**Supplementary Material**

MATERIALS AND METHODS

***Real time RT-PCR***

An iScript cDNA Synthesis kit (Bio-Rad), and iTaq SYBR Green Supermix with ROX (Bio-Rad) were used for reverse transcription of RNA and real-time PCR amplification of cDNA. The sequences of the primers used for PCR are shown in *SM*. Changes in mRNA levels were calculated by the 2-ΔΔC*T* method using GAPDH or 18S rRNA for normalization.

Sequences of the primers used for qRT-PCR (5’ to 3’ direction).

|  |  |  |
| --- | --- | --- |
| Gene | Forward | Reverse |
| *DNMT1* | AAGGGAAGGGCAAGGGAAAAGG | AGAAAACACATCCAGGGTCCGCAG |
| *DNMT3A* | CCATAAAGCAGGGCAAAGAC | AGTGGACTGGGAAACCAAATAC |
| *DNMT3B* | AATGTGAATCCAGCCAGGAAAGGC | ACTGGATTACACTCCAGGAACCGT |
| *EVI2A* | TACAAATGTCAGCCAATGAC | TCTAGCTAGTATAGGTAGGGC |
| *CCL19* | CCAACTCTGAGTGGCACCAA | TGAACACTACAGCAGGCACC |
| *PDHX* | GCAAATGCCAGATGTTAATG | GCAATTTCCTGGATACCTTTAG |
| *UBE4A* | GAGAATCAAGTCTCTTGCAG | ACTACGGTTAAAGGGATCTG |
| *IRF9* | CTCAGAAAGTACCATCAAAGC | TCATTATTGAGGGAGTCCTG |
| *FXYD6* | TATGATTACCAGACCCTGAG | CTGCGACTTAGGATAAGGAG |
| *GAPDH* | AGCCACATCGCTCAGACAC | GCCCAATACGACCAAATCC |

***Cell culture***

SKOV3 cells were maintained in media containing 1:1 MCDB 105 (Sigma-Aldrich, St. Louis, MO) and M199 (Cellgro, Herndon, VA, USA). OVCAR5 cells were cultured in DMEM, A2780 and OVCAR3 in RPMI-1640 (ATCC # 30-2001). Media were supplemented with 10% (20% for RPMI-1640) fetal bovine serum, 100 units/mL penicillin, and 100 μg/mL streptomycin. Cisplatin resistant A2780, SKOV3 and OVCAR5 cells were developed by treating parental cells with increasing concentrations of cisplatin. Cells received 3 rounds of cisplatin treatment and were given 3-4 weeks of recovery after each treatment. To estimate IC50 values, cells were exposed to cisplatin for 24 h, cisplatin was removed from the media (wash off), and cell survival was measured 72 h later using a CCK8 kit (Dojindo Molecular Technologies, Rockville MD). The IC50 values for parental and cisplatin-resistant SKOV3 cells were 3.98 µM, and 6.30 µM, A2780 cells were 7.24 µM and 53.85 µM, and for OVCAR5 1.35 µM and 6.94 µM, respectively. Cells were treated with guadecitabine (Astex Pharmaceuticals, Inc.) at 100 nM for 3 days before harvest for specific gene expression validation.

***DNA and RNA extraction***

DNA and total RNA were extracted from approximately 25 mg tumor tissue or 5 million cultured cells using AllPrep DNA/RNA/Protein Mini kit (Qiagen, Valencia CA). Alternatively, DNA was extracted using a QIAamp DNA Mini kit (Qiagen) and total RNA with TriReagent (Sigma-Aldrich) in some experiments. The procedures employed followed the manufacturers’ instructions. DNA and RNA concentrations were determined by using the absorbance at 260 nm, and purity was assessed based on the 260/280 nm absorbance ratio.

**Figure legends**

**Figure S1.** (a) Treatment administration schema and timing of tumor biopsies. (b) A flow chart of data analysis and sample comparisons.

**Figure S2**. (a) Pathways and (b) networks (red molecules represent hypermethylated, while the green molecules represent hypomethylated) enriched/altered by differentially methylated genes (*P*adj<0.05, |Δβ|>0.1) between primary OC and human ovarian surface epithelia cells (HOSE).

*(Related data Supplementary Table 1. Differentially methylated gene list between primary OC and HOSE)*

**Figure S3**. (a) Pathways and (b) networks (red molecules represent hypermethylated, while the green molecules represent hypomethylated) enriched by differentially methylated genes (*P*adj<0.05, |Δβ|>0.1) between recurrent OC and human ovarian surface epithelia cells (HOSE).

*(Related data Supplementary Table 2. Differentially methylated gene list between platinum resistant OC and HOSE)*

**Figure S4**. (a) Pathways and (b) networks (red molecules represent hypermethylated, while the green molecules represent hypomethylated) enriched by differentially methylated genes (*P*adj<0.05, |Δβ|>0.1) between guadecitabine-treated OC and human ovarian surface epithelia cells (HOSE).

*(Related data Supplementary Table 3. Differentially methylated gene list between guadecitabine-treated OC and HOSE)*

**Figure S5**. Estimations of half maximal inhibitory concentrations (IC50) of cisplatin in OVCAR5, SKOV3, and A2780 epithelial ovarian cancer cells.

**Figure S6**. Baseline levels of DNMT1, DNMT3A, and DNMT3B mRNAs in OC tumors (a), and correlations between DNMT mRNA levels (b,c,d) (n=19) .