## Supplementary Figure S1



Figure S1.
HCT116 cells were treated with $2 \mu \mathrm{~g} / \mathrm{ml}$ of ADR for 2 h . At 36 h after treatment, cells were harvested. Whole cell extracts were subjected to immunoblotting with anti-modified citrulline (MC) or anti-p53 antibody.

## Supplementary Figure S2



Figure S2.
At 6 h after transfection with each siRNA, U373MG cells were infected with Ad-p53 or Ad-LacZ. At 36 h after infection, mRNA levels of PADI3 (upper) and PADI4 (lower) were analyzed by qPCR analysis. Relative expression was shown with standard deviations ( $\mathrm{n}=2$ ).

## Supplementary Figure S3



Figure S3.
Results of luciferase assay of p53BS-AB (PADI4) are shown. Luciferase activity is indicated relative to the activity of mock vector with standard deviations ( $n=2$ ). Reporter plasmids which contain $p 53$-responsive elements of BAX, FAS, and p21 WAF1 were used as positive controls.

## Supplementary Figure S4



Figure $\mathbf{S 4}$.
HEK293T cells were transfected with plasmid expressing HA-PADI4 (lane 1 and 2). U373MG cells were infected with Ad-p53 at 20 MOI (lane 3 and 4). At 48 h after treatment, cells were cultured with or without $5 \mu \mathrm{M}$ of A23187 for 1 h , followed by immunoblotting with anti-MC or anti-NPM1 antibody. Arrowhead on the right indicates the band corresponding to NPM1 protein.

## Supplementary Figure S5



Figure $\mathbf{S 5}$.
Generation of antibody against citrulinated NPM1 at arginine 197 residue. Antibody titers and specificity were determined by enzyme-linked immunosorbent assay (ELISA) using peptide Cit197 that was used for immunization and peptide Arg 197 that was used for adsorption.

## Supplementary Figure S6



Figure $\mathbf{S 6}$.
A, Subcellular distribution of NPM1 proteins was examined by immunocytochemistry. At 36 h after cotransfection of plasmid expressing myc-NPM1 with p53, H1299 cells were fixed and stained with anit-myc antibody (Alexa fluor 488) and anti-p53 antibody (Alexa fluor 594). B, Subcellular distribution of Nucleolin proteins was examined by immunocytochemistry. At 36 h after transfection with plasmid expressing HAPADI4, HEK293T cells were fixed and stained with anit-nucleolin antibody (Alexa fluor 488) and anti-HA antibody (Alexa fluor 594). C, Subcellular distribution of NPM1 proteins was examined by immunocytochemistry. At 36 h after co-transfection of plasmid expressing myc-NPM1 or myc-NPM R197K with HA-PADI4, HEK293T cells were fixed and stained with anit-myc antibody (Alexa fluor 488) and anti-HA antibody (Alexa fluor 594).

## Supplementary Table S1

Sequences of primers and RNA nucleotides

| Site-directed mutagenesis | Forward | Reverse |
| :--- | :--- | :--- |
| PADI4D350A | GACCAGTGGATGCAGGCTGAAATGGAGATCGGC | GCCGATCTCCATTTCAGCCTGCATCCACTGGTC |
| PADI4D473A | TCCGTGGGCCACGTGGCCGAGTTCCTGAGCTTT | AAAGCTCAGGAACTCGGCCACGTGGCCCACGGA |
| siRNA oligonucleotides | Sense | Antisense |
| siNPM1 | GGACAAGAAUCCUUCAAGATT | UCUUGAAGGAUUCUUGUCCTT |
| siPADI3 | CCACAAACUUGUCCUCCAUTT | AUGGAGGACAAGUUUGUGGTT |
| siPADI4 | GGUCCUGCUACAAACUGUUTT | AACAGUUUGUAGCAGGACCTT |
| sip53 | GACUCCAGUGGUAAUCUACTT | GUAGAUUACCACUGGAGUCTT |
| siEGFP | GCAGCACGACUUCUUCAAGTT | CUUGAAGAAGUCGUGCUGCTT |


| Quantitative real-time PCR | Forward | Reverse |
| :--- | :--- | :--- |
| PADI1 | CATGACGCCCAACACTCA | GAGCCATGAGTGTCCATCAC |
| PADI2 | AGAGCCTTGTGCAGGAGAAC | AGGATGTCACGGTTCCAGTC |
| PADI3 | GCCTTCTTCCCTGACTTGGT | AGCAGCAGCCATTGATGA |
| PADI4 | TTCTCTAAGGCGGAAGCTTTT | AGCAGGGAACACACCTTCTC |
| PADI6 | CAGGTGCCTCTGGAGGTTTA | TCGTCACTGTGTCCACAAAAC |
| B2M | TTCTGGCCTGGAGGCTATC | TCAGGAAATTTGACTTTCCATTC |


| ChIP assay / Gene reporter assays | Forward | Reverse |
| :--- | :--- | :--- |
| PAD4BS-A | TGGGTGTGAAGGAAATGACA | CCAAGGCCATTGAATGAGTT |
| PAD4BS-B | CACCTCAAGCCACACAGCTA | AAGTGATTTGCCCATCTTGG |
| PAD4BS-AB | TGGGTGTGAAGGAAATGACA | AAGTGATTTGCCCATCTTGG |
| BAX | AGGCTGAGACGGGGTTATCT | AGGCTGGGCCTGTATCCTAC |
| FAS | GAATTGAAGCGGAAGTCTGG | TTAACCACTGCTTCGGTGC |
| p21 ${ }^{\mathrm{WAF} 1}$ | ACCTTTCACCATTCCCCTAC | GCCCAAGGACAAAATAGCCA |

