

SUPPLEMENTARY INFORMATION

MOF and histone H4 acetylation at lysine 16 are critical for DNA Damage Response

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Supplementary Fig. 1. Western blot analysis for hMOF, H4K16ac and some common DDR associated proteins in cells with and without depletion of hMOF. **(A)**. 293 cells were transfected with either hMOF-siRNA or Tip60-siRNA. Tip60 knock down had no effect on H4K16ac levels analyzed after 72 h post transfection with Tip60-siRNA. hMOF knock down showed an effect on H4K16ac levels even after 36 h post transfection of hMOF-siRNA. **(B)** 293 cells were transfected with Tip60-siRNA and Tip60 levels were

determined after 72 h of transfection. (C) 293 cells transfected with hMOF-siRNA were examined for different proteins after 72 h of transfection.

Supplementary Fig. 2. Cells at high magnification with and quantification of γ -H2AX foci observed post irradiation at various time points post-irradiation (1.5 Gy exposure) in cells with and without depletion of hMOF. (A) Cells were transfected with Cy3 labeled hMOF-siRNA and analyzed for γ -H2AX foci. (B) Cells were transfected with Cy3 labeled Tip60-siRNA and analyzed for γ -H2AX foci. (C, D) Cells were transfected with hMOF-siRNA (C) or Tip60-siRNA (D) (without Cy-3 labeling) and analyzed for γ -H2AX foci. For each time point, 100 cells were analyzed. Each experiment was repeated thrice with the mean number of cells with more than 2 foci are plotted against time. Each experiment was repeated thrice and the mean number of foci is plotted against time.

Supplementary Fig. 3. Frequency of cells with γ -H2AX foci following exposure to IR in cells with different knock down of both MOF and Tip60 in 293 cells for appearance (A) and disappearance (B) of cells with foci.

Supplementary Fig. 4. Frequency of cells with γ -H2AX foci following exposure to IR in cells with overexpression of wild type or mutant MOF. (A) Cells with overexpression of hMOF or hMOF knockdown and (B) cells with expression of HAT dead mutant (Δ) hMOF (2) with and without knock down of hMOF were irradiated with 1.5 Gy and analyzed for the appearance of IR-induced γ -H2AX foci.

Supplementary Fig. 5. HL60 cells were treated with DMSO for different time periods and examined for telomerase activity by the described procedure (1, 6).

Supplementary Fig. 6. Effect of ATM specific inhibitor (KU-55933) in 293 cells with and without hMOF knock down for appearance (A) and disappearance (B) of IR-induced γ -H2AX foci. Cells were irradiated with 1.5 Gy and each experiment was repeated thrice.

Supplementary Fig. 7. High magnification of the fluorescence of γ -H2AX, MOF and DNA at defined DNA DSB in mouse NIH2/4 cells.

