Supplemental Information

Cooperation between SMYD3 and PC4 drives a distinct transcriptional program in cancer cells

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Supplementary Figure Legends

Figure S1. Effects of SMYD3 knockdown on proliferation and invasion of bladder and colon cancer cells.
(A) Whole cell extracts were prepared from colon normal (CDC-18Co) and cancer (HCT116, CaCO2, and HT29) cells, bladder normal (UROtsa) and cancer (J82, T24 and RT4) cells, breast normal (MCF-10-2A) and cancer (MCF7 and MDA-MB-231) cells, and prostate normal (MLC) and cancer (LNCaP and DU145) cells. Equal amounts of extracts were analyzed by immunoblotting with anti-SMYD3 and anti-Actin antibodies. (B) HCT116, CaCO2, J82 and T24 cancer cells were transfected with lentiviruses expressing control shRNA (Control sh) or SMYD3-specific shRNA (SMYD3 sh), and knockdown efficiency was determined by immunoblotting. (C) HCT116, CaCO2, J82 and T24 cancer cells were transfected with control shRNA or SMYD3 shRNA, and their proliferation rates were determined by MTT colorimetric assays over a period of 4 days. Results represent the means of three independent experiments ± SD. (D) Control or SMYD3-depleted HCT116, CaCO2, J82 and T24 cancer cells were detached and seeded onto the upper chamber coated with Matrigel, and then allowed to invade toward 10% FBS in the lower chamber. The graph depicts the average number of invaded cells per four fields. Results represent the means ± SD of three independent experiments.

Figure S2. Validation of microarray data.
Microarray data were validated by qRT-PCR of seventeen genes whose expression was decreased upon SMYD3 knockdown and which are related to cell growth and invasion, and two unaffected genes. Primer sequences are listed in Table S3. The values are expressed as fold changes from the mRNA levels in control cells after normalization with ACTB. Data represent the means ± SD of three independent experiments.

Figure S3. Target gene occupancy of SMYD3 and H3K4me3.
(A) SMYD3-depleted HCT116 colon cancer cells were infected with lentiviruses expressing RNAi-resistant FLAG-SMYD3 wild-type (wt) or enzymatic dead mutant (F183A), and whole cell extracts were analyzed by immunoblotting with the indicated antibodies. (B) ChIP-qPCR experiments were performed as in Figure 1D, but using primers specific for the MFGE8 gene. The vertical red bar represents the putative SMYD3 binding site.

Figure S4. Effects of SMYD3 knockdown on target gene expression.
Expression levels of the genes down-regulated (FNBP1, MFGE8, PDLIM7, and WNT3A) or unaffected (KRT81 and GAPDH) were analyzed by qRT-PCR on total RNA isolated from SMYD3-depleted CaCO2, J82 and T24 cancer cells.

Figure S5. Effects of SMYD3 knockdown on H3K4me3.
(A) Potential SMYD3 binding sites in target gene promoters are indicated in red. The nucleotides in blue indicate primer sequences used for ChIP-qPCR. (B) Diagram depicting the positions of the PCR amplicons used for ChIP assays. The numbers represent the start and end positions of amplicons relative to the transcription start site (TSS). The putative SMYD3 binding sites are shown as vertical red bars. (C and D) ChIP experiments determining SMYD3 localization (C) and H3K4me3 enrichment (D) at the four SMYD3 target genes (FNBP1, MFGE8, PDLIM7,
and two control genes (KRT81 and GAPDH) in SMYD3-depleted HCT116, CaCO2, J82 and T24 cancer cells using primers depicted in (B).

**Figure S6. Interaction of SMYD3 N-terminal deletion mutants with PC4.**
GST alone or GST-PC4, immobilized on glutathione Sepharose beads, was incubated with His-tagged SMYD3 deletion mutants. After washing with washing buffer, the bound SMYD3 proteins were immunoblotted with His antibody. Of the input proteins, 5% were examined by immunoblotting.

**Figure S7. Circular dichroism spectra of SMYD3 and PC4.**
Spectra of SMYD3 (black) and PC4 (red) are shown. In addition, the spectra of the SMYD3-PC4 complex is depicted in green, which matches the sum of the individual SMYD3 and PC4 spectra, shown in blue. Spectra were recorded at protein concentrations of 5 µM in 5 mM K2HPO4/KH2PO4, pH 7.4, 25 mM KCl at 25 °C using a JASCO J-810 spectropolarimeter. The observed ellipticity in millidegrees, \( \theta \), was converted into the mean residue ellipticity, \( \theta_{MRW} \).

