**Materials and Methods**

**RNA interference**

For stable shRNA knockdowns, cells were seeded in a six-well plate to 60-80% confluency and subsequently transduced with 200-500 μl lentiviral particles expressing shRNAs (obtained from Open Biosystems/Thermo Scientific, listed in Table III) in a total volume of 2 ml of appropriate media supplemented with ~6-10 µg/ml polybrene. Media was replaced after overnight incubation and cells were subjected to puromycin selection (2 µg/ml) for at least 3 days as has been described previously (1).

**Quantitative RT-PCR (qRT–PCR)**

Total RNA was isolated and reverse transcribed using Superscript II Reverse Transcriptase (Invitrogen). Quantitative real-time PCR was performed as has been described previously (1) using primers listed in Table II. Gene expression was normalized to *Gapdh.* Controls lacking reverse transcriptase were carried out in parallel to rule out the possibility of DNA contamination. Each sample was analyzed at least two independent times and the results from at least three different biological triplicates are presented.

**TRIM37, p53, E2F1, STAT1 and STAT3 overexpression**

A TRIM37 cDNA clone (Origene) was sub-cloned into the vector pcDNA3.1(-) (Sigma) using Not1 and BamH1 sites, and verified by full-length sequencing. TRIM37 expression vector was linearized with PciI and transfected into cells using either 4D Amaxa nucleofection system (Lonza) or Effectene reagent (Qiagen). The p53 (provided by Marty Mayo), E2F1 (provided by Marty Mayo), STAT1 (Addgene) or STAT3 (provided by Gholam Mohi) cDNA clones were transfected into cells using Effectene reagent (Qiagen). After 24 hrs., cell medium was replaced by appropriate selection markers and stable cells were selected.

**Comet assay**

Cells were treated with 1 µM of Dox, and harvested at 24 hrs. Neutral comet assays with ethidium bromide staining (Invitrogen) were performed as has been described previously (2). The quantification of tail DNA was done using OpenComet software (3).

**DSB repair efficiency assay**

NHEJ and HR assays were designed and performed as described previously (4,5). MDA-MB-468 cells were stably transfected with pimEJ5GFP (Addgene) for the NHEJ assay or pDRGFP (Addgene) for the HR assay. Stable cell lines were infected with control or TRIM37 shRNA lentivirus, followed with transfection with pCBASceI (Addgene). After 24 hrs., cells were analyzed for GFP expression by flow cytometry and normalized to non-silencer (NS) control for each biological replicate.

**Clonogenic assay**

Cells were plated in 6-well plates after indicated treatment and cultured until visible colonies were observed. Cells were treated with chemotherapeutic drugs as indicated at the following concentrations: 0.1-1 µM doxorubicin (Cayman Chemical), 0.1-1 µM Daunorubicin (Cayman Chemical), 250-500 µM temozolomide (Cayman Chemical), 0.5-25 µM etoposide (Cayman Chemical) and 1-10 µM cisplatin (Cayman Chemical) for 24hrs. Colonies were fixed (100% methanol, 37oC) and stained with 0.1% crystal violet dissolved in 20% methanol/80% PBS. Cells were imaged using a Chemidoc (BioRad) and colonies were counted.

**Caspase 3 activity assay**

The Caspase 3 assay was performed using Caspase 3 assay kit (BD Pharmingen) according to the manufacturer’s instructions.

**Directed-ChIP assays**

ChIP assays were performed as has been described previously (1) using cell extracts prepared 1 day after Cas9 transduction or inhibitor treatment, and antibodies against BMI1 (Abcam), EZH2 (Cell Signaling Technology), H2Aub (Cell Signaling Technology), TRIM37 (Abcam), STAT1 (Cell Signaling Technology), STAT3 (Cell Signaling Technology), E2F1 (Cell Signaling Technology), KU80 (Cell Signaling Technology), KU70 (Cell Signaling Technology), RAD51C (Cell Signaling Technology), NBS1 (Cell Signaling Technology) and as a negative control, IgG. The sequences of primers used for amplifying ChIP products are listed in Table II. Each ChIP experiment was carried out at least three independent times and the results from at least three biological experiments, with technical duplicates, are shown.