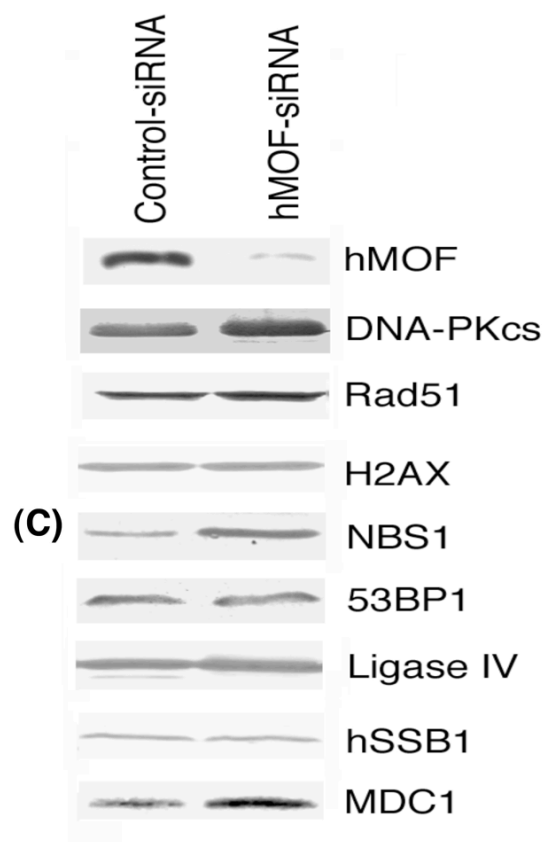
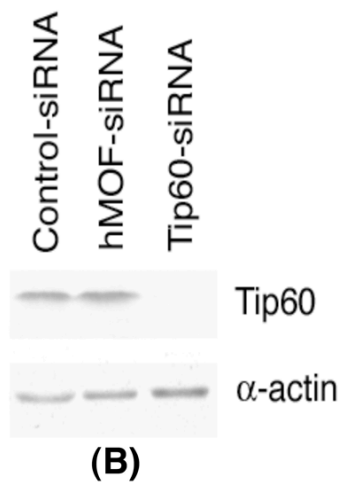
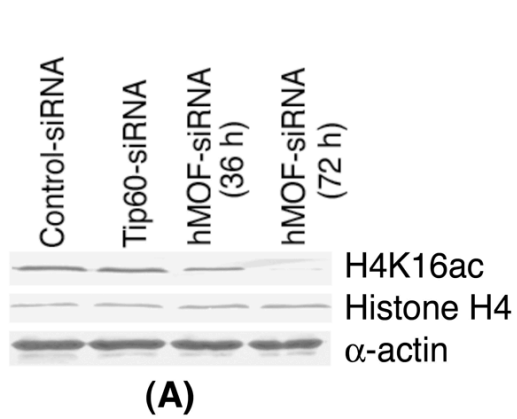
Supplementary Fig. 8. ChIP analysis in NIH2/4 cells with and without knock down of MOF. Cells with and without MOF depletion were treated with TA and examined for levels of bound MOF (A), H4K16ac (B) and histone H4 (C) at mice MOF locus by using the described procedure (5).

Supplementary Fig. 9. Influence of MOF on DNA-PKcs at damage site. (A) Interaction of hMOF with DNA-PKcs. hMOF-TAP is purified with distinct sets of associated proteins. Peptides corresponding to hMSL's and other proteins were identified (number of peptides shown in parenthesis). (B) Depletion of MOF influence on the accumulation and dispersal of DNA-PKcs at the DNA damage site. 293 cells expressing YFP-DNA-PKcs were irradiated and fluorescence intensity was measured. The results are mean of the three experiments.

