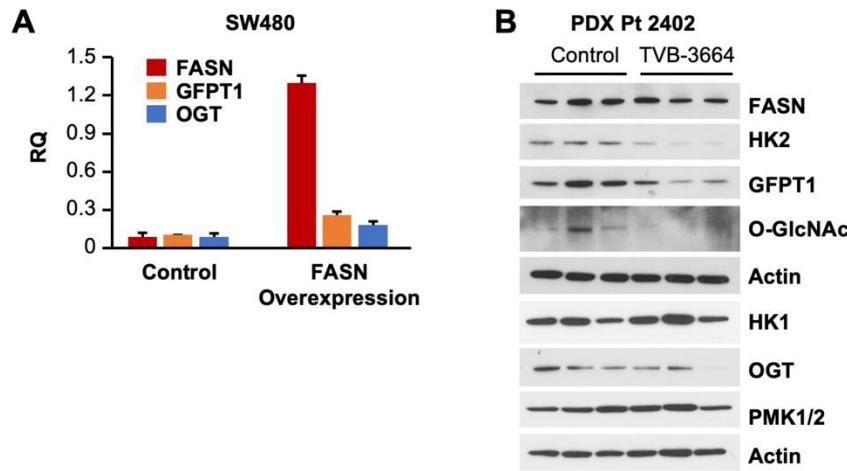
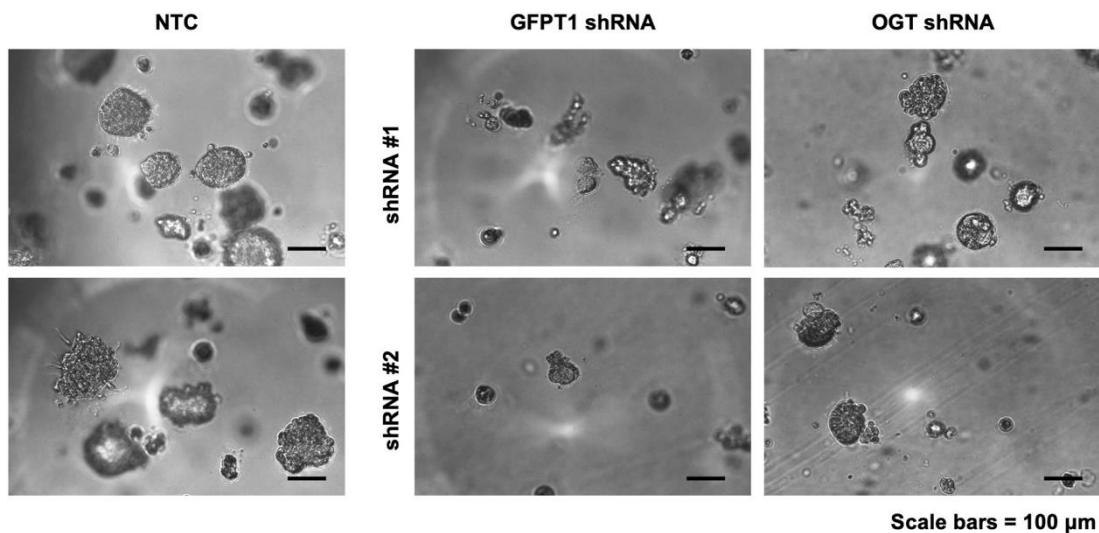


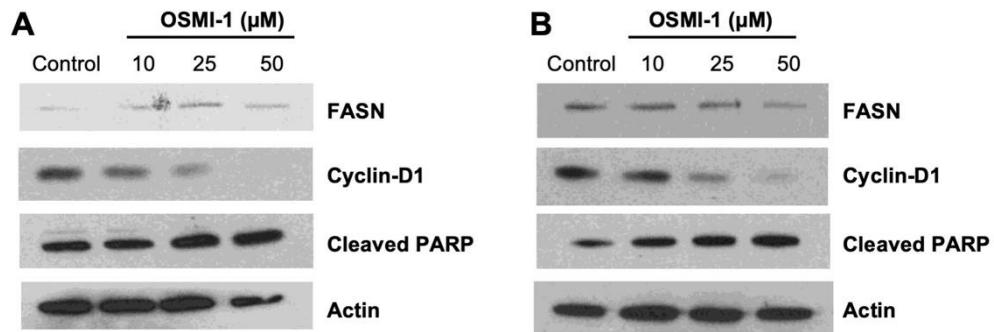
**Supplemental Figure S1. ERT2-mediated deletion of FASN decreases expression of OGT and GFPT1.**  
**(A)** IHC staining of *Apc*/Cre intestinal adenoma tissues for FASN, GFPT1 and OGT. Protein **(B)** and mRNA **(C)** expression of FASN, GFPT1 and OGT in intestinal tissues in *Apc*/VillinCreERT2 and *Apc*/*Fasn*<sup>+/−</sup>/VillinCreERT2 mice.



