The CDO1 gene contains 5 exons dispersing over a length of about 15 kb on chromosome 5q23.2. The 5'-flanking region is GC rich (72%) and contains several putative regulatory elements including an antioxidant response element (ARE) and reactive oxygen intermediate target (CArG box)-like sequences, which mediate the transcriptional induction of genes involved in antioxidant defense and detoxification. CDO1 encodes a protein of 200 amino acids with a molecular weight of 23 kDa. CDO1 is a non-heme iron-containing dioxygenase (EC 1.13.11.20) and the key enzyme in the homeostasis of cysteine and glutathione.
Supplementary Figure 2

(A) DNA methylation and mRNA expression status of CDO1, assayed by MSP and RT-PCR, in three representative cell lines of each cancer type. GAPDH expression was assayed as control. Basal expression of CDO1 was confirmed in normal tissue of each cancer type. (B) Overall DNA methylation frequency (in %) of CDO1 in primary samples of ovarian (n = 65), lung (n = 65), pancreas (n = 30), and hepatocellular cancer (n = 21). (C) Scatter plots depicting correlation between expression log base 2 values (y-axis) (analyzed on Agilent 244K Custom Gene Expression G4502A-07 platform) and DNA methylation β values (x-axis) (analyzed on Illumina HumanMethylation 27k platform) of CDO1 in primary samples of lung (n = 104) and ovarian cancer (n = 584) from TCGA Data Portal. (D) Box plots demonstrating a statistically significant correlation between tumor stage and CDO1 methylation frequency in TCGA lung but not in ovarian cancers. A p < 0.05 was considered statistically significant.

Supplementary Figure 2 - DNA methylation-associated silencing of CDO1 occurs in multiple types of cancer

(A) DNA methylation and mRNA expression status of CDO1, assayed by MSP and RT-PCR, in three representative cell lines of each cancer type. GAPDH expression was assayed as control. Basal expression of CDO1 was confirmed in normal tissue of each cancer type. (B) Overall DNA methylation frequency (in %) of CDO1 in primary samples of ovarian (n = 65), lung (n = 65), pancreas (n = 30), and hepatocellular cancer (n = 21). (C) Scatter plots depicting correlation between expression log base 2 values (y-axis) (analyzed on Agilent 244K Custom Gene Expression G4502A-07 platform) and DNA methylation β values (x-axis) (analyzed on Illumina HumanMethylation 27k platform) of CDO1 in primary samples of lung (n = 104) and ovarian cancer (n = 584) from TCGA Data Portal. (D) Box plots demonstrating a statistically significant correlation between tumor stage and CDO1 methylation frequency in TCGA lung but not in ovarian cancers. A p < 0.05 was considered statistically significant.
Supplementary Figure 3- Tumor-specific point mutations within the *CDO1* gene.
Protein damaging missense mutations within the *CDO1* gene in primary breast tumor tissue of four patients. Arrows indicate the position of the mutation. Tumor-specificity of mutations was confirmed in non-tumor tissue of same patients.