

# **DNA damage invokes mitophagy through a pathway involving Spata18**

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## **Supplementary Figures and Figure Legends**

**FigureS1. Effects of IR on cell proliferation and mitophagy.**

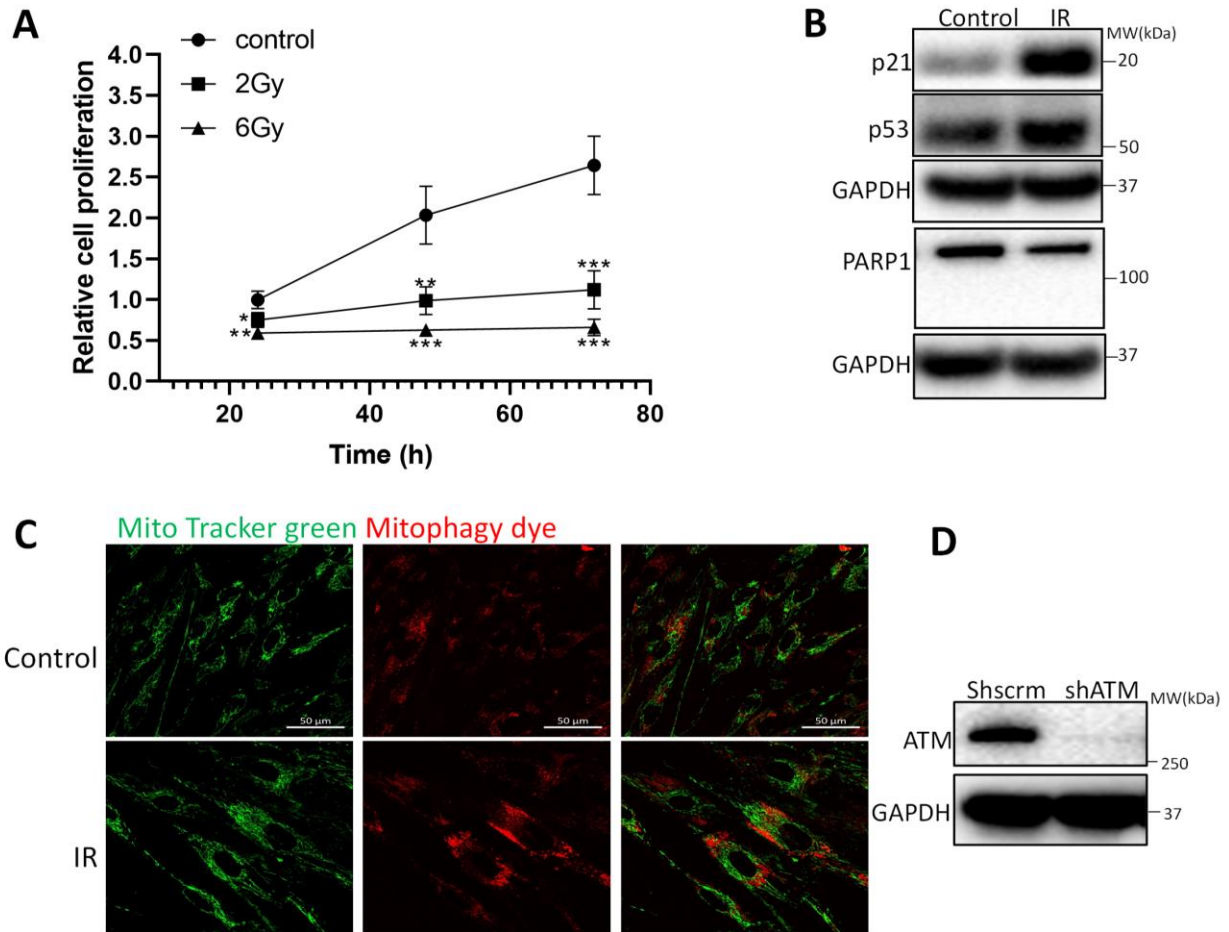
**FigureS2. Changes of pathways in cerebellum, cerebrum, heart and liver after IR.**

