SUPPLEMENTARY MATERIAL - FIGURE LEGENDS

<u>Fig. 1.</u> CO at 100 μ M does not confer protection against astrocytic apoptosis. Primary cultures of astrocytes cultured in 24 well-plates were pre-treated with 100 μ M of CO for 3h, following apoptosis induction by 18h exposure to (A, B) the pro-oxidant, t-BHP (from 0 to 280 μ M). The apoptotic hallmarks were assessed by flow cytometry. In (A) the percentage of cells presenting high mitochondrial potential, detected by DiOC₆(3), is expressed. In (B) the percentage of cells containing intact plasma membrane (viable cells) is presented, assessed with PI fluorochrome. All values are mean ± SD, n = 4 (A, B).

Fig. 2. CO confers protection against apoptosis in delayed periods of time. Primary cultures of astrocytes cultured in 24 well-plates were pre-treated with 50 μ M of CO for 3h, following apoptosis induction by the pro-oxidant, *t*-BHP (160 μ M) for 48 h. The percentage of viable cells (assessed by PI) is presented at 0, 6, 12, 24 and 48 h after *t*-BHP addition,. All values are mean ± SD, n = 3.

Fig. 3. ANT glutathionylation in mitochondria treated with diamide. Isolated non-synaptic mitochondria were pre-treated with CO at 10 or 50 μ M or with GSSG at 1 μ M for 15 minutes, followed by diamide (100 μ M) incubation at 37°C for 30 minutes; then glutathionylated proteins (α -GSH) were immunoprecipitated and ANT was immunodetected by Western blot from the immunoprecipitated proteins. This experiment has been repeated three times, with similar results.





t-BHP (μM)





Figure 3