**Figure S8. Interdependent localization of SMYD3 and PC4 at target genes.**
(A) SMYD3-depleted HCT116 colon cancer cells were infected with lentiviruses expressing FLAG-tagged SMYD3 wild-type (wt) or K78/D82/R85 mutant (mt) lacking interaction with PC4, and cell lysates were prepared and analyzed by immunoblotting with anti-SMYD3, anti-FLAG, anti-PC4, and anti-Actin antibodies, as indicated. (B) ChIP-qPCR experiments were performed as in Figure 3C, but using primers specific for the MFGE8 gene. (C) Exogenous expression of FLAG-tagged PC4 wild-type (wt) or SMYD interaction-deficient Q65/R75 mutant (mt) in PC4-depleted HCT116 colon cancer cells was confirmed by immunoblotting. (D) ChIP-qPCR assays were done with PC4-depleted HCT116 cells complemented with PC4 wild-type (wt) or mutant (mt) as in (B). The vertical red bar in (B) and (D) represents the putative SMYD3 binding site.

**Figure S9. RT-PCR and ChIP of the PC4 target gene SMUG1.**
(A) SMYD3-depleted HCT116 cells were transfected with SMYD3 wild-type (wt) or K78/D82/R85 mutant (mt), and the expression of the known PC4 target gene SMUG1 was determined by qRT-PCR. (B) SMYD3-depleted HCT116 cells were complemented with SMYD3 wild-type (wt) or K78/D82/R85 mutant (mt) as in (A), and the levels of SMYD3, PC4 and H3K4me3 along the SMUG1 gene were determined by ChIP-qPCR. (C) Wild type (wt) or Q65/R75-mutated (mt) PC4 was expressed in PC4-depleted HCT116 cells, and SMUG1 gene expression was determined by qRT-PCR. (D) ChIP signals for SMYD3, PC4 and H3K4me3 along the SMUG1 gene were determined as indicated.

**Figure S10. dCas9-based activation of SMYD3 target genes.**
qRT-PCR experiments were performed as in Figure 5C, but using sgRNAs specific for the MFGE8 gene.

**Figure S11. dCas9-based accumulation of SMYD3 and H3K4me3 at target genes.**
ChIP-qPCR experiments were performed as in Figure 6, but using sgRNAs and primers specific for the MFGE8 gene.
### Supplementary Table S2. sgRNA target sites and oligo primer sequences.

| Name   | Target site            | Forward (5'-3')          | Reverse (5'-3')          |
|--------|------------------------|--------------------------|--------------------------|
| FNBP1-G1 | CCCGGGGGAGGGGGCCCCGGCC | AGGGGAGGGGGCCCCGGCC      | AAAACGCCGGCCCCCTCCCCCCTC |
|         | G1                     | ACACCCCCGGGGGAGGGGCCCCG  | CCCGGGGGCCCCCTCCCCCCTCC  |
| FNBP1-G2 | CCGCCCCCTCCCCCGCTCTCC | AGGGGAGGGGGCCCCCTCCCCCCTCC | AAAACGGAGAGGGGGCCCAGGCGGG |
|         | G2                     | ACACCCCCGGGGGCCCCCTCCCCCTCC | AAAACGGGAGGCCCCGCCCCAGG |
| FNBP1-G3 | CCGAGCTCTGGGGGGCTCTGGGC | AAAACCGGGCTGGGAGGGAATTGGTTG |
|         | G3                     | AAAACCTACACTACCCAGAGCTCGGG |
| FNBP1-G4 | CTCGCGCGCTGGGGCTCTTCGG | AAAACCTACACTACCCAGAGCTCGGG |
|         | G4                     | AAAACCTACACTACCCAGAGCTCGGG |
| MFGE8-G1 | ACCAAATTCCCTCCCCAGGCC | ACCAAACCCGCCCCCTCCCTCCCAGG |
|         | G1                     | ACACCCCCGGGGGAGGGGCCCCG  | AAAACCTACACTACCCAGAGCTCGGG |
| MFGE8-G2 | GCCAGCTTGGGCGAGCCGCGCA | AAAACCTACACTACCCAGAGCTCGGG |
|         | G2                     | AAAACCTACACTACCCAGAGCTCGGG |
| MFGE8-G3 | TAAGGGGAAGGGCGGAGGAGT  | AAAACCTACACTACCCAGAGCTCGGG |
|         | G3                     | AAAACCTACACTACCCAGAGCTCGGG |
| MFGE8-G4 | GGAAGAGGGCTTGGTGGTGGGT | AAAACCTACACTACCCAGAGCTCGGG |
|         | G4                     | AAAACCTACACTACCCAGAGCTCGGG |
**Supplementary Table S3.** Primer sequences for qRT-PCR.