**FigureS3. Quantification of different mitophagy regulation proteins.**

**FigureS4. Effects of Spata18 KD on mitophagy after DNA damage.**

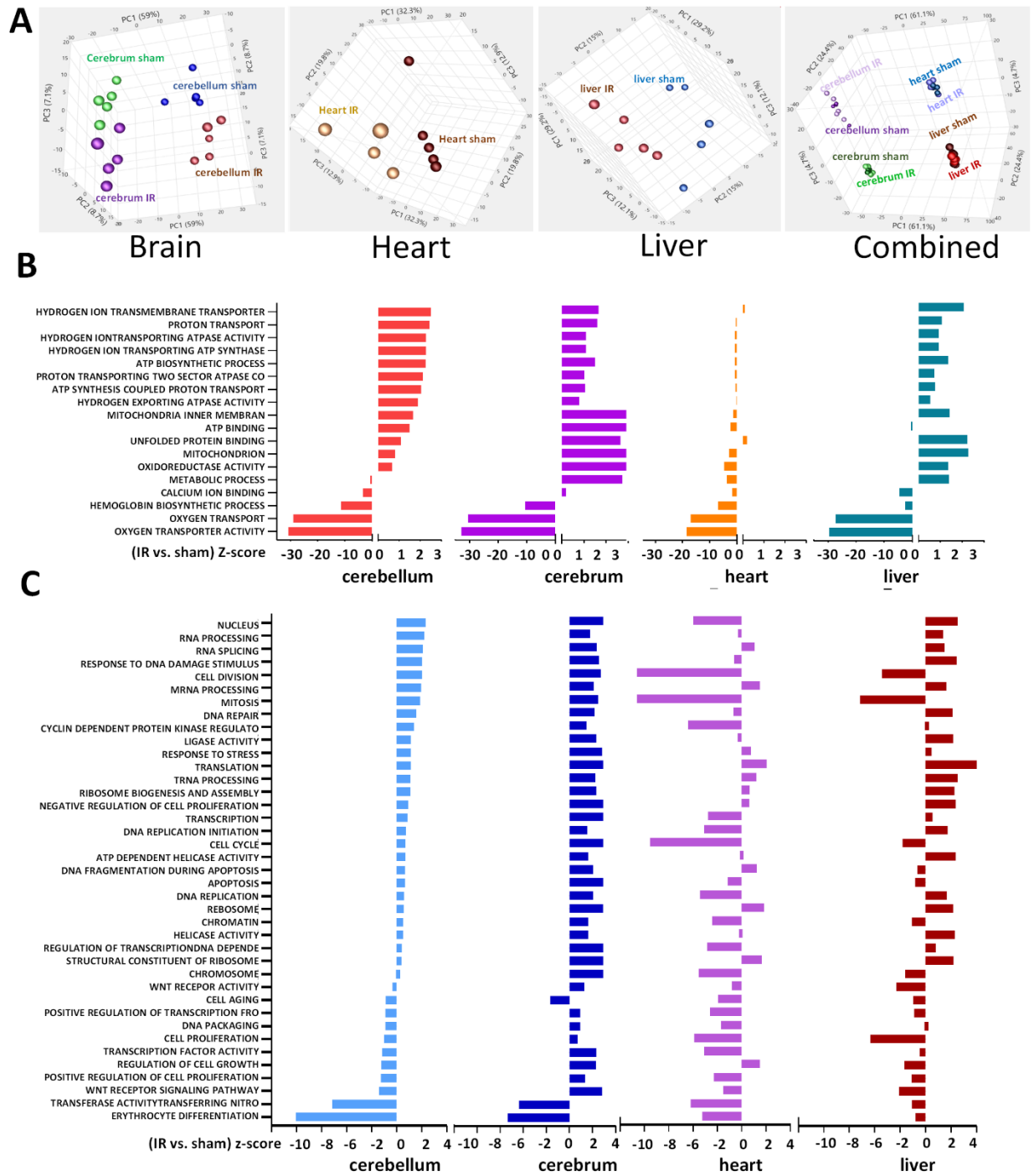
**FigureS5. Effects of Spata18 KD on basal DNA damage and DNA repair after IR.**

