

Supplementary Figures

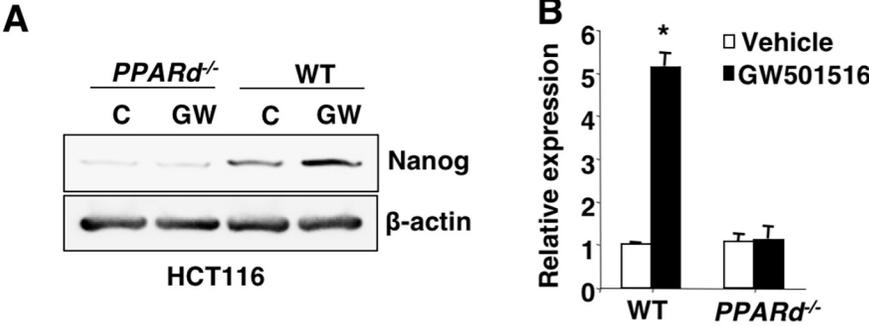


Figure S1. Activation of PPAR δ induces Nanog expression

(A-B) HCT-116/WT and HCT-116/*PPARd^{-/-}* cells were treated with vehicle or GW501516. After treatment, Nanog expression at protein (A) and mRNA levels (B) were measured as described in the Method. The error bar indicates \pm SEM. *p<0.05.

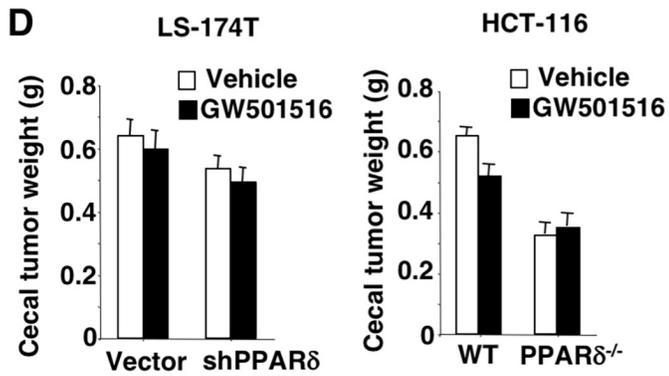
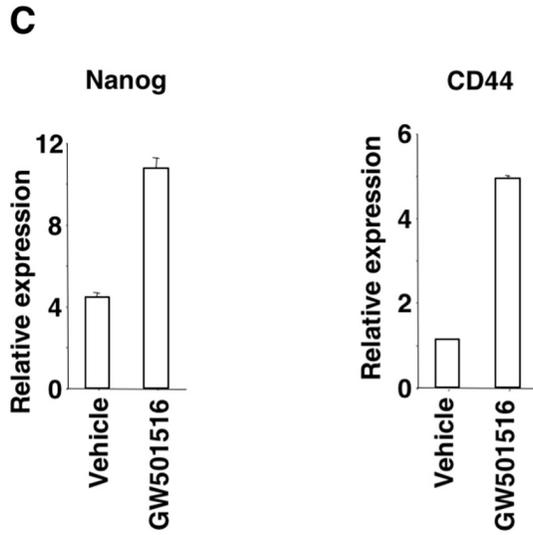
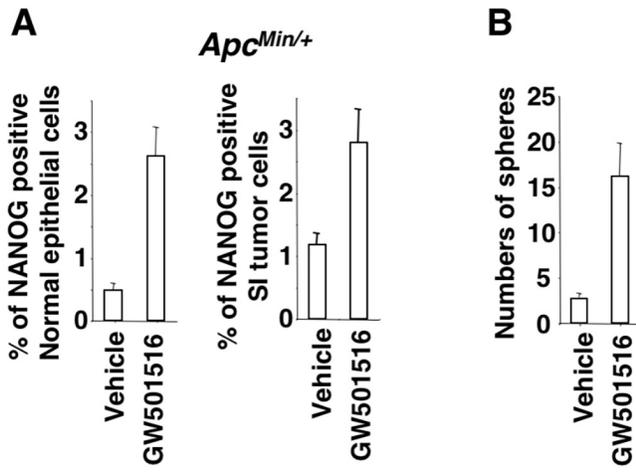


Figure S2. Activation of PPAR δ induces intestinal CSCs by inducing Nanog expression

(A) The *Apc*^{Min/+} mice were fed with vehicle or GW501516. After treatment, cells isolated from normal tissues and intestinal tumors of indicated group of mice were subjected into flow cytometry analysis. For flow cytometry analysis, data represents the percentage of EpCAM⁺CD45⁻Nanog⁺ cells in total intestinal epithelial cells. (B) Purified epithelial tumor cells by flow cytometry sorting were subjected to *in vitro* sphere-forming assays. (C) The levels of Nanog and CD44 expression at mRNA levels in purified epithelial tumor cells of indicated group of mice. (D) The NSG mice were treated with vehicle or GW501516 after LS-174T/vector, LS-174T/shPPAR δ , HCT-116/WT, or HCT-116/PPAR δ ^{-/-} cells were injected into cecum. After treatment, the weight of cecal tumor taken from indicated group of mice as described in Figs. 2-3. The error bar indicates \pm SEM. *p<0.05.

