Legends to Supplemental Figures

Supplemental Figure S1. Increased PARI expression does not alleviate HU-induced replication stress. For PARI overexpression, the commercial lentiviral construct pLV-Puro-TRE>hPARPBP (Vector Builder Inc.) was used. As PARI expression is doxycycline-inducible, HL60 cells were first transduced with the pLV[Exp]-Neo-UBC>tTS/rtTA_M2 lentiviral construct (Vector Builder Inc.) which expresses the reverse tetracycline-controlled transactivator (rtTA). **A.** RT-qPCR experiment showing PARI expression in control and PARI-overexpressing HL60 cells. Bars represent the average of 3 experiments with standard deviations shown as error bars. **B.** Cells were treated with the indicated dose of HU for 4 days and cellular viability was measured using the CellTiterGlo reagent. The average of 3 experiments is shown, with standard deviations indicated as error bars.

Supplemental Figure S2. Increased differentiation and reduced proliferation upon RAD51 depletion in U937 cells. For RAD51 knockdown, U937 cells were infected with a commercial lentiviral construct, namely pLV-EGFP:T2A:Neo-U6>hRAD51[shRNA#1] (targeting sequence: TTAGAGCAGTGTGGCATAAAT) obtained from VectorBuilder Inc., and subjected to G418 selection **A.** Quantifications of CD14 differentiation marker on the surface of U937 cells by flow cytometry. The mean value of CD14 staining is shown, normalized to control cells. The average of 4 experiments is shown, with error bars indicating standard deviations. Asterisks indicate statistical significance. **B.** Proliferation of RAD51-knockdown U937 cells was measured using the CellTiterGlo reagent, after 4 days in culture (normalized to day 0). The average of 3 experiments is shown, with error bars indicating standard deviations. Asterisks indicate statistical significance. **C**. Western blot showing reduced RAD51 levels in the knockdown cells.

Supplemental Figure S3. Increased NFκB activation in PARI-knockdown cells in response to DNA damage. Western blot showing increased p65 phosphorylation in PARI-depleted U937 cells under normal (no drug) growth conditions, and upon treatment with camptothecin (1μM for 24h) or bleomycin (100μg/ml for 24h).

Supplemental Figure S4. Depletion of p21 rescues the proliferation defect of PARI-knockdown U937 cells. For p21 knockdown, U937 cells were infected with a commercial lentiviral construct, namely pLV-EGFP:T2A:Neo-U6>hCDKN1A[shRNA#1] (targeting sequence: GAGCGATGGAACTTCGACTTT) obtained from VectorBuilder Inc., and subjected to G418 selection. **A**. RT-qPCR experiment showing p21 expression in control and p21-knockdown U937 cells. Bars represent the average of 3 experiments with standard deviations shown as error bars. **B.** Proliferation of U937 cells with PARI and/or p21 knockdown was measured using the CellTiterGlo reagent, after 4 days in culture (normalized to day 0). The average of 3 experiments is shown, with error bars indicating standard deviations. Asterisks indicate statistical significance.

Figure S1.

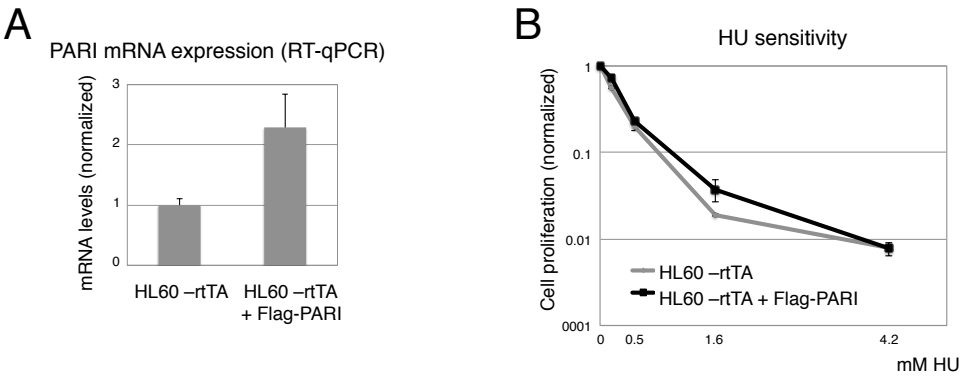

A
PARI mRNA expression (RT-qPCR)
mRNA levels (normalized)
3
2
1
0
HL60 –rtTA
HL60 –rtTA
+ Flag-PARI
B
HU sensitivity
Cell proliferation (normalized)
1
0.1
0.01
0001
HL60 –rtTA
HL60 –rtTA + Flag-PARI
0
0.5
1.6
4.2
mM HU

Figure S2.

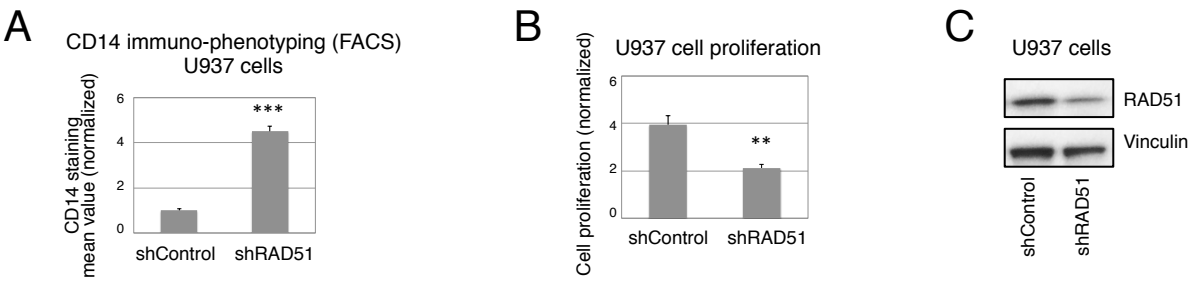

A
CD14 immuno-phenotyping (FACS)
U937 cells
CD14 staining
mean value (normalized)
6
4
2
0
***
shControl
shRAD51
B
U937 cell proliferation
Cell proliferation (normalized)
6
4
2
0
**
shControl
shRAD51
C
U937 cells
RAD51
Vinculin
shControl
shRAD51

Figure S3.

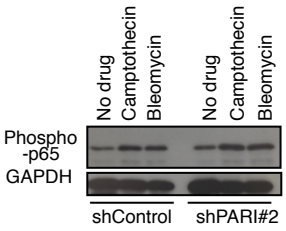
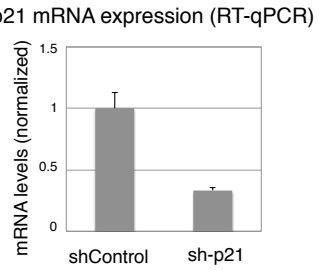
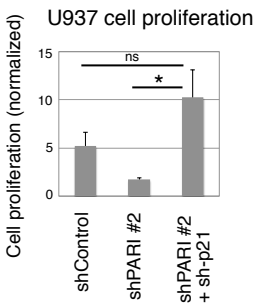

No drug
Camptothecin
Bleomycin
No drug
Camptothecin
Bleomycin
Phospho
-p65
GAPDH
shControl
shPARI#2

Figure S4.

A


p21 mRNA expression (RT-qPCR)
mRNA levels (normalized)
1.5
1
0.5
0
shControl
sh-p21

B


U937 cell proliferation
Cell proliferation (normalized)
15
10
5
0
ns
*
shControl
shPARI #2
shPARI #2
+ sh-p21