**Figure S1**



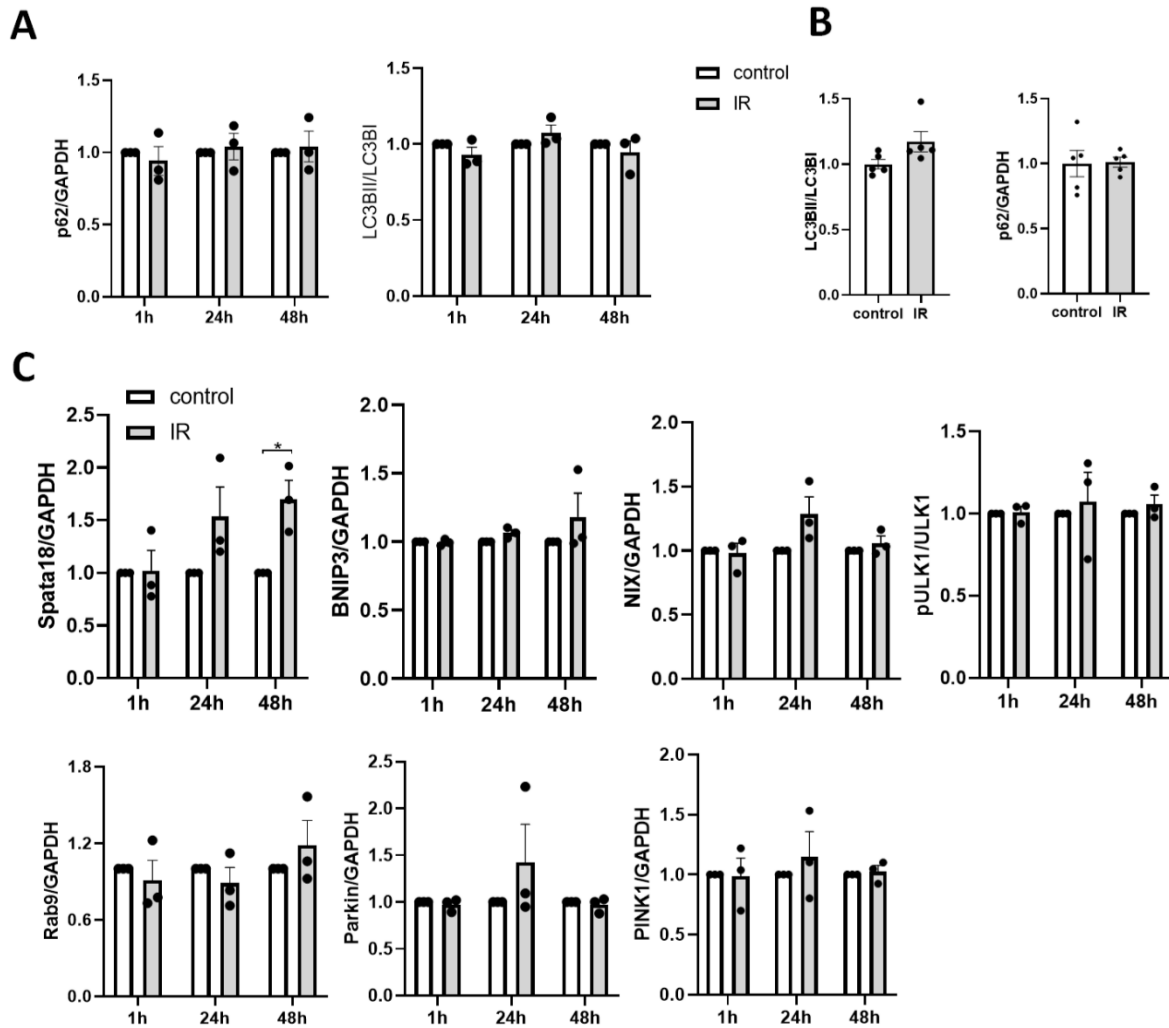
**FigureS1. Effects of IR on cell proliferation and mitophagy. (A)** Cell proliferation of HFLF after 2Gy or 6Gy IR. Data shown is normalized to control cells at 24h post radiation and the graph was made based on the average of three independent biological experiments. **(B)** Western blot showing the protein level of p21, p53 and PARP1. Samples were collected from HFLF cells 48h after 6Gy IR. **(C)** Representative images showing mitophagy induction 48h after 6Gy IR. The scale bars represent 50  $\mu$ m. **(D)** Western blot showing the protein level ATM. N=3, mean  $\pm$  S.E.M. \* P < 0.05, \*\* P < 0.01, \*\*\* P < 0.001.

