

Description of CPTAC dataset

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The experiment was conducted as part of the multilaboratory investigation of the Clinical Proteomic Technology Assessment for Cancer network of the National Cancer Institute (NCI-CPTAC) (Addona *et al.*, 2009). The investigation prepared controlled mixtures according to three mixing and digestion protocols. Here we focus on Study III, which contained sources of technical variation closest to a real-life experiment. 7 proteins were spiked in 3 biological replicates of human plasma at 9 levels of concentration (i.e. diluted 2-fold 8 times), resulting in 9 conditions. Therefore the study had a group comparisons design. Since all the proteins of interest change in concentrations between the conditions, the dataset can be used to evaluate the sensitivity but not the specificity of detecting changes in protein abundance.

The endogenous human plasma samples were mixed with an equal amount of proteins from heavy-labeled reference AQUA peptides (Gerber *et al.*, 2003). The samples were shipped to eight participating sites, which independently quantified the 7 spiked proteins in 4 technical replicates, resulting in a total of 108 mass spectrometry runs per site. Each protein was represented by 1-3 peptide, and each peptide by 2-3 transitions. Peaks were detected and quantified automatically with MultiQuant (Applied Biosystems). This manuscript focuses on the dataset from site 52. No reference transitions were missing by design, but 6%(2/33) of endogenous transitions and 6%(2/33) of reference transitions were missing in some runs for reasons other than the design.

References

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- Gerber, S. A., Rush, J., Stemman, O., Kirschner, M. W., and Gygi, S. P. (2003). Absolute quantification of proteins and phosphoproteins from cell lysates by tandem MS. *PNAS*, **100**, 6940–6945.