| Primers | Forward (5’-3’) | Reverse (5’-3’) |
|---------|----------------|----------------|
| ACTB    | TCACCGAGCGCGGCT | TAATGTCAACGCACGATTTCCC |
| ARAF    | GAATGAGATGCAGGTGCTCA | CCACATGCAGGTGATGGTAG |
| GRINA   | CGGGAGAATGTCTGGACCTA | CTGCATGGAGAAGATGACGA |
| MAP2K3  | AGCTACCTGGAGCTGATGGA | AGCTTATGGTGTTGGGTGAGC |
| MAPK3   | ACAGTCTCTGCCCCCTCCAAGA | CTCATCCGTGGGTGCTAGT |
| P4HB    | CATCGTGAACTGGCTGAAGA | CTCCACGTCTTGAAGACGC |
| PLCG2   | AAGAACCAGCAGAAACCAAG | TTTCTCCTTTGGGTGAGACG |
| DERL1   | GCTTTGCAGCTGGAGACCAG | TGGTAGCCAGAAGCGCATGT |
| FNBP1   | TGGAGGAAAGGAGGATTGTG | CGCTTCATGGCTGATGT |
| FTL     | AGGCCCTTTTGGATCTTCTCAT | CAGGTGGTCACCCCATTTCT |
| GAL     | AAGGAAAAACGAGGCTGGAC | GGACCTGTCAAAGCTCCTG |
| MFGE8   | ACCTGTGGAGACCCCTGCTG | GGTTCAGCCTGAAAGATGC |
| PDLIM7  | CAGCCTTCCACCTACCTCACC | AAAGAGCAAGGGCAGTGTCA |
| SLC9A1  | TCTTCACCTCTCTTGATCAG | AAGGTGGTCCAGGAACGTGT |
| SNCA    | TGTGGGCCCAGCTGACATTT | CCACAAATCCACACGAAC |
| TOB1    | ACAGCCCCCTTAACCTCAGT | GCCCGTGACACCTATTGT |
| VGF     | AGCCTCCTCTCTTCCTAGCT | GAAAAGCTTCCTCCCTCCTAG |
| WNT3A   | CAAGATTGGGCATCCAGGAGT | ATGACGCCTGTCACCTGAAAG |
| KRT81   | TAGGCACCCCAACTCAAGTC | AAGTGCGGGGATCACACAGAG |
| GAPDH   | GGCCTCAAGGAGAATGAC | AGGGGAGATTCAGTGTG |
| SMUG1   | AGCTGGGGGAAAGGATGTAGT | GCCAACATGGTGAACACTG |
### Supplementary Table S4. Primer sequences for qPCR.