**Sucrose gradient fractionation**

Sucrose gradient sedimentation analysis was performed as described previously (1). Briefly, 10–40% gradients were formed by layering 200 μl NEB1 buffer containing 10%, 20%, 30% or 40% sucrose in a 13 x 51 mm centrifuge tube (Beckman) and allowed to equilibrate at room temperature for 2 hrs. Gradients were chilled, loaded with 500 mg HCC1806 extract (adjusted to a volume of 200 μl) or 200μl molecular weight markers (Sigma MW-GF-1000), and centrifuged in a Beckman SW 55 Ti rotor at 214,000g for 14 hrs. Twenty-five fractions of 200μl were collected. For the markers, 20μl of each fraction was electrophoresed and Coomassie stained. For the gradient fractions, 20μl of fractions was analyzed by immunoblotting using TRIM37 (Abcam), KU80 (Cell Signaling Technology), KU70 (Cell Signaling Technology), RAD51C (Cell Signaling Technology), MRE11 (Cell Signaling Technology) and NBS1 (Cell Signaling Technology).

**Immunoblotting**

Cell extracts were prepared by lysing cell pellet in RIPA buffer supplemented with 1 mM sodium ortho-vanadate and 10 mM PMSF. To prepare protein extract from mouse tissue, it was homogenized in lysis buffer (1% SDS, 1 mM sodium ortho-vanadate, 10 mM Tris pH 7.4 and protease inhibitor). Immunoblots were probed using antibodies against TRIM37 (Abcam), PARP (Cell Signaling Technology), p21 (Cell Signaling Technology), γ-ph-H2AX (Cell Signaling Technology), ph-p53 (Cell Signaling Technology), RAD51C (Invitrogen), ph-ATM (Cell Signaling Technology), ph-STAT1 (Cell Signaling Technology), ph-STAT3 (Cell Signaling Technology), ph-STAT5 (Cell Signaling Technology), BIM (Abcam), STAT 3 (Cell Signaling Technology) , STAT1 (Cell Signaling Technology) , GAPDH (Cell Signaling Technology), FOLR1 (R&D) and α-tubulin (Invitrogen).

**Co-immunoprecipitation**

MDA MB 468 nuclear extract (~1mg) was incubated with either TRIM37 (Abcam), XRCC5 (Cell Signaling Technology), KU80 (Invitrogen), KU70 (Invitrogen), NBS1 (Invitrogen), or RAD51C (Invitrogen) antibody at 4oC for 24 hrs. in the presence of ethidium bromide (100 µg/ml). Immune complexes were captured on protein A/G-Agarose (GenDEPOT), washed three times in NEB1 buffer, and eluted by boiling for 10 min in SDS sample buffer. Immunoprecipitated proteins were analyzed by immunoblotting as has been described earlier. Input lanes represent ~1-5% of extract loaded in the immunoprecipitation lanes.

**Chemical inhibitor treatment**

MDA-MB-468 cells were treated for 24hrs. with 10 µM Dox and small molecule inhibitors at the following concentrations: 10 µM KU000553 (Cayman Chemical), 50 µM AG490 (Cayman Chemical), and 40 µM HLM006474 (Sigma-Aldrich) for 24 hrs.

**MTT cell proliferation assay**

MTT was performed using MTT Cell Proliferation Assay Kit (Trevigen) according to the manufacturer’s instructions.

**Antibody Purification**

Transfected cultures were harvested after 10 days and filtered through 0.2-mm PES membrane filters (Millipore Express Plus). Cleaning-in-place was performed for each column using 0.2M NaOH wash (20 min.). Following cleaning, columns were washed 3 times with Binding buffer (20 mM sodium phosphate, 0.15 M NaCl, pH7.2). Filtered supernatant containing recombinant antibodies or antigens were passed through the columns at 4°C. Prior to elution in 0.1 M sodium citrate pH 3.0–3.6, the columns were washed 3 times with binding buffer, pH 7.0. The pH of eluted antibodies was immediately neutralized using 3M sodium acetate, pH 9.0. After protein measurements at 280 nm, antibodies were dialyzed in PBS using Slide-A-Lyzer 3.5K (Thermo Scientific). Antibodies were run on gel filtration columns to analyze the percent monomers. Whenever necessary a second step size exclusion chromatography was performed.

