## SUPPLEMENTARY DATA

# Myeloperoxidase modulates hydrogen peroxide mediated cellular damage in murine macrophages. 

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[^0]Table S1. Mus primer sequences used for qPCR.

| Gene | Forward sequence $\left(5^{\prime}-3^{\prime}\right)$ | Reverse sequence $\left(5^{\prime}-3^{\prime}\right)$ |
| :---: | :---: | :---: |
| TATA-box binding protein (TBP) | CACAGGAGCCAAGAGTGAAGA | CACAAGGCCTTCCAGCCTTA |
| 18S ribosomal RNA (18S) | GTAACCCGTTGAACCCCATT | CCATCCAATCGGTAGTAGCG |
| Mus beta-2 microglobulin (B2M) | TGGTCTTTCTGGTGCTTGTC | GGATTTCAATGTGAGGCGGGT |
| Heme oxygenase 1 (HMOX1) | ACAGCCCCACCAAGTTCAAA | TCTGCAGGGGCAGTATCTTG |
| Superoxide dismutase 2, mitochondrial (SOD2) | ACAACTCAGGTCGCTCCTCAG | GATAGCCTCCAGCAACTCTCC |
| Glutathione Peroxidase 1 (GPx1) | CCACCGTGTATGCCTTCTCC | AGAGAGACGCGACATTCTCAAT |
| Glutathione S- <br> Transferase Pi 1 (GSTP1) | ATGCCACCATACACCATTGTC | GGGAGCTGCCCATACAGAC |
| Glutamate-Cysteine Ligase Catalytic Subunit (GCLc) | GGACAAACCCCAACCATCC | GTTGAACTCAGACATCGTTCCT |
| Glutamate-Cysteine Ligase Modifier Subunit (GCLm) | CTTCGCCTCCGATTGAAGATG | AAAGGCAGTCAAATCTGGTGG |
| NAD(P)H Quinone <br> Dehydrogenase 1 (NQO1) | AGGATGGGAGGTACTCGAATC | TGCTAGAGATGACTCGGAAGG |
| Glutathione synthetase (GS) | GGTATCTTCCCTCAGCAGCCTT | GCTTCCATTCCCACACTCCAAA |



Figure S1: Effect of the glucose/GO/MPO enzymatic system on J774A. 1 metabolic activity.
J774A. 1 cells $\left(1 \times 10^{5}\right)$ were treated with $\mathrm{GO}\left(0-200 \mathrm{mU} \mathrm{mL}^{-1}\right)$ in HBSS with or without glucose ( 5.6 mM ), in the absence and presence of MPO ( $0-100 \mathrm{nM}$ ) for either (a) 1 h or (b) 4 h before reincubation in MTS containing cell media for 4 h . Data are expressed as the percentage of metabolic activity compared to the non-treated group and represent the mean $\pm$ S.E.M from 3 independent experiments. * shows a significant difference ( $p<0.05$ ) compared to the non-treated cells; \# shows a significant difference ( $p<0.05$ ) compared to the glucose/GO group without MPO, by a 2-way ANOVA with a Tukey's multiple comparison test.


Figure S2: Effect of the glucose/GO/MPO enzymatic system on intracellular thiols in J774A. 1 cells.

J774A. 1 cells $\left(5 \times 10^{5}\right)$ were treated with $\mathrm{GO}\left(50 \mathrm{mU} \mathrm{mL}^{-1}\right)$ in the absence and presence of MPO (20 and 50 nM ) in HBSS containing glucose ( 5.6 mM ) for 1 h before re-incubation in cell media for 24 h. Intracellular thiols were quantified using ThioGlo 1 and normalised to the total protein concentration measured by BCA assay. Data are shown as the fold change of thiols compared to non-treated group and represent mean $\pm$ S.E.M from three independent experiments. Analysis by 1-way ANOVA with Dunnett's multiple comparison test showed no significant changes compared to the non-treated control (HBSS).


Figure S3. Cytoplasmic Nrf2 and c-JUN are not altered in J774A. 1 cells on exposure to the glucose/GO/MPO enzymatic system.

J774A. 1 cells ( $2 \times 10^{6}$ ) were incubated in HBSS containing glucose ( 5.6 mM ) (black bars), with GO ( $50 \mathrm{mU} \mathrm{mL}^{-1}$ ), GO/MPO ( 50 nM ) or GO/MPO/SCN $(200 \mu \mathrm{M})$ for 1 h at $37^{\circ} \mathrm{C}$ before re-incubation in cell media for 24 h . The cytoplasmic protein was extracted using a commercial kit and $10 \mu \mathrm{~g}$ protein was loaded. $\beta$-actin was used as a loading control. Images are representative of 3 independent experiments (A). Panels $\mathbf{B}$ and $\mathbf{C}$ show the densitometry analysis of $\mathrm{Nrf2}$ ( $\mathbf{B}$ ) and c JUN (C) following normalization to $\beta$-actin. Data are expressed as the fold change compared to the respective non-treated group.


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