| Primers        | Forward (5’-3’)       | Reverse (5’-3’)       |
|----------------|-----------------------|-----------------------|
| PDLIM7-TSS     | GAAAGTCTAGTGGGCGTGGT  | AGTGCCCTTTTGTGTCCTC   |
| WNT3A-TSS      | CACAGACCAGGAGCGAGAG   | GTGTCGGTGTTGCTGAGGAC  |
| KRT81-TSS      | GCCAGGCTCTACTCCCTCT   | GGGGAGTCAGCATGTTCAT   |
| FNPB1-A        | GCTGTCCTCCCTCTCTCT    | TTTGAAAACCGTACATGCA   |
| FNPB1-B        | GGGGAAAGGGCTAGTGATTC  | CAGGCTCTATTATATCAGCA  |
| FNPB1-C        | TACTGCGAGCAGACAGTGGT  | CAAATGGCCCGAGGAAG     |
| FNPB1-D        | CGAGCTCTGGGTGAGTGAG   | TGCCTCCGAAGGACAAAG    |
| FNPB1-E        | CAAAGGATGTTGAGAGGAGAA| TATGCCCCTCAGGATTTGG   |
| FNPB1-F        | GTGGGTCAGGCCTATATTC   | TCCAGATATTCCTCCAAGCTG|
| MFGE8-A        | CCACCATGCTCTTACTTTT   | GAAGGGAGCTTCTCAAACC   |
| MFGE8-B        | GCACCCAGCCAAACACTAT   | CTGTTTGGCCGAAAGGAATA  |
| MFGE8-C        | CCAACAGACTCAAGACTCG   | CCTTGGGACCGGAATAAAT   |
| MFGE8-D        | GGCTGGACAGTTCTCCATTG  | GTCAGGAGACTCCCACAGC   |
| MFGE8-E        | CCAAGCTCTCGTCTGCTGCA  | GGGGCTGCGGAGCTTAAGT   |
| MFGE8-F        | CCCACTCCCATCTCAACCT   | TGGGAGGATCTGAAGAATG   |
| SMUG1-A        | GGCTGTAGGAGTGGATGAT   | GGTGCAGTAGGAGCTTCAGG  |
| SMUG1-B        | GGCTCTGGGACTTAGCAGTG  | TCTCCCCGGCTTGCTGTAT   |
| SMUG1-C        | ACAGGCTGTGGCTTTTCACT  | GCTAAGGACAGTGGGAGTCG  |
| SMUG1-D        | CACCAGTGGGACTATAATT   | GACCTCGAGCCAGCGCTGAC  |
| SMUG1-E        | ATGTCAAAGGTTGCGCTCAA  | GGGGAAGTGGGAGGAAGAAG  |
| SMUG1-F        | ACGGGACCTACTTGGGTTGG  | CCCAACCTGTCACATATCC   |
**Supplementary Table S5.** GSEA of 9 gene sets significantly enriched in the SMYD3-knock down condition (normalized (NOM) p<0.01 and false discovery rate (FDR) q<0.05).

| NAME                                           | SIZE | ES       | NES      | NOM p-val | FDR q-val |
|------------------------------------------------|------|----------|----------|-----------|-----------|
| BENPORATH PROLIFERATION                        | 78   | 0.423409 | 4.035999 | 0         | 0         |
| POOLA INVASIVE BREAST CANCER UP                | 83   | 0.252012 | 2.556257 | 0         | 3.18E-04  |
| ROSTY CERVICAL CANCER PROLIFERATION CLUSTER   | 84   | 0.544624 | 5.226195 | 0         | 0         |
| CHIANG LIVER CANCER SUBCLASS PROLIFERATION UP  | 85   | 0.397859 | 4.045592 | 0         | 0         |
| PUIFFE INVASION INHIBITED BY ASCITES DN        | 65   | 0.326433 | 2.170377 | 0.005865  | 0.015505  |
| PEART HDAC PROLIFERATION CLUSTER DN           | 40   | 0.352734 | 1.989805 | 0.013921  | 0.020701  |
| PEART HDAC PROLIFERATION CLUSTER UP           | 56   | 0.305071 | 1.880649 | 0.012019  | 0.029155  |
| PUIFFE INVASION INHIBITED BY ASCITES UP       | 35   | 0.296035 | 1.724907 | 0.010392  | 0.036908  |
| WANG TUMOR INVASIVENESS UP                    | 147  | 0.195778 | 1.674531 | 0.014035  | 0.04197   |
**Supplementary Table S6.** Molecular and cellular function analysis of IPA in SMYD3 knockdown repressed genes.

| Name                                      | Number of genes | p-value          |
|--------------------------------------------|-----------------|------------------|
| Cell death and Survival                    | 77              | 5.98E-06 - 1.27E-02 |
| Cellular Growth and Proliferation          | 72              | 4.45E-04 - 1.23E-02 |
| Cellular Function and Maintenance          | 71              | 1.03E-04 - 1.37E-02 |
| Cellular Assembly and Organization         | 54              | 1.50E-04 - 1.41E-02 |
| Cellular Development                       | 51              | 4.45E-04 - 1.23E-02 |
| Molecular Transport                        | 50              | 2.79E-04 - 1.41E-02 |
| Small Molecule Biochemistry                | 41              | 3.54E-05 - 1.41E-02 |
| Cell-To-Cell Signaling and Interaction     | 31              | 4.45E-04 - 1.41E-02 |
| Cellular Compromise                        | 26              | 1.03E-04 - 1.41E-02 |
| Carbohydrate Metabolism                    | 23              | 9.58E-05 - 1.23E-02 |
| Post-Translational Modification            | 14              | 3.54E-05 - 1.23E-02 |
| Protein Trafficking                        | 10              | 2.79E-04 - 1.23E-02 |
| Amino Acid Metabolism                      | 5               | 3.54E-05 - 1.23E-02 |
**Supplementary Table S7.** Molecular and cellular function analysis of IPA in SMYD3 knockdown activated genes.