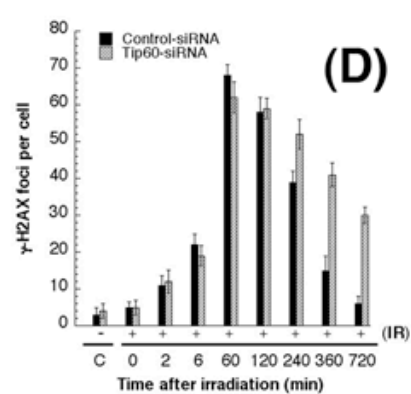
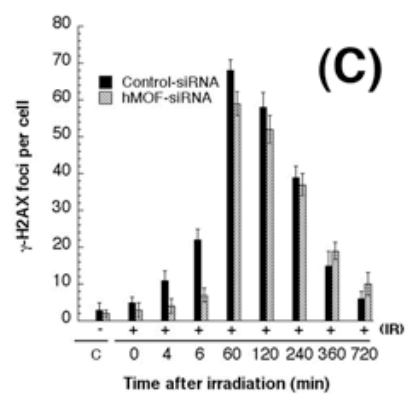
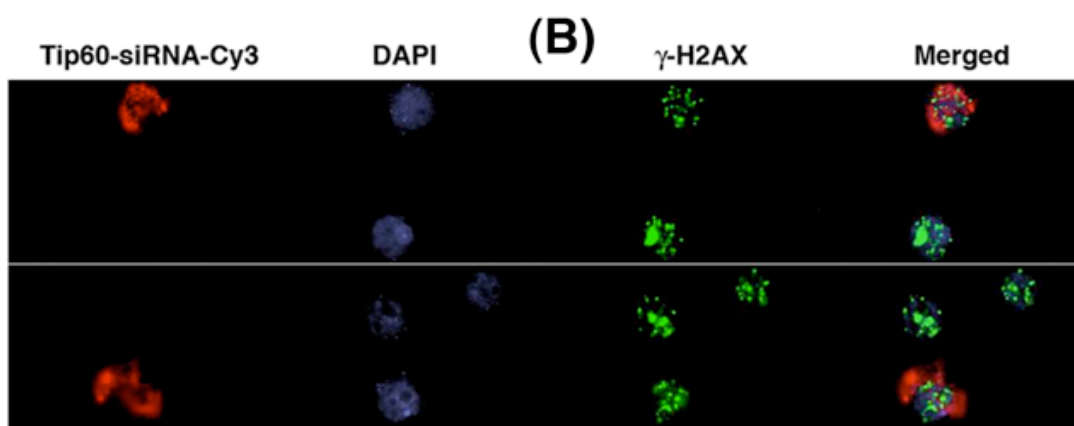
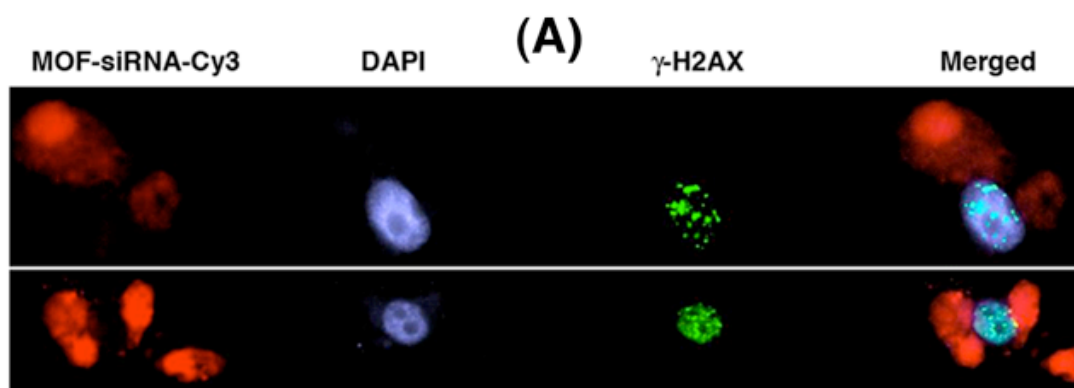
Supplementary Fig. 10. Cells with and without depletion of hMOF were treated with mitomycin C and examined for sister chromatid exchange frequency at metaphase by the described procedure (3, 4).

References:

