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Article

TLR4 Signaling and Heme Oxygenase-1/Carbon Monoxide Pathway Crosstalk Induces Resiliency of Myeloma Plasma Cells to Bortezomib Treatment

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Supplemental Material.

1. Supplemental Method

shHO-1 MM cells generation and XTT proliferation assay

U266 cells were silenced for HO-1 as previously described [1]. To assay their proliferation cells were plated in a 96-well plate and treated with 2 ug/mL LPS for 24 and 48 hours. XTT assay was then performed as already reported[2].

Supplemental Figures

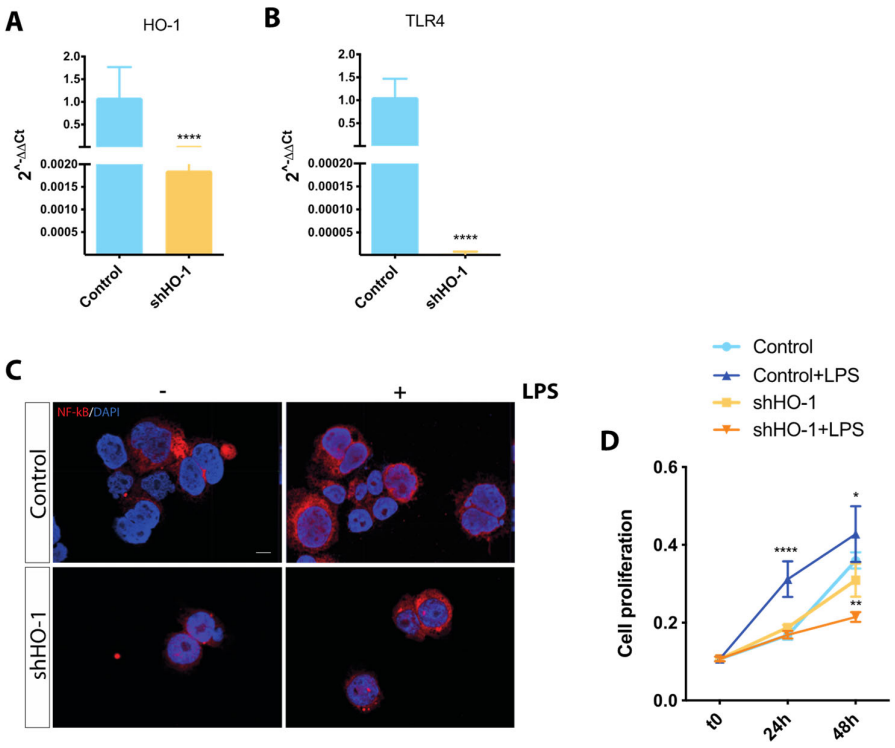

Figure S1. HO-1 knockdown impairs TLR4 expression and its signalling. (**A-B)** HO-1 and TLR4 expression analysis by qPCR analysis. shRNA against HO-1 efficiently mediate its knockdown. In parallel also TLR4 expression turns to be downregulated (**C)** NF-kB nuclear traslocation analysis by immunofluorescence. Upon HO-1 silencing, Nf-kB nuclear translocation turns to be imapired. (**D)** XTT assay on shHO-1 cells. Cells transfected with shRNA against HO-1 did not respond to LPS-induced proliferation as control cells.

Supplemental References

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2. Maugeri, G.; D'Amico, A.G.; Giunta, S.; Giallongo, C.; Tibullo, D.; Bucolo, C.; Saccone, S.; Federico, C.; Scollo, D.; Longo, A.; et al. Activity-Dependent Neuroprotective Protein (ADNP)-Derived Peptide (NAP) Counteracts UV-B Radiation-Induced ROS Formation in Corneal Epithelium. *Antioxidants (Basel)* **2022**, *11*, doi:10.3390/antiox11010128.