Supplementary Information

to

Metal homeostasis disruption and mitochondrial dysfunction in hepatocytes exposed to sub-toxic doses of zinc oxide nanoparticles

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Figure S1. Size distributions of a) ZnO-NP-FBS and b) ZnO-NP-silane preparations



a)

Figure S2. Measurement of SOD and CAT activities after a 6 hour incubation with Zn acetate, ZnO-NP-FBS or ZnO-NP-silane at 90 μ M. Results were expressed in enzymatic units/mg of protein as the average + SEM of 4 independent experiments. P>0.05



Figure S3. Quantification of acidic vesicular organelles with acridine orange staining. Results are presented as changes in mean of green fluorescence expressed as a percentage of the mean fluorescence in control cells (three independent experiments). Fluorescence measurements were carried out on a MoFlo cell sorter. ***: p<0.001



Figure S4. Quantitative PCR analysis of DRP1, MFN1 and MFN2 mRNA expression in HepG2 cells after a 6 hour incubation with Zn acetate, ZnO-NP-FBS or ZnO-NP-silane at 90 μ M. Results are presented as relative expression changes after normalizing with HPRT and 36B4 mRNA. Each value represents the mean of relative expression +SEM of three independent experiments. P>0.05



Table S1. Quantitative real time polymerase chain reaction primer sequences

	Formeral	Deverage
gene	Forward	Reverse
Met1X	GCT TCT CCT TGC CTC GAA A	TGA CGT CCC TTT GCA GAT G
HMOX1	ATG ACA CCA AGG ACC AGA GC	GTG TAA GGA CCCA TCG GAG A
HSPA6	AGG AGG TGG AGA GGA TGG TT	TGT CCT CTT CGG GAA TCT TG
SOD1	TGG CCG ATG TGT CTA TTG AA	ACC TTT GCC CAA GTC ATC TG
SOD2	GTG GAG AAC CCA AAG GGG AG	GAA ACC AAG CCA ACC CCA AC
САТ	TTG CCA CAG GAA AGT ACC CC	TGA GGC CAA ACC TTG GTG AG
ZnT1	CGC AGC CAT GGG GTG TTG GG	CAT CGC CAG CGA CGA GGT CA
ZnT2	AAC TTT GGC TGG CAG AGA GC	TGG TGA AGG GTC AAC CCC AT
ZnT7	GTG CCT GAA CCT CTC TTT CG	CGG AAA TCA GCC TAA GCA G
Zip1	GCC AGG AGC TAA CCA TGA AG	ATG GCC AGG ATG AAC TCT TG
Hamp	ACC AGT GGC TCT GTT TTC CC	CAG ATG GGG AAG TGG GTG TC
GCLM	CCT CCT GCT GTG TGA TGC CAC C	CGT GCG CTT GAA TGT CAG GAA TGC
EGR1	TAC GAG CAC CTG ACC GCA GAG TC	CAT GAA GCT CCT CGC TGT GCG TG
MTF1	CGA AGG AGA AGC CAT TTG AG	ATT TGC TGC AGC CTT CAG AT
DRP1	TCA CCC GGA GAC CTC TCA TT	TCT GCT TCC ACC CCA TTT TCT
MFN1	TGT TTTGGT CGG AAA CTC TG	CTGTCTGCGTACGTCTTC CA
MFN2	TAT CCA CTT CCC TCC TCC CG	CCACCAGTCCTGACT TCA CC
HPRT	ATG GAC AGG ACT GAA CGT CTT GCT	TTG AGC ACA CAG AGG GCT ACA ATG
36B4	GAA ATC CTG GGT GTC CGC AAT GTT	AGA CAA GGC CAG GAC TCG TTT GTA