Supplementary Data



SUPPLEMENTARY FIG. S1. Disulfram induced S-glutathionylation in liver lysate and subcellular compartment, including cytosol (Cyto), endoplasmic reticulum (ER) and mitochondria (Mito) from Gstp1/p2^{+/+} and $Gstp1/p2^{--}$ mice. (A) Liver lysates were freshly pre-pared from $Gstp1/p2^{++}$ and $Gstp1/p2^{--}$ mice. Thirty micrograms of control or disulfiram-treated (10 μM , 30 min at 37°C) proteins was separated by SDS-PAGE under nonreducing conditions and blotted for anti-PSSG. Based on this result, protein bands of interest, defined as those with Sglutathionylation levels that quantitatively differed between $Gstp1/p2^{+++}$ and $Gstp1/p2^{+--}$ mice (see *arrows*), were excised from the Brilliant Blue G-colloidal stained gel and subject to digestion. The peptides were further analyzed and identified by mass spectrometry at the Proteomics Core Facility of the Medical University of South Carolina. (B) Liver tissue homogenates were treated with $10 \,\mu M$ disulfiram for 30 min at 37°C and free thiols were blocked by adding 5 mM NEM for 30 min. Subcellular fractionation was performed as described in "Materials and Methods" and proteins were separated by SDS-PAGE under nonreducing conditions and blotted for anti-PSSG. GSTP, glutathione S-transferase Pi; NEM, N-ethylmaleimide; PSSG, protein S-glutathionylation; SDS-PAGE, sodium dodecyl sulfate-polyacrylamide gel electrophoresis.