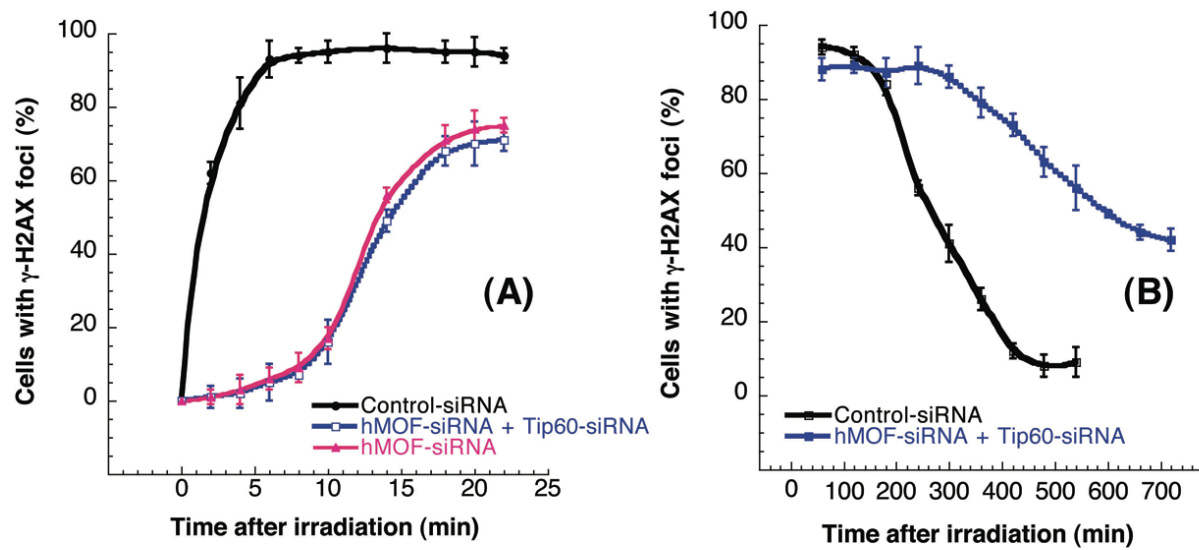
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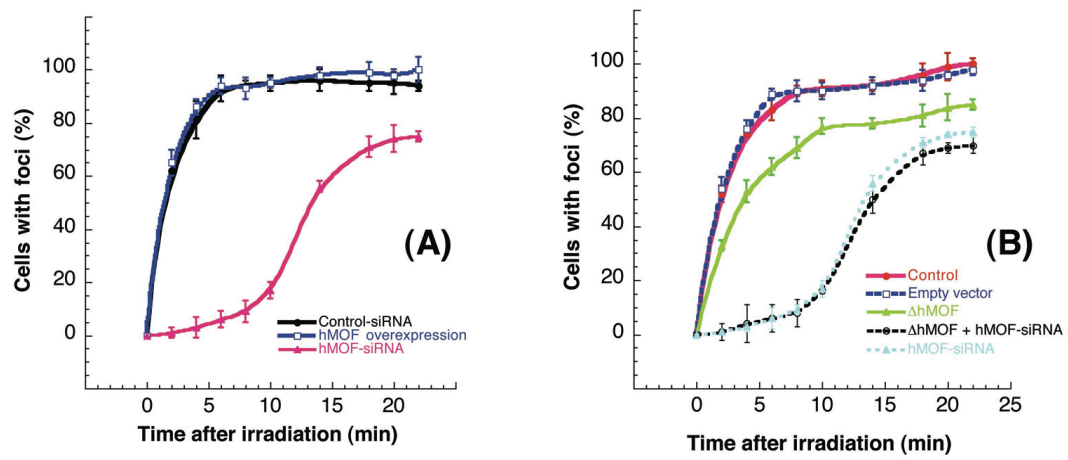
Supp Fig. 1