| Name                                         | Number of genes | p-value               |
|----------------------------------------------|-----------------|-----------------------|
| Cell death and Survival                      | 31              | 2.12E-06 – 2.18E-02   |
| Cell Cycle                                   | 21              | 3.00E-05 – 2.24E-02   |
| Cellular Growth and Proliferation            | 20              | 3.44E-05 – 2.24E-02   |
| DNA Replication, Recombination, and Repair   | 15              | 4.49E-05 – 2.18E-02   |
| Cellular Development                         | 31              | 1.13E-04 – 2.30E-02   |
| RNA Post-Transcriptional Modification        | 7               | 1.99E-04 – 1.69E-02   |
| Cellular Compromise                          | 5               | 1.01E-03 – 1.35E-02   |
| Cell Morphology                              | 20              | 1.87E-03 – 1.68E-02   |
| Cellular Function and Maintenance            | 12              | 2.99E-03 – 2.02E-02   |
| Cellular Movement                            | 13              | 3.10E-03 – 2.27E-02   |
| Immune Cell Trafficking                      | 8               | 3.10E-03 – 1.58E-02   |
| Inflammatory Response                        | 8               | 3.10E-03 – 1.58E-02   |
| Amino Acid Metabolism                        | 1               | 3.40E-03 – 3.40E-03   |
| Carbohydrate Metabolism                      | 4               | 3.40E-03 – 1.69E-02   |
Figure S1

A

| Colon cell lines | Bladder cell lines | Breast cell lines | Prostate cell lines |
|------------------|--------------------|-------------------|---------------------|
| CCD-18Co | HCT116 | CaCO2 | HT29 | UROtsa | J82 | T24 | RT4 |
| anti-SMYD3 | | | | | | | |
| anti-Actin | | | | | | | |

B

| HCT116 | CaCO2 | J82 | T24 |
|--------|-------|-----|-----|
| Control sh | SMYD3 sh | Control sh | SMYD3 sh | Control sh | SMYD3 sh | Control sh | SMYD3 sh |
| anti-SMYD3 | | | | | | | |
| anti-Actin | | | | | | | |

C

| HCT116 | CaCO2 | J82 | T24 |
|--------|-------|-----|-----|
| Absorbance (570 nm) | Absorbance (570 nm) | Absorbance (570 nm) | Absorbance (570 nm) |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |

D

| HCT116 | CaCO2 | J82 | T24 |
|--------|-------|-----|-----|
| Invaded cell numbers | Invaded cell numbers | Invaded cell numbers | Invaded cell numbers |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
Figure S2

![Bar graph showing relative mRNA levels for various genes under control sh and SMYD3 sh conditions.](image_url)
Figure S3

A

|            | Control sh | SMYD3 sh | SMYD3 sh + FLAG-SMYD3 wt | SMYD3 sh + FLAG-SMYD3 F183A |
|------------|------------|----------|--------------------------|----------------------------|
| SMYD3      | +          | -        | -                        | -                          |
| FLAG-SMYD3 | -          | -        | +                        | -                          |
| FLAG-SMYD3 F183A | -    | -        | -                        | +                          |

- Control sh + − − − −
- SMYD3 sh − + + +
- FLAG-SMYD3 wt − − + −
- FLAG-SMYD3 F183A − − − +

B

MFG8

-2 -1 TSS 1 2 3 kb

SMYD3

% of Input

H3K4me3/H3

Arbitrary unit

0

2

4

6

0.0

0.1

0.2

0.3

0.4

A B C D E F

A B C D E F

A B C D E F
Figure S4
Figure S5

**A**

**FNBP1**

```
-300  gctcagcactgccgaagcctggtcgaagcagctag
  -100  gccgaggaagtcgctgcggcctggtgccg
       +1  gacagtggtggcctgcggccgtccctcacacct
```