**Figure S2**



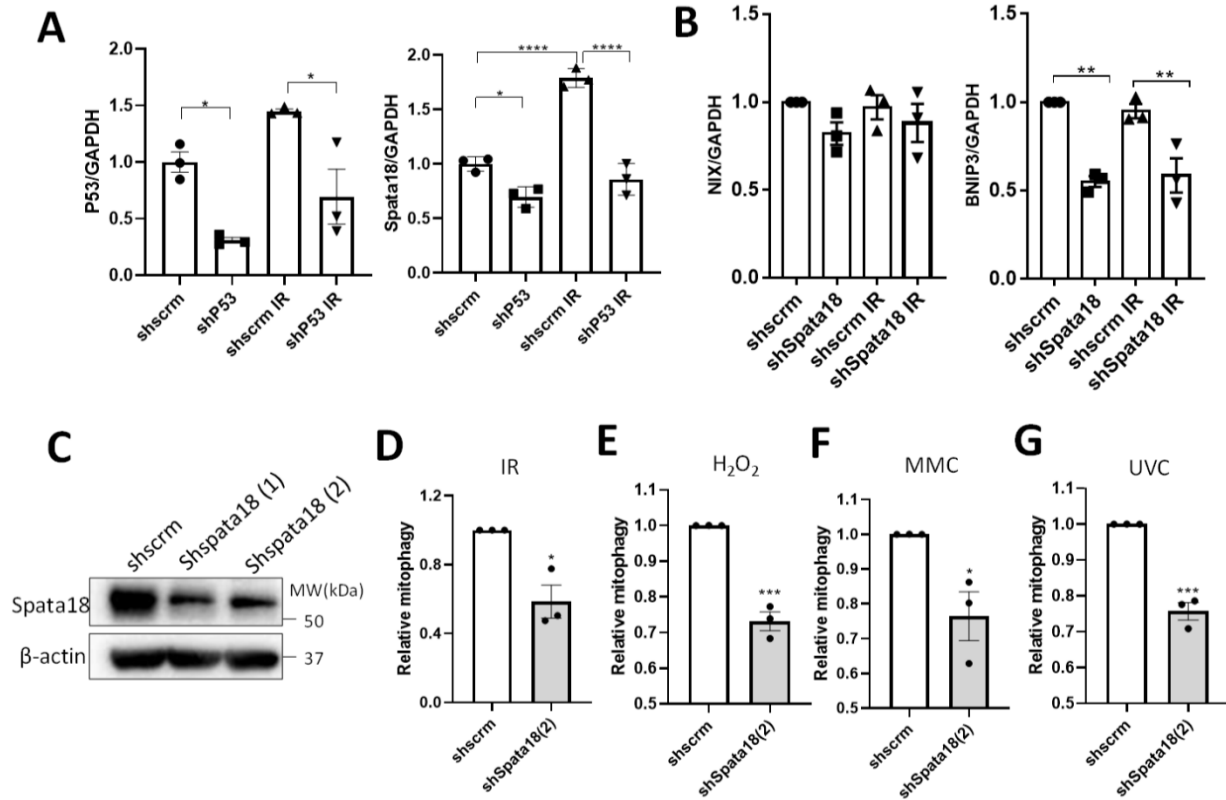
**FigureS2. Changes of pathways in cerebellum, cerebrum, heart and liver after IR. (A)** 3D scatter plot of Principal Component Analysis (PCA) results. Summary of significantly altered pathways related to **(B)** mitochondria and metabolism and **(C)** DNA damage response.