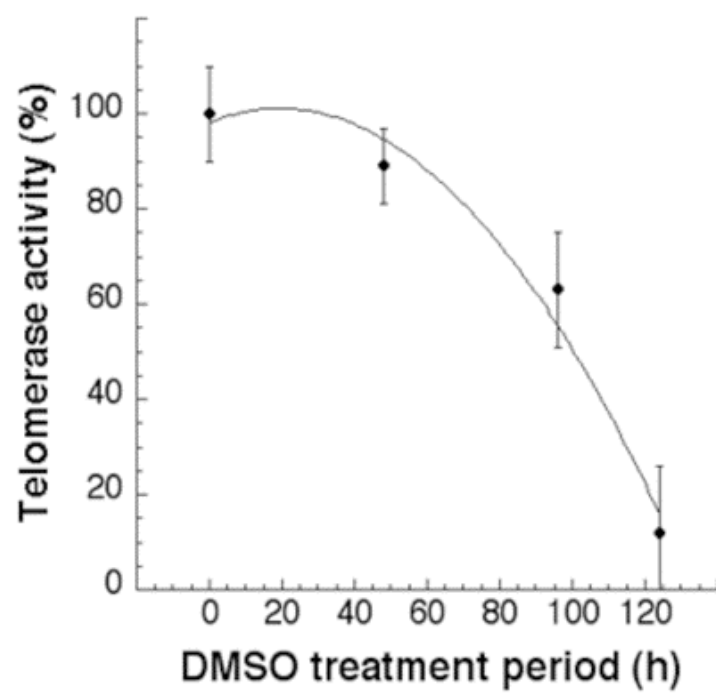
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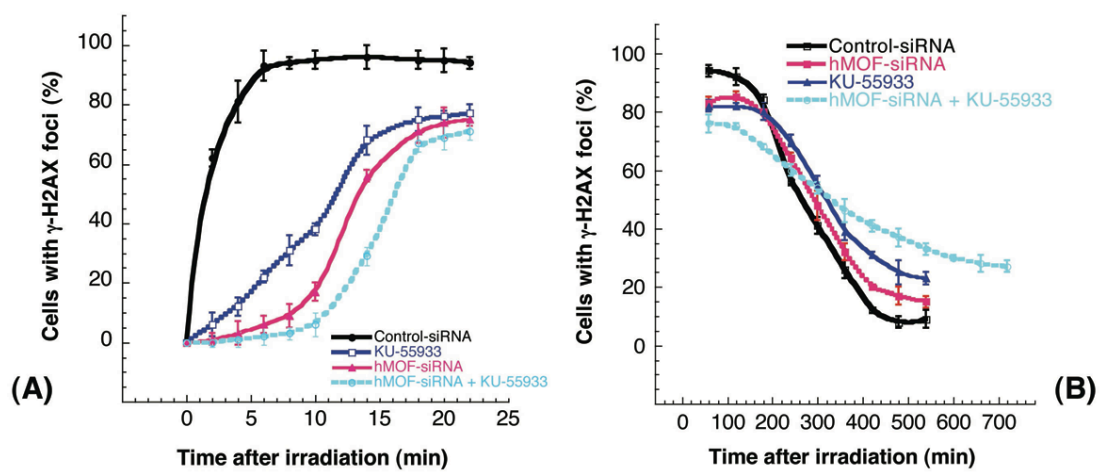
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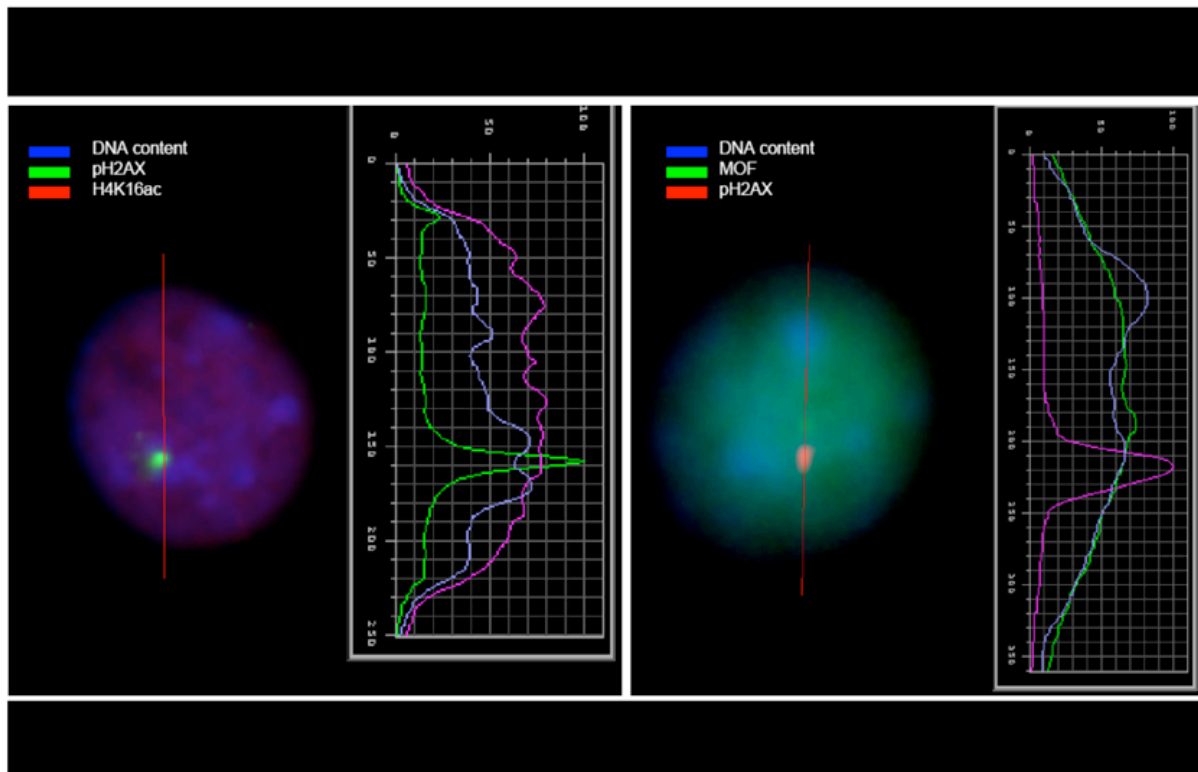
