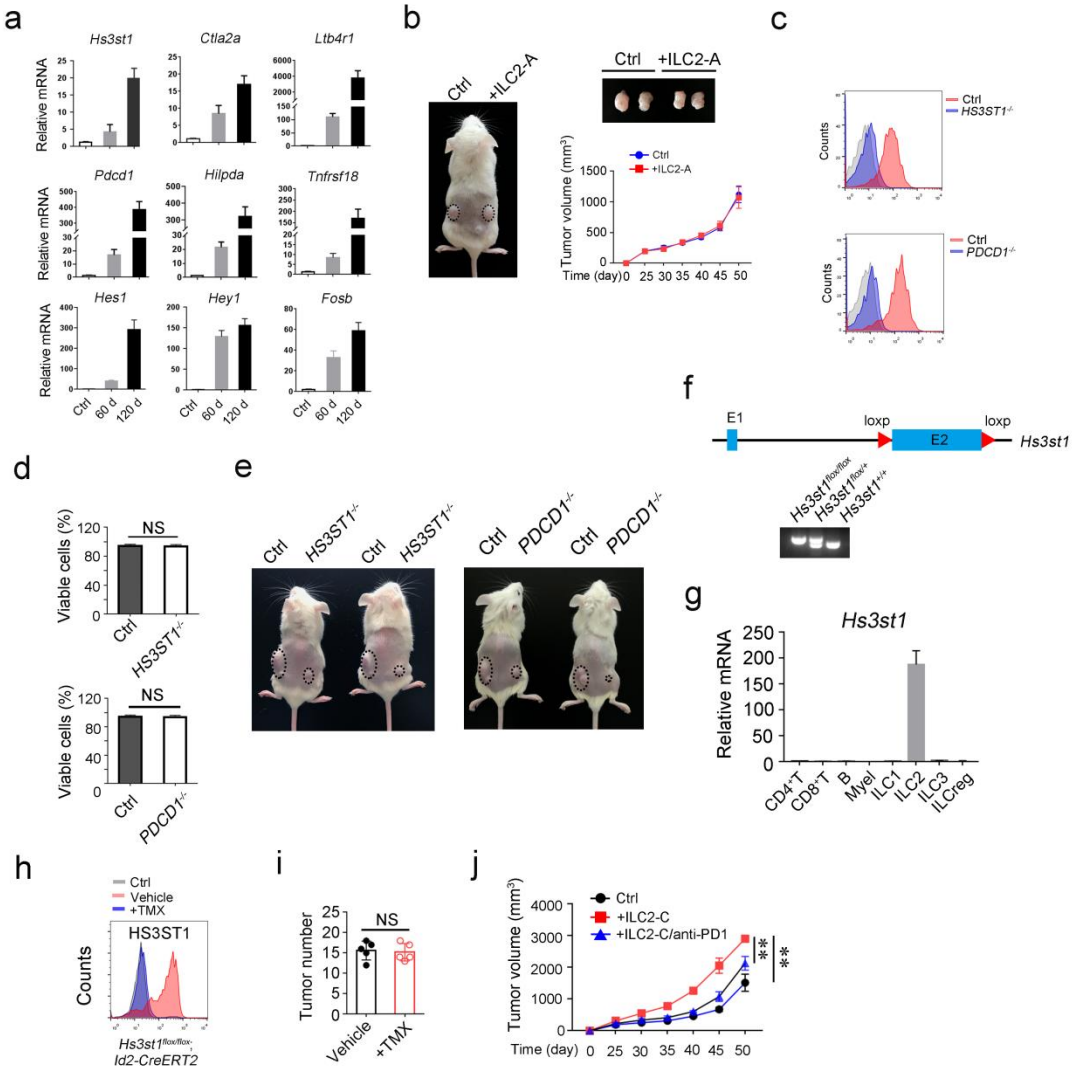
Supplementary information Fig.S3



Supplementary information Fig.S3. ILC2-C cells promote colon tumor progression. (a) Signature genes of tumor infiltrating ILC2s were analyzed by RT-PCR and shown as means±SD. (b) ILC2-A cells have no effect on tumor growth.1×10^5 ILC2-A cells ($PD1^{low}$) (ILC2-A=$Lin^-CD45^+CD127^+CRTH2^+PD1^{low}$) were isolated from stage II CRC patient samples and subcutaneously injected into B-NSG mice together with 1×10^6 primary CRC tumor cells. After 50 days, tumor photo was shown. Tumor volumes were measured at indicated time points and calculated as means±SD. n=6 for each group. (c) ILC2-C cells were isolated from stage IV CRC patient samples by FACS and transfected with virus coding CRISPR/Cas9 elements targeting *HS3ST1* or *PDCD1* gene. After 3 days, expression of HS3ST1 or PD1 in ILC2-C cells was analyzed by flow cytometry. Cells infected with virus bearing empty vector of CRISPR/Cas9 system served as negative control (Ctrl). (d) Ctrl and *HS3ST1*$^{-/-}$ or *PDCD1*$^{-/-}$ ILC2-C cells were isolated and stained with 7-AAD followed by flow cytometry. Percentage of viable cells were calculated and shown as means±SD. NS, no significant by Two-tailed unpaired Student's *t*-test. (e)

Control or *HS3ST1*$^{-/-}$ or *PDCD1*$^{-/-}$ ILC2-C cells together with primary tumor cells from stage IV CRC patient samples were subcutaneously injected into B-NSG mice. After 50 days, photos of tumors on mice were shown. (f) Conditional knockout of *Hs3st1* gene. *Hs3st1* gene containing loxP sites flanking exon 2 of *Hs3st1* was generated by CRISPR/Cas9 technology. Genotyping of successful target sites was shown in the lower panel. (g) mRNA expression levels of *Hs3st1* in indicated cells were analyzed by RT-PCR. Myel, Myeloid cells. (h) *Hs3st1*$^{flox/flox}$*;Id2-CreERT2* mice were subjected to AOM/DSS treatment. At day 60, mice were treated with TMX as shown in Fig. 4h. Tumor infiltrating ILC2s were analyzed for HS3ST1 expression by flow cytometry. (i) WT mice were subjected to AOM/DSS treatment. At day 60, mice were treated with TMX or corn oil (Vehicle) as described in Fig. 4h. At day 120, numbers of colon tumors were calculated and shown as means±SD. NS, not significant by Two-tailed unpaired Student's *t*-test. n=5 per group. (j) ILC2-C cells were isolated from stage IV CRC patient tumors and were subcutaneously injected into B-NSG mice with tumor cells. After 25 days, 1 mg/kg anti-human PD1 antibody was injected into tumors every 1 week. PBS was used as negative control (Ctrl). Tumor volumes were measured at indicated time points and calculated as means±SD. n=5 for each group. **, $P<0.01$ by One Way ANOVA. Data are representative of at least four independent experiments.