ***P<0.001, ****P<0.0001.

Figure S21: *In vivo* tumor-bearing experiments using two shRNA-knockdown tumor cell lines. (A) Flow chart of the study. (B) Tumor growth curve among different groups. (C-D) Tumor images (C) and weight (D) of individual groups. ns, P>0.05, **P<0.01, ***P<0.001.

Figure S22: Gating strategies of flow cytometry. (A) Gating strategy of mouse T cells co-cultured with MB49-NC and MB49-shLRFN2. (B) Gating strategy of mouse in vivo studies.

Figure S23: Additional multicolor fluorescence results related to Figure 7. The multicolor fluorescence merge plots of five patients in the Response group (A) and five patients in the Resistance group (B).

Figure S24: Schematic diagram of the study.