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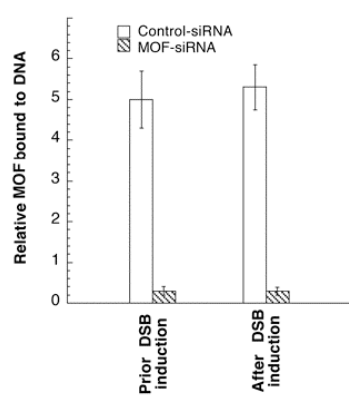
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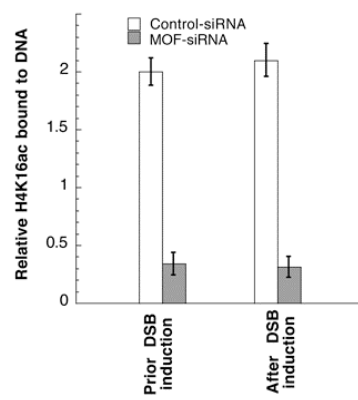
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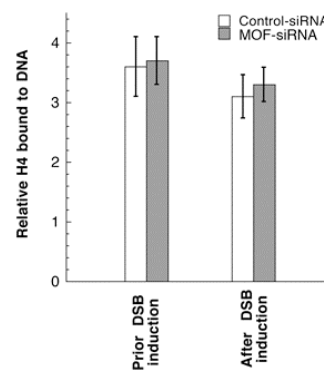
Supp Fig. 7