**Figure S3**



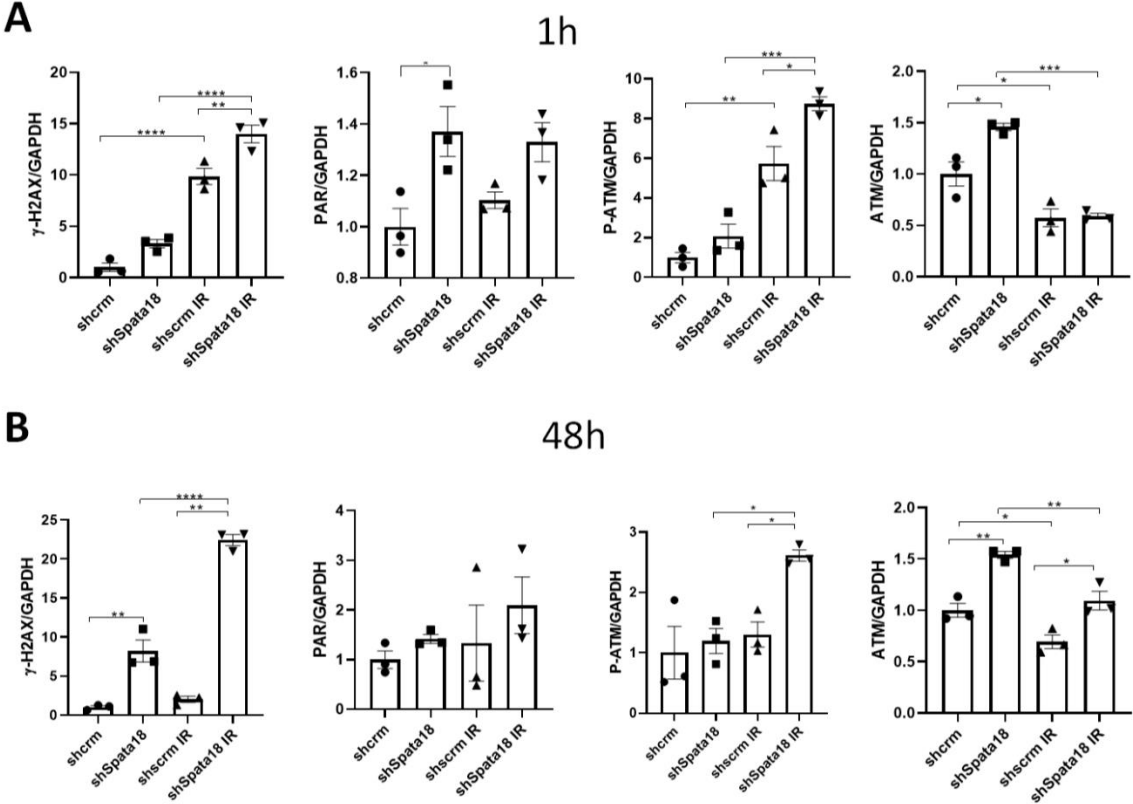
**FigureS3. Quantification of different mitophagy regulation proteins.** Quantification of p62 and LC3BII/LC3BI of HFLF cells in (A) and of mice cerebellum in (B) after 6Gy IR. (C) Quantification of Spata18, PINK1, Parkin, NIX, BNIP3, p-ULK1, ULK1 and RAB9a in HFLF cells after 6Gy IR. N=3, mean  $\pm$  S.E.M. \* P < 0.05.

**Figure S4**



**FigureS4. Effects of Spata18 KD on mitophagy after DNA damage.** (A) Quantification of p53 and Spata18 protein levels in p53 KD cells 48h after 6Gy IR. (B) Quantification of NIX and BNIP3 protein levels in Spata18 KD cells 48h after 6Gy IR. (C) Western blot showing the protein level of Spata18 in HFLF targeted by scrambled, shSpata18(1) or shSpata18(2). (D-G) Relative mitophagy levels of HFLF cells with Spata18 KD by shSpata18(2) after the treatment with 6Gy IR in D, 0.25 mM H<sub>2</sub>O<sub>2</sub> in E, 2 μM MMC in F and 10 J/m<sup>2</sup> UVC in G. Three independent biological repeats. Data: mean ± S.E.M. \* P < 0.05, \*\* P < 0.01, \*\*\* P < 0.001.

**Figure S5**



**FigureS5. Effects of Spata18 KD on basal DNA damage and DNA repair after IR.** Quantification of  $\gamma$ -H2AX, PAR, P-ATM and ATM of scrambled and shSpata18 HFLF cells 1h (A) and 48h (B) after IR. N=3, mean  $\pm$  S.E.M. \* P < 0.05, \*\* P < 0.01, \*\*\* P < 0.001.