**Size exclusion chromatography**

The percent monomer of purified antibodies was determined by size exclusion chromatography as described previously (26).

**Binding studies by ELISA**

Binding specificity and affinity of described *Farletuzumab* was determined by ELISA using the recombinant FOLR1 as described previously (6). Briefly, recombinant FOLR1 protein was coated on 96-well plate by incubation overnight at 4oC. Different amounts of nanoparticles and free antibody were incubated in the coated plates and binding was detected by two-component peroxidase substrate kit (BD Biosciences).

**Particle Characterization**

The particle size was determined by dynamic light scattering (DLS) and the zeta potential of NPs was determined using a Zetasizer ZS (Malvern). The morphology of NPs was characterized by transmission electron microscope (JEOL JEM 1400 instrument, JEOL Ltd., Japan) at a voltage of 120 kV using CLSM (Zeiss LSM510 instrument, Carl Zeiss) at the Electron Microscopy Core. Briefly, nanoparticles were dissolved in 0.01 M PBS (pH 7.4) and were negatively stained with 1% phosphotungstic acid as described previously (7,8). A dialysis method was used to measure the release kinetics of ASO-loaded nanoparticles. In brief, 5 mg of drug-loaded NPs was dissolved in PBS with 0.1% (v/v) Tween 80, and dialyzed against 5 mL of that same buffer using ready-to-use dialysis tubes (MWCO 12,000-14,000; Fisher Scientific) under continuous stirring at 37oC. At predetermined time points, 10 μL aliquot was removed for quantification and replaced with fresh buffer. The concentration of the drug was determined by Nanodrop (Thermo Scientific).

**FOLR1-overexpression cellular target and uptake**

HCC1806RR cells with either MCF7 or MCF10A were seeded onto an 8 well-chamber slide (Thermo Fisher Scientific Inc.) at a ratio of 1:1 with cell concentration of 30,000 per well in 100 μl of medium and cultured overnight at 37oC. The original medium was replaced with fresh medium containing 8.3x 106 nanoparticles. Cells were incubated for indicated times and imaged for nanoparticle uptake by Zeiss Axio Observer 200 inverted microscope (Zeiss).

**Immunofluorescence**

Cells were stained with an anti-phospho γH2AX (Santa Cruz Biotechnology), TRIM37 (Abcam), NBS1 (Cell Signaling Technology), RAD51c (Novus Biologics), XRCC5 (Invitrogen) and XRCC6 (Invitrogen). Cells were visualized on a Zeiss Axio Observer Live-Cell microscope and images were adjusted for contrast and brightness using AxioVision Software. For quantification, 100–500 cells in at least 10 different fields from biological and technical replicates were counted and scored for positive signal.

**Patient sample analysis**

Patients with triple negative breast cancers treated between 2001 and 2009 at Saint Louis hospital (Paris), by neoadjuvant chemotherapy regimen and with available pre and post-chemotherapy frozen tumor tissues were selected.  Frozen tumor tissue sections were provided by the biological resource center of St Louis Hospital (agreement # DC 2009-929), following the Ethics and Legal national French rules for the patients' information and consent (ANAES, HAS and INCA). Total RNAs were extracted using phenol/chloroform method (29). The p53 status was determined by the FASAY yeast functional assay, as previously reported (30,31).

**Tissue distribution studies**

FOLR1 antibody was tagged with IRDye® 800 CW NHS Ester fluorochrome as described previously (26). To examine the tumor accumulation of smart nanoparticles, 6-8 weeks old NSG mice bearing ~200-500 mm3 xenograft tumors were injected with a single dose of smart nanoparticles. Tissue distribution was monitored using fluorescent imaging at the indicated times. A subset of animals was euthanized and spleen, kidney, liver, lungs and tumor were isolated. Different tissues were exposed directly to the excitation wavelength (772 nm) to monitor tissue specific fluorescent signal for smart nanoparticles.

**AST/ALT assays**

To examine the hepatoxicity of the smart nanoparticle treatment, serum was isolated from blood samples collected from the treated mice at the termination of tumor studies. Samples were assessed for aspartate aminotransferase and alanine aminotransferase (ALT) levels using Liquid AST (SGOT) reagent set (Pointe Scientific) and EnzyChrom Alanine Transaminase Assay Kit (Bioassay Systems) as per manufacturer’s instruction.