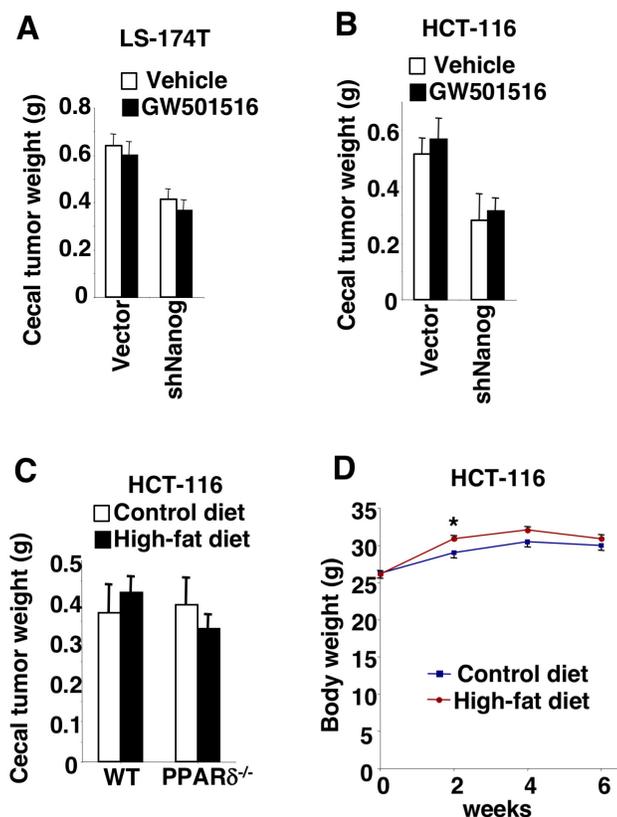


Figure S3. GW501516 and a high-fat diet did not affect cecal tumor growth

(A-B) The NSG mice were treated with vehicle or GW501516 after LS-174T/vector, LS-174T/shNanog (A), HCT-116/vector, or HCT-116/shNanog cells (B) were injected into cecum. After treatment, the weight of cecal tumor taken from indicated group of mice as described in Fig. 4. (C-D) The NSG mice were treated with a control diet or a high-fat diet after HCT-116/WT or HCT-116/*PPAR $\delta^{-/-}$* cells were injected into cecum. After treatment, the weight of cecal tumor (C) and body weight (D) of indicated group of mice as described in Fig. 5. The error bar indicates \pm SEM. * $p < 0.05$.

Supplementary Tables

Supplementary Table S1. Tumorigenicity of CRC cells treated with vehicle or GW501516

	No. of injected cells	Number of mice with tumors/total injected mice (% of mice with tumors)			
		21 days	28 days	34 days	44 days
LS-174T					
Vehicle	1,000	2/7 (29)	2/6 (33)	5/6 (83)	6/6 (100)
	200	0/8 (0)	2/8 (25)	4/8 (50)	8/8 (100)
	50	0/8 (0)	0/8 (0)	1/8 (12.5)	2/8 (25)
GW501516	1,000	3/8 (50)	5/6 (83)	6/6 (100)	6/6 (100)
	200	2/8 (25)	5/8 (63)	7/8 (88)	8/8 (100)
	50	0/8 (0)	1/8 (13)	4/8 (50)	7/8 (88)
HCT-116					
Vehicle	2,500	1/6 (16.7)	4/6 (66.7)	6/6 (100)	6/6 (100)
	500	1/8 (12.5)	4/8 (50)	5/8 (62.5)	8/8 (100)
	100	0/8 (0)	1/8 (12.5)	2/8 (25)	2/8 (25)
GW501516	2,500	3/6 (50)	6/6 (100)	6/6 (100)	6/6 (100)
	500	2/8 (25)	6/8 (75)	8/8 (100)	8/8 (100)
	100	0/8 (0)	4/8 (50)	6/8 (75)	7/8 (88)

Supplementary Table S1. LS-174T and HCT-116 cells were treated with vehicle or GW501516. After treatment, these cells were subjected to limiting dilution transplantation assays.

Supplementary Table S2. Tumorigenicity of cecal tumor cells from mice treated with a control diet or a high-fat diet

HCT-116	No. of injected cells	Number of mice with tumors/total injected mice			
		(% of mice with tumors)			
		26 days	30 days	35 days	42 days
Vehicle	1x10 ⁴	1/8 (12.5)	1/8 (12.5)	3/8 (37.5)	4/8 (50)
	2,500	0/8 (0)	0/8 (0)	0/8 (0)	1/8 (12.5)
	500	0/8 (0)	0/8 (0)	0/8 (0)	0/8 (0)
High-fat	1x10 ⁴	5/8 (62.5)	8/8 (100)	8/8 (100)	8/8 (100)
	2,500	1/8 (12.5)	2/8 (25)	6/8 (75)	8/8 (100)
	500	0/8 (0)	0/8 (0)	1/8 (12.5)	3/8 (37.5)

Supplementary Table S2. The NSG mice were treated with a control diet or a high-fat diet after HCT-116 cells were injected into cecum. Tumor epithelial cells isolated from cecal tumor cells of indicated group of mice were subjected to limiting dilution transplantation assays.

Supplementary Table S3. Tumorigenicity of cecal tumor cells from mice treated with a control diet or a high-fat diet

LS-174T	No. of injected cells	Number of mice with tumors/total injected mice (% of mice with tumors)			
		26 days	33 days	37 days	44 days
Vehicle	5,000	0/6 (0)	2/6 (33.3)	4/6 (66.7)	5/6 (83.3)
	1,000	0/8 (0)	0/8 (0)	3/8 (37.5)	4/8 (50)
	200	0/8 (0)	0/8 (0)	0/8 (0)	1/8 (12.5)
High-fat	5,000	3/6 (50)	5/6 (83.3)	6/6 (100)	6/6 (100)
	1,000	0/8 (0)	4/8 (50)	7/8 (88)	8/8 (100)
	200	0/8 (0)	0/8 (0)	3/8 (37.5)	5/8 (62.5)

Supplementary Table S3. The NSG mice were treated with a control diet or a high-fat diet after LS-174T cells were injected into cecum. Tumor epithelial cells isolated from cecal tumor cells of indicated group of mice were subjected to limiting dilution transplantation assays.