**Supplementary Figure S1.**

5FU and CPT11 display opposite effects on β-catenin localization in CRC colons. Colons isolated from normal and CRC-mice untreated (CRC) or treated with 5FU or CPT11 were subjected to immunohistological staining with anti-β-catenin antibodies, as described in the methods. A representative section from 4 independent animals analyzed in each experimental group is shown here, original magnification, x200.

**Supplementary Figure S2.**

5FU and CPT11 display opposite effects on MDSC accumulation and CD247 expression in the colons of CRC-mice. (**A** and **B**). Cells were isolated from the lamina propria (left) and epithelium (right) of the colons from each experimental group and analyzed by flow cytometry for MDSC accumulation (**A**) and CD247 expression levels (MFI), gating on CD3+ cells (**B**). Graphs (means of triplicates ± s.e.m., *n*=4) are representative of a typical experiment out of three independent performed. \*, *P*<0.05;\*\*, *P*<0.01; ns = non-significant.

**Supplementary Figure S3.**

5FU or CPT11 chemotherapies do not alter the elevated Treg levels in spleens from CRC-mice. Levels of Tregs from each experimental group were evaluated after fixation/permeabilization and double staining for CD4+Foxp3+. Graph (means of triplicates ± s.e.m., *n*=4) is representative of a typical experiment out of three performed.\*, *P*<0.05.

**Supplementary Figure S4.**

A combined 5FU and CPT11 therapy preserves immunosuppression. (**A** and **B**) MDSCs isolated from spleens of CRC-mice (*n*=5) were cultured *ex vivo* in the presence of 2.5µmol/L of 5FU, CPT11 or 5FU and CPT11 for 24h. Cultured MDSCs were analyzed for NO- and ROS production by flow cytometry analysis gaiting on (**A**) CD11b+Gr1+ MDSCs, (**B**) CD11b+Ly6Chigh Ly6G- monocytic MDSCs (M-MDSCs) and CD11b+Ly6ClowLy6G+ granulocytic MDSCs (G-MDSCs) sub-populations. (**C**) Splenocytes were labeled with CFSE and activated with anti-CD3 and anti-CD28 antibodies or left non-activated. The proliferative response was assessed by monitoring cell divisions of gated CFSE-labeled CD8+ T-cells. Representative histograms of proliferative activity are shown (left), and the percent of proliferating cells was calculated and compared to steady-state levels of non-activated cells in each group (right). (D) Splenocytes from the experimental groups were analyzed for CD247 expression levels gating on CD8+ cells and indicated by MFI. Graphs (means of triplicates ± s.e.m., *n*=5) are representative of a typical experiment out of three independent performed. \*, *P*<0.05; \*\*, *P*<0.01; \*\*\*, *P*<0.001; non-significant.

**Supplementary Figure S5.**

A schematic representation of mouse model for CRC in which MDSCs were *in vivo* depleted in CPT11 treated mice by Gr1 mAb administration every 3 days.

**Supplementary Figure S6.**

5FU and CPT11 differently affect monocytic/granulocytic MDSC sensitivity to apoptosis. (**A**) MDSCs isolated from spleens of CRC-mice (*n*=5) were cultured *ex vivo* in the presence of 2.5µmol/L of 5FU, CPT11 or 5FU and CPT11 for 3 days. Cultured MDSCs were analyzed for cleaved caspase-3 expression (MFI) by flow cytometry analysis gaiting on CD11b+Ly6Chigh Ly6G- monocytic MDSCs (M-MDSCs) (left) and CD11b+Ly6ClowLy6G+ granulocytic MDSCs (G-MDSCs) (right) sub-populations. (**B**) 5FU or CPT11 treatments do not alter cleaved caspase-3 expression in T (CD3+) (left)- and B (B220+) (right)-lymphocytes. Splenic T- and B- cells from each group were analyzed for the expression of activated (cleaved) caspase-3 by flow cytometry analysis. All *ex vivo* experiments involved 5 and *in vivo* experiments 6 mice per group and were repeated three times yielding similar results. Graphs (means of triplicates ± s.e.m., *n*=6) are representative of a typical experiment out of three performed.

**Supplementary Figure S7.**

5FU and CPT11 treatments display opposite effects on mRNA expression levels of pro-inflammatory molecules. (**A**) mRNA levels of S100A8/9 in the colon of CRC-mice treated with 5FU or CPT11 relative to the expression in untreated CRC-mice were evaluated by Real Time PCR analysis. (**B-C**) To assess the effect of 5FU and CPT11 on pro-inflammatory molecules generated by MDSCs, MDSCs isolated from spleens of CRC-mice (*n*=4) were cultured *ex vivo* in the presence of scaled-doses (0, 1.25, 2.5, 5 and 10µmol/L) of 5FU or CPT11 for 3days. TNFα (**B**) and S100A9 (**C**) mRNA levels were then evaluated by Real Time PCR analysis performed on the primary MDSCs. Graphs (means of triplicates ± s.e.m., *n*=3) are representative of a typical experiment out of three independent performed. \*, P<0.05; \*\*, *P*<0.01.

**Supplementary Figure S8.**

5FU and CPT11 treatments differently affect the immunological profile of mice exhibiting a pathology free chronic inflammation. MDSC accumulation in the spleen was evaluated by flow cytometry analysis. Graph represents the absolute number of MDSCs within the spleen of normal, inflamed and inflamed mice treated with 5FU, CPT11 or 5FU/CPT11 (**A**). Tregs derived from the spleens of each experimental group were evaluated by measuring CD4+Foxp3+cells (**B**). T-cell proliferative response was assessed by monitoring cell divisions of gated CFSE-labeled CD8+ T-cells upon TCR-CD28 mediated activation. The percent of proliferating cells was calculated and compared to steady-state levels of non-activated cells in each group (**C**). Splenocytes (left) and PBLs (right) from each experimental group were stained for CD247 (**D**). CD247 expression levels were measured in gated CD3+ cells and are presented as the expression in the experimental group relative to normal mice (as 100%). Splenocytes from each group were double stained for CD247 and NCR1. CD247 expression levels were measured in NK (NCR1+) cells and are presented as the expression in the experimental group relative to normal mice (as 100%) (**E**). Graph (means of triplicates ± s.e.m., *n*=4) is representative of a typical experiment out of three independent performed. \*, *P*<0.05; \*\*, *P*<0.01; \*\*\*, *P*<0.001; non-significant.