**Supplemental Figure S2.** FASN regulates expression of GFPT1 and OGT in human CRC cells. **(A)** qRT-PCR showing that overexpression of FASN upregulates GFPT1 and OGT mRNA levels. **(B)** Pharmacological inhibition of FASN using TVB-3664 decreases the expression of GFPT1 and OGT and the level of O-GlcNAc in the PDX mouse model.



**Supplemental Figure S3.** Representative images of HCT116, NTC, GFPT1 shRNA and OGT shRNA, cells grown in Matrigel for 10 days.



**Supplemental Figure S4. Effects of O-GlcNAc transferase inhibitor on CRC cells.** Western blot showing dose-dependent inhibition of cyclin D and induction of PARP cleavage in Pt130 (A) and Pt93 (B) cell lines by OSMI-1.

**Supplemental Table S1.** Summary of correlations between FASN mRNA level and HK2, GFPT1 and OGT mRNA levels using data from five different publicly available databases.

Data set	Gene pair	Probe use	P value	Spearman Coefficient ( $\rho$ )	Correlation
TCGA-COAD	FASN vs HK2	-	0.324899608	1.13723E-12	Significant
Bittner_Colon	FASN vs HK2	Maximum	0.25718059	2.20077E-06	Significant
Bittner_Colon	FASN vs HK2	Average	0.226399107	3.3079E-05	Significant
Jorissen_Colorectal_3	FASN vs HK2	Maximum	0.203873119	0.011210113	Significant
Jorissen_Colorectal_3	FASN vs HK2	Average	0.209525452	0.009108027	Significant
Kaiser_Colon	FASN vs HK2	Maximum	0.35548088	0.000284065	Significant
Kaiser_Colon	FASN vs HK2	Average	0.393497711	5.11824E-05	Significant
Ki_Colon	FASN vs HK2	Maximum	0.102808071	0.463832084	Insignificant
Ki_Colon	FASN vs HK2	Average	0.015280102	0.913524096	Insignificant
TCGA-COAD	FASN vs GFPT1	-	-0.003472998	0.941042518	Insignificant
Bittner_Colon	FASN vs GFPT1	Maximum	0.417345681	2.43541E-15	Significant
Bittner_Colon	FASN vs GFPT1	Average	0.222043779	4.71686E-05	Significant
Jorissen_Colorectal_3	FASN vs GFPT1	Maximum	0.213324856	0.00789863	Significant
Jorissen_Colorectal_3	FASN vs GFPT1	Average	0.1567461	0.052217361	Insignificant
Kaiser_Colon	FASN vs GFPT1	Maximum	0.321671062	0.001100603	Significant
Kaiser_Colon	FASN vs GFPT1	Average	0.438814514	4.95166E-06	Significant
Ki_Colon	FASN vs GFPT1	Maximum	0.333588679	0.014645743	Significant
Ki_Colon	FASN vs GFPT1	Average	0.333588679	0.014645743	Significant
TCGA-COAD	FASN vs OGT	-	0.025906755	0.581091482	Insignificant
Bittner_Colon	FASN vs OGT	Maximum	0.383606995	5.19062E-13	Significant
Bittner_Colon	FASN vs OGT	Average	0.128322574	0.019706026	Significant
Jorissen_Colorectal_3	FASN vs OGT	Maximum	-0.147712599	0.067523682	Insignificant
Jorissen_Colorectal_3	FASN vs OGT	Average	0.043321258	0.593706068	Insignificant
Kaiser_Colon	FASN vs OGT	Maximum	0.246299802	0.013503282	Significant
Kaiser_Colon	FASN vs OGT	Average	0.094174265	0.351348935	Insignificant
Ki_Colon	FASN vs OGT	Maximum	0.08208519	0.559000311	Insignificant
Ki_Colon	FASN vs OGT	Average	0.08208519	0.559000311	Insignificant