Data in Brief 5 (2015) 53–58

Contents lists available at ScienceDirect

Data in Brief

journal homepage: www.elsevier.com/locate/dib

Data Article

SILAC-based quantification of changes in protein tyrosine phosphorylation induced by Interleukin-2 (IL-2) and IL-15 in T-lymphocytes

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ARTICLE INFO

Article history:
Received 18 June 2015
Received in revised form
10 August 2015
Accepted 13 August 2015
Available online 22 August 2015

Keywords:
SILAC
Interleukin
Cell signaling
Phosphotyrosine
T-lymphocytes

ABSTRACT

This data article presents the first large-scale quantitative phosphoproteomics dataset generated to decipher the signaling networks initiated by IL-2 and IL-15 in T-lymphocytes. Data was collected by combining immunoprecipitation of tyrosine phosphorylated proteins and TiO_2-based phosphopeptide enrichment with SILAC-based quantitative mass spectrometry. We report all the proteins and phosphotyrosine-containing peptides identified and quantified in IL-2- and IL-15-stimulated T-lymphocytes. The gene ontology analysis of IL-2 and IL-15 effector proteins detected in the present work is also included. The data supplied in this article is related to the research work entitled "Simultaneous dissection and comparison of IL-2 and IL-15 signaling pathways by global quantitative phosphoproteomics" [1]. All mass spectrometry data have been deposited in the ProteomeXchange with the identifier PXD001129.

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http://dx.doi.org/10.1016/j.dib.2015.08.007

Specifications Table

Subject area	Cell signaling and immunology
More specific subject area	Interleukin signaling and quantitative phosphoproteomics
Type of data	Mass spectrometry (MS) data
How data was acquired	MS data was acquired in a Q-Exactive (Thermo) mass spectrometer.
Data format	Raw (*raw), excel files (.xlsx)
Experimental factors	Kit225 T-cells were grown in light (Arg0/Lys0), medium (Arg6/Lys4) and heavy (Arg10/Lys8) media. Differentially SILAC-labeled T-cells were kept unstimulated, treated with IL-2 or stimulated with IL-15, respectively prior cell lysis.
Experimental features	After stimulation, cells were lysed and protein extracts derived from the three different experimental conditions were combined and enriched in tyrosine phosphorylated proteins using two antibodies. Immune complexes were fractionated in a SDS-PAGE and in-gel digested using trypsin. Resulting peptides were either directly analyzed by LC-MS/MS or enriched in phosphorylated peptides using TiO_2 beads prior MS analysis.
Data source location	Odense, Denmark
Data accessibility	All MS data presented in this article are deposited in the ProteomeXchange with the identifier PXD001129 (http://proteomecentral.proteomexchange.org/datastet/PXD001129). List of all proteins identified in each of the two replicas performed are provided in Supplementary material linked with this article.

Value of the data

- The first simultaneous dissection of the signaling pathways triggered by IL-2 and IL-15 in CD4+ T-cells provides extensive data that allows discerning between the proteins that are regulated or not by tyrosine phosphorylation upon cytokine stimulation.
- The detection of numerous cytokine-dependent and – independent tyrosine phosphorylation events enables constructing a more precise molecular snapshot of the ongoing events on T-cells treated with IL-2 and IL-15.
- The identification of previously not reported phosphorylated tyrosine residues corresponding to distinct proteins serves as the starting point to characterize their biological relevance.

1. Data, experimental design, materials and methods

In the present work we provide the data generated to unveil the signaling pathways initiated by IL-2 and IL-15 in T-lymphocytes [1]. We include two tables containing quantitative and qualitative information about all the proteins and phosphotyrosine (pY)-containing peptides identified and quantified in the pY-immune complexes isolated from IL-2- and IL-15-treated T-lymphocytes, as well as the gene ontology analysis was performed.

To assess the signaling networks initiated downstream of IL-2/IL-2R and IL-15/IL-15R complexes in T-lymphocytes, we followed the experimental workflow shown in Fig. 1. (A) Kit225 T-cells were grown in media containing either light (0/0), medium (6/4) or heavy (10/8) version of arginine and lysine until their proteome was completely labeled. Then, cells grown in light media were kept unstimulated and thus served as control whereas cells grown in medium and heavy media were stimulated with IL-2 and IL-15, respectively. After protein extraction, differentially labeled cell lysates were combined in 1:1:1 ratio. (B) Tyrosine phosphorylated proteins were enriched using phospho-specific antibodies and (C) fractionated on a SDS-PAGE. After protein in-gel digestion and peptide extraction, resulting peptides where either (D) enriched in phosphopeptides using TiO_2 beads prior mass spectrometry (MS) analysis or (E) directly analyzed by a QExactive mass spectrometer. (F) Acquired raw mass spectra data were analyzed using the MaxQuant software and further data analysis was performed using David bioinformatics tools.