



Article

Iron-Storage Protein Ferritin is Upregulated in Endometriosis and Iron Overload Contributes to a Migratory Phenotype

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Methods

MTT Assay

To analysis cell viability, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-tetrazolium bromide (MTT) assay was carried out. Cells were plated in a 96-well plate (5×10^4 cells/well) and incubated overnight to allow adhesion of the cells. After treatment with FeSO_4 and FAC, the cells were incubated for 24 h. Then, 50 μL of MTT solution (1 mg/mL stock solution) was added into each well and the plate was incubated for another 3 h. Formazan crystal formed in the cells was dissolved in DMSO after removal of culture medium. The absorbance at 540 nm was measured in a microplate reader (SpectraMax; Molecular Devices, Sunnyvale, CA, USA).

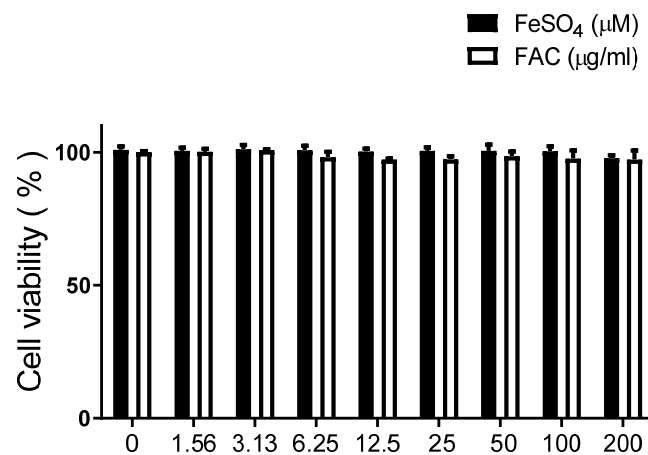


Figure S1. Effect of FeSO_4 and FAC on viability of 12Z cells. 12Z cells were treated with various concentration of FeSO_4 and FAC for 24. MTT assay were performed.

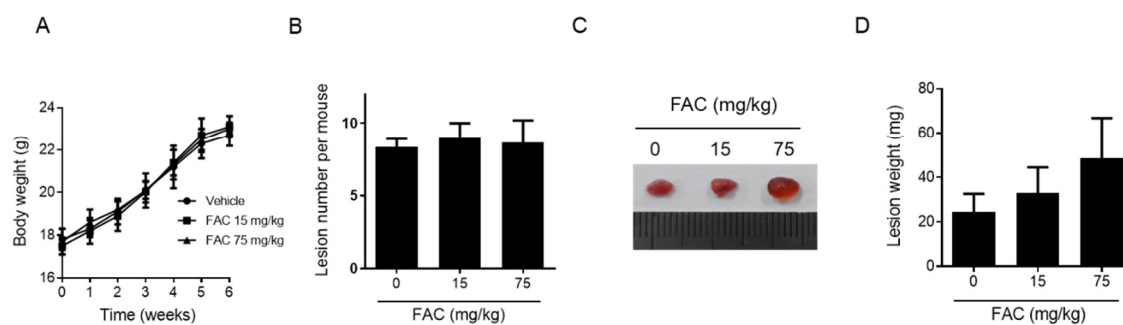


Figure S2. Effect of FAC on endometriotic lesion formation in female BALB/c mice. Female BALB/c mice were intraperitoneally administered with vehicle (0.05% CMC), FAC (15mg/kg) or FAC (75 mg/kg) three times per week for 5 weeks. **(A)** Body changes were measured every week. **(B)** Numbers of endometriotic lesions on the peritoneum and visceral organs. **(C)** Representative image of endometriotic lesions **(D)** Weight of endometriotic lesions.

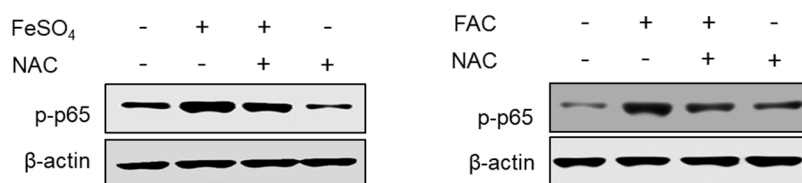


Figure S3. Effect of NAC on iron-induced activation of NFκB in 12Z cells. Human endometriotic 12Z cells were treated with iron (FeSO₄: 100 μM, FAC: 100 μg/ml) in the presence of NAC for 24 h. p-p65 expression was determined by Western blot analysis. β-Actin was used as loading control. Representative images of three independent experiments are shown.