**Hematoxylin and eosin, Ki67, Caspase 3 and TRIM37 staining**

Animals were either perfused with 10% neutral buffered formalin or organs were collected and then fixed in 10% neutral buffered formalin and embedded in paraffin. H&E staining was performed by Research Histology Core. Ki67, Caspase 3 and TRIM37 (Abcam) staining was performed by Biorepository and Tissue Research Facility.

**Identification of tumor and metastasis suppressors**

Candidate TRIM37 target genes were systematically queried against Tumor suppressor database (https://bioinfo.uth.edu/TSGene).

**RNA-seq analysis**

231-2b cells were transfected with control or TRIM37-ASO twice and RNA was isolated with RNeasy Mini Kit (Qiagen) at day 7 post-transfection. RNA-seq was performed by Novogene genome Sequencing Company using Illlumina Novoseq platform with paired-end 150 bp sequencing strategy. RNA-Seq data was aligned to the human genome assembly GRCm38/hg38 using STAR software package. Using DESeq2 R package, differential expression of genes in control and TRIM37-ASO cells was determined with a
significant criterion padj<0.05. GSEA (10) was used for gene set
enrichment analysis for all genes with FPKM>1. GeneTrail2 (11) was used for analysis of enriched KEGG pathways. MA plot and heatmap of gene expression clusters were drawn using R.

1. Bhatnagar S, Gazin C, Chamberlain L, Ou J, Zhu X, Tushir JS*, et al.* TRIM37 is a new histone H2A ubiquitin ligase and breast cancer oncoprotein. Nature **2014**;516:116-20

2. Zhang Y, He Q, Hu Z, Feng Y, Fan L, Tang Z*, et al.* Long noncoding RNA LINP1 regulates repair of DNA double-strand breaks in triple-negative breast cancer. Nat Struct Mol Biol **2016**;23:522-30

3. Gyori BM, Venkatachalam G, Thiagarajan PS, Hsu D, Clement MV. OpenComet: an automated tool for comet assay image analysis. Redox Biol **2014**;2:457-65

4. Bennardo N, Cheng A, Huang N, Stark JM. Alternative-NHEJ is a mechanistically distinct pathway of mammalian chromosome break repair. PLoS Genet **2008**;4:e1000110

5. Pierce AJ, Johnson RD, Thompson LH, Jasin M. XRCC3 promotes homology-directed repair of DNA damage in mammalian cells. Genes Dev **1999**;13:2633-8

6. Shivange G, Urbanek K, Przanowski P, Perry JSA, Jones J, Haggart R*, et al.* A Single-Agent Dual-Specificity Targeting of FOLR1 and DR5 as an Effective Strategy for Ovarian Cancer. Cancer Cell **2018**;34:331-45 e11

7. Lou S, Zhao Z, Dezort M, Lohneis T, Zhang C. Multifunctional Nanosystem for Targeted and Controlled Delivery of Multiple Chemotherapeutic Agents for the Treatment of Drug-Resistant Breast Cancer. ACS Omega **2018**;3:9210-9

8. Chen L, Watson C, Morsch M, Cole NJ, Chung RS, Saunders DN*, et al.* Improving the Delivery of SOD1 Antisense Oligonucleotides to Motor Neurons Using Calcium Phosphate-Lipid Nanoparticles. Front Neurosci **2017**;11:476

9. Jenkins DE, Hornig YS, Oei Y, Dusich J, Purchio T. Bioluminescent human breast cancer cell lines that permit rapid and sensitive in vivo detection of mammary tumors and multiple metastases in immune deficient mice. Breast Cancer Res **2005**;7:R444-54

10. Subramanian A, Tamayo P, Mootha VK, Mukherjee S, Ebert BL, Gillette MA*, et al.* Gene set enrichment analysis: a knowledge-based approach for interpreting genome-wide expression profiles. Proc Natl Acad Sci U S A **2005**;102:15545-50

11. Stockel D, Kehl T, Trampert P, Schneider L, Backes C, Ludwig N*, et al.* Multi-omics enrichment analysis using the GeneTrail2 web service. Bioinformatics **2016**;32:1502-8