Supplementary Figure Legends

Supplementary Figure S1: (A) Processing of tumors and marginal tissues. Work flow of tissue processing for *ex vivo* explant culture and subsequent functional analyses. Ex vivo culture was done for 30 min for NF-\(\kappa\)B, analysis, 24 hours for RNA and 48 hours for ELISA (B) CCL5, CXCL10 and CCL22 mRNA expression in tumors (\(n = 6\) tumors from 6 different patients) was analyzed in freshly-harvested tumor tissue (fresh) or in tumor tissues cultured for 48 hrs in medium alone (untreated) or in the presence of indomethacin + IFN\(\alpha\) + poly-I:C (treated). The treated and untreated tumor samples were harvested simultaneously at 48 hrs of culture. Note that the 48 hr-long cultures did not significantly affect the spontaneous pattern of chemokine production in the (untreated) tumor tissues compared to fresh tumor samples (\(P > 0.05\) for all three chemokines).

Supplementary Figure S2: Presence of T\(_{\text{reg}}\) and T\(_{\text{eff}}\) markers in tumors correlate with intra-tumoral expression of T\(_{\text{eff}}\)- and T\(_{\text{reg}}\)-attracting chemokines. (A) Expression of an alternative CXCR3 ligand, CXCL9, is correlated with local expression of CXCL10 and with T\(_{\text{eff}}\) markers, CD8 and GZMB. (B) Correlation between CCL22 and COX-2 (C) Example: Lack of correlation between CCL22 and CXCL13. (D) Confocal analysis of CD8, CCL5 and CXCL10 expression in colon tumor sections. Expression of CXCR3, CCR5 (E) and Granzyme B (F) in CD8\(^+\) TILs grown from tumor biopsies of 3 different patients. The biopsies were cultured for 2-3 weeks in 1000 units of IL-2 to obtain TILs (See M&M).

Supplementary Figure S3 (A-B): Combination of indomethacin, IFN\(\alpha\) and poly-I:C induces the optimal pattern of chemokine expression in isolated cell cultures. (A) Dose-dependent impact of IFN\(\alpha\) and poly-I:C on the production of T\(_{\text{eff}}\)- and T\(_{\text{reg}}\)-attracting chemokines by *in vitro* generated macrophages (see M&M) and in fibroblasts
(obtained from Cascade Biological). Data from one representative experiment of three.

(B) Effects of indomethacin on T_{eff.} and T_{reg.-attracting} chemokines produced by macrophages (N=3) by ELISA analysis. The concentrations of chemokines in 48 hr cultures were analyzed by ELISA. Note the suppression of CCL22 production by indomethacin. 

**(C-D): Combination of IFN{\alpha}, poly-I:C and cyclooxygenase blockade is needed for the optimal and consistent modulation of chemokine production in metastatic colorectal cancer lesions.** (C) Heterogeneous response of different tumor lesions from the same patient (CCL5 and CXCL10 production) to the individual components of the chemokine-modulating cocktail. (D) Indomethacin and celecoxib enhance the IFN{\alpha}/poly-I:C-induced T_{eff.-attracting} chemokine expression, but suppress T_{reg.-attracting} chemokine expression in colorectal cancer lesions. All cultures were for 48hr. Combined data from the tumors of 3 different patients (n=3).

**Supplementary Figure S4: CCL22 is predominantly expressed by HLA-DR\(^+\) APC, whereas CXCL10 and CCL5 are expressed by both HLA-DR positive and negative cells.** Immunohistochemistry analysis of HLA-DR\(^+\) protein (brown staining) and in-situ hybridization analysis of chemokine mRNA (black silver grains) in tumor tissues. To achieve the optimal levels of expression of each of the three chemokines, CXCL10 and CCL5 expression was analyzed in the treated tissues, while CCL22 expression was in untreated tissues.

**Supplementary Figure S5: Elevated expression of CXCL10 and CCL5 in liver metastases compared to normal liver tissues: role of NF-\(\kappa\)B.** Matched samples of marginal liver tissues and liver-metastatic colorectal cancer tissues (3 biopsies in 1ml in 24 well plate), were cultured for 24hrs either untreated or treated with IFN{\alpha} + poly-I:C + indomethacin and (A) analyzed for CCL5 and CXCL10 expression by Taqman (see matched protein data in Fig 4). (B) Tissues were untreated or treated with IFN{\alpha} + poly-I:C + indomethacin in absence or presence of 20\(\mu\)M CAY10470 (NF-\(\kappa\)B inhibitor). The supernatants were analyzed for CCL5 production by ELISA (see matched CXCL10 data
in Fig 4). (C) CAY10470 (20μM) effects on liver glycogen phosphorylase mRNA expression in matched marginal liver tissues and liver-metastatic colorectal cancer. (D) Example: single and composite images of p65 nuclear translocation in fibroblasts

**Supplementary Figure S6: Tregs are preferentially attracted by untreated tumors.** Negatively-isolated total CD4⁺ T cells were allowed to migrate towards the supernatants from either untreated or treated tumor tissues. Treg migration was analyzed either by (A) flow cytometry analysis of the percentage of FOXP3⁺ cells or (B) Taqman analysis of GITR mRNA in migrated cells.