**Chemoproteomics analysis of LPS-stimulated macrophages (Figure 3, Figures S4-S7)**

We used SAMPEI to map protein modifications induced during the response of mouse RAW264.7 macrophage cells to lipopolysaccharide (LPS), a potent inducer of macrophage activation and differentiation that involves extensive protein and metabolic signaling. Cell proteomes were extracted, proteolyzed using trypsin, and fractionated using SCX chromatography prior to LC/MS analysis. Spectra were analyzed using X!tandem for conventional database search and generation of target spectra, followed by SAMPEI analysis for unbiased identification of modifications.

In addition, spectra were re-analyzed using X!Tandem with variable 73.0051 and 89.0000 Da modification on carbamidomethylated cysteine residues.

Raw MS data are publicly available via the PRIDE repository with dataset identifiers PXD019853.

**Files**

The dataset contains the following files:

- RAW264p7_LPS_tryp_SCX_all.txt: X!tandem output from MS raw data
- RAW264p7_LPS_tryp_SCX_SAMPEI_input.txt: target spectra submitted to SAMPEI.
- RAW264p7_LPS_tryp_SCX_SAMPEI_output.txt: SAMPEI output, containing the identified modified peptides.
- RAW264p7_LPS_tryp_SCX_SAMPEI_var130_var146.txt: X!Tandem output with peptides identified (FDR<0.01) setting variable +130 and +146 Da modification on carbamidomethylated cysteine residues