Integration of proteomics and metabolomics data in a novel cellular knock out model of methylmalonic acidemia

Michele Costanzo[1], Marianna Caterino[1], Armando Cevenini[1], Vincent Jung[2], Ida Chiara Guerrera[2], Margherita Ruoppolo[1]

[1] Dipartimento di Medicina Molecolare e Biotecnologie Mediche, Università degli Studi di Napoli "Federico II", Naples, Italy
[2] Plateforme Protéomique 3P5-Necker, Structure Fédérative de Recherche Necker and Université Paris Descartes, INSERM U24, Paris, France

Corresponding Author:
Michele Costanzo[1]
Via Pansini, 5, 80131, Naples, Italy
Email address: michele.costanzo@unina.it

Background. Methylmalonic acidemia is a rare inborn error of metabolism caused by mutations in methylmalonyl–CoA mutase (MUT) gene. As intermediate of propionate metabolism, MUT converts methylmalonyl–CoA into succinyl–CoA, which enters the Krebs cycle. Downstream MUT deficiency, methylmalonic acid accumulates in body fluids as biomarker of disease. The long-term complications of the disease can include cognitive and neurological impairment, chronic kidney disease, liver failure, and death.
Methods. In order to create a valid cellular model to study the disease, MUT gene was knocked out (KO) in HEK293 cell line by using CRISPR-CAS9 technology. Methylmalonic acid was measured in MUT-KO and wild type (WT) cells by multiple reaction monitoring. A quantitative proteomics analysis was carried out using a label-free mass spectrometry-based approach. Data were processed using MaxQuant software. Moreover, a targeted metabolomics analysis was performed in order to measure an entire panel of amino acids and acylcarnitines.
Results. Methylmalonic acid resulted increased in KO cells if compared with WT ones. The proteomic dataset showed a number of 69 differentially expressed proteins, of which 39 down-regulated and 30 up-regulated in the MUT-KO condition. Gene Ontology analysis revealed an enrichment in energy and lipid metabolism categories. The variations in the metabolomic profile are indicative of alterations in fatty acid oxidation processes and lipid metabolism.