**PDLIM7**

```
-300  gccgaggaagtcgctgcggcctggtgccg
       +1  gacagtggtggcctgcggccgtccctcacacct
```

**MFGE8**

```
-200  gctcagcactgccgaagcctggtcgaagcagctag
  -100  gccgaggaagtcgctgcggcctggtgccg
       +1  gacagtggtggcctgcggccgtccctcacacct
```

**B**

**FNBP1 (F)**

-261 +33

**MFGE8 (M)**

-185 +43

**PDLIM7 (P)**

-95 +151

**WNT3A (W)**

-434 -260

**KRT81 (K)**

-330 -210

**C**

**HCT116**

- **Control sh** - **SMYD3 sh**

**CaCO2**

- **Control sh** - **SMYD3 sh**

**J82**

- **Control sh** - **SMYD3 sh**

**T24**

- **Control sh** - **SMYD3 sh**

**D**

**HCT116**

- **Control sh** - **SMYD3 sh**

**CaCO2**

- **Control sh** - **SMYD3 sh**

**J82**

- **Control sh** - **SMYD3 sh**

**T24**

- **Control sh** - **SMYD3 sh**
Figure S6

[Diagram showing protein interactions and bands for different SMYD3 constructs with His-SMYD3 and anti-His antibody.]
Figure S7

![Graph showing wavelength (nm) vs. $\theta_{\text{res}} \times 10^2$ (deg cm$^2$/dmol)]
Figure S8

A

Control sh + – – –
SMYD3 sh – + + +
FLAG-SMYD3 wt – – + –
FLAG-SMYD3 mt – – – +

anti-SMYD3
anti-FLAG
(FLAG-SMYD3)
anti-PC4
anti-Actin

B

Control sh
SMYD3 sh
SMYD3 sh + FLAG-SMYD3 wt
SMYD3 sh + FLAG-SMYD3 mt

MFG8

-2 -1 TSS 1 2 3 kb

SMYD3

% of Input

0.3

0.2

0.1

0.0

A B C D E F

PC4

% of Input

0.5

0.4

0.3

0.2

0.1

0.0

A B C D E F

H3K4me3/H3

Arbitrary unit

2

3

4

5

0

A B C D E F

C

Control sh + – – –
PC4 sh – + + +
FLAG-PC4 wt – – + –
FLAG-PC4 mt – – – +

anti-PC4
anti-FLAG
(FLAG-PC4)
anti-SMYD3
anti-Actin

D

Control sh
PC4 sh
PC4 sh + FLAG-PC4 wt
PC4 sh + FLAG-PC4 mt

MFG8

-2 -1 TSS 1 2 3 kb

PC4

% of Input

0.5

0.4

0.3

0.2

0.1

0.0

A B C D E F

SMYD3

% of Input

0.3

0.2

0.1

0.0

A B C D E F

H3K4me3/H3

Arbitrary unit

2

3

4

5

0

A B C D E F
Figure S9

A

Relative mRNA level

B

SMUG1

C

Relative mRNA level

D

SMUG1

SMYD3

PC4

H3K4me3/H3
Figure S10

MFGE8

TSS 1 kb
sgRNA: G1 G2 (M) G3 G4 (M)

dCas9-FLAG
dCas9-FLAG-SMYD3 wt
dCas9-FLAG-SMYD3 F183A

MFGE8

Relative mRNA level

FNBP1

Relative mRNA level

PDLIM7

Relative mRNA level

WNT3A

Relative mRNA level
Figure S11

TSS  
\text{MGE8}  
1\ kb  
sgRNA :  
G1 G2  
C  
G3 G4  
D

\begin{figure}
\begin{center}
\begin{tabular}{ccc}

\text{dCas9-FLAG-SMYD3 wt} & \text{dCas9-FLAG-SMYD3 F183A} & \text{dCas9-FLAG + G1,2} \\
\text{dCas9-FLAG-SMYD3 wt + G1,2} & \text{dCas9-FLAG-SMYD3 F183A + G1,2} & \\
\text{dCas9-FLAG-SMYD3 wt + G3,4} & \text{dCas9-FLAG-SMYD3 F183A + G3,4} & \\
\end{tabular}
\end{center}
\end{figure}