(A)

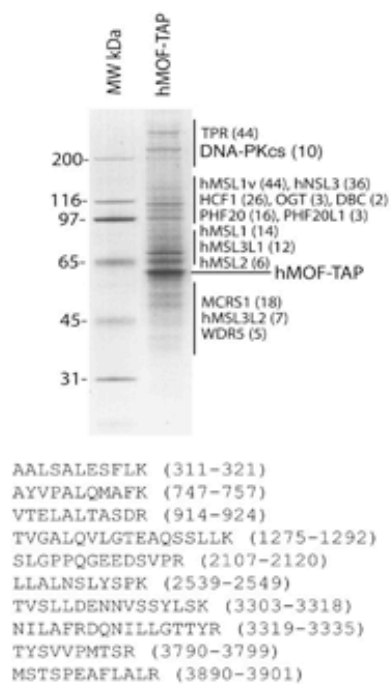


(B)

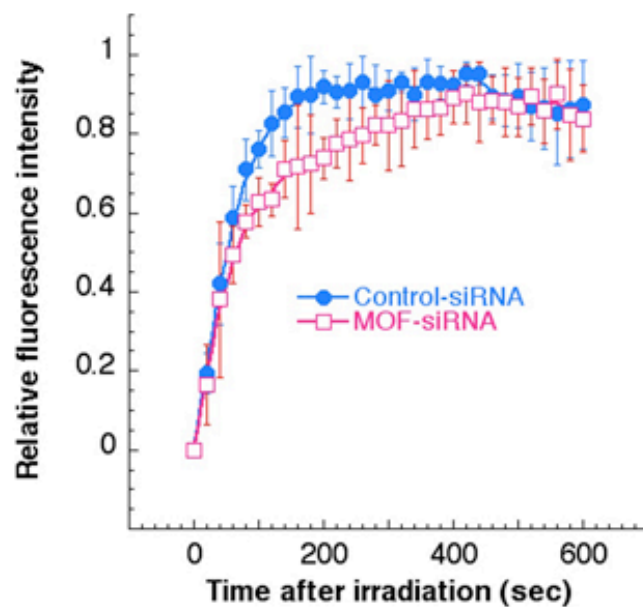


(C)

Supp Fig. 8

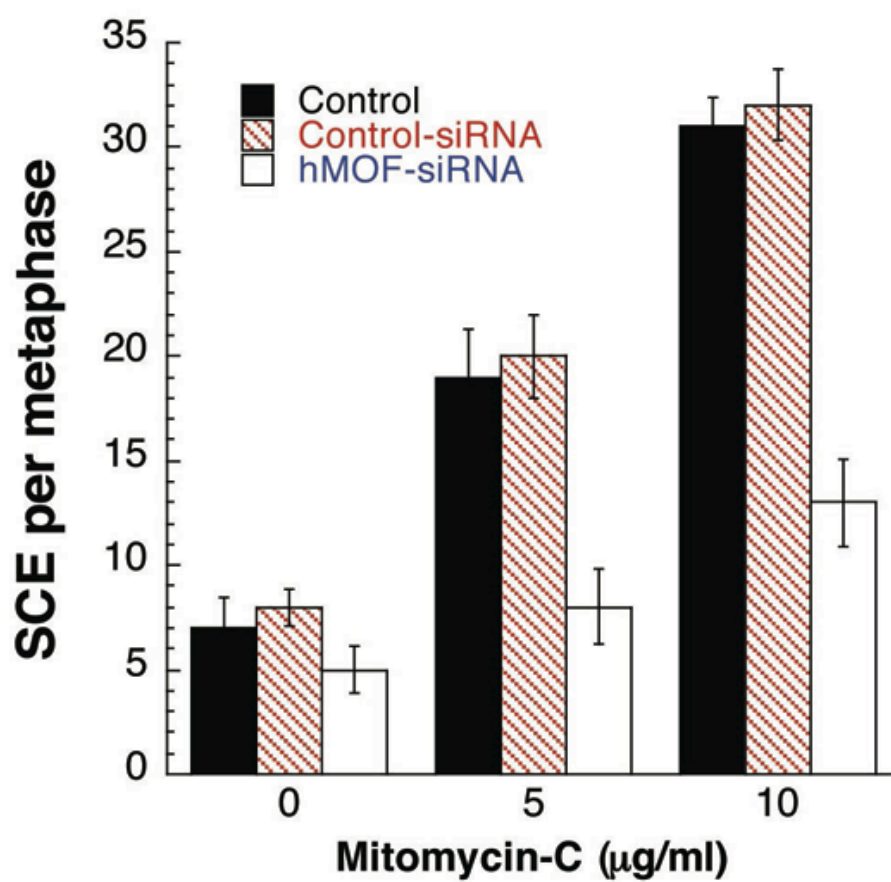


(A)



(B)

Supp Fig. 9



Supp Fig. 10