Plan Overview

A Data Management Plan created using DMPTool

**Title:** Proteomics and degradomics applied to the study of the effects of mannose in mutant and non-mutant B-RAF melanoma cell lines

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**Creator:** André Zelanis - **ORCID:** 0000-0003-3066-6527

**Affiliation:** Universidade Federal de São Paulo (UNIFESP)

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**Template:** Digital Curation Centre

**Project abstract:**

The high energy demand imposed by the oncogenic process has important consequences on the metabolism of transformed cells, as well as having a significant impact on diagnostic methods and therapies associated with the treatment of neoplasias. Mannose is an epimer of glucose and therefore is absorbed by the same transporters as glucose. Recent work using tumor cell lines has shown that this glucose epimer accumulates in cells in the form of the intermediate mannose-6-phosphate (M6P), due to the low expression of the enzyme phosphomannose isomerase (PMI), which converts M6P to fructose-6-phosphate (F6P), an intermediary of the glycolytic pathway. This feature has a significant impact on glucose metabolism through the glycolytic pathway, the tricarboxylic acid cycle, and the pentose phosphate pathway. Recent data from our group indicate that the metabolic stress resulting from the administration of mannose to cell lines of metastatic human melanoma results in a decrease in the growth rate, as well as in the activation of lysosomal degradation pathways and, possibly, of autophagy. In this context, the main objective of this project is to evaluate the effects of mannose administration on the proteome of malignant human melanoma cells, as well as on the repertoire of activated protease substrates (degradome) in human melanoma cell lines with two distinct metabolic/mitogenic phenotypes (B-RAF mutant, A375 lineage, and non-mutant, WM 1366 lineage), correlating the results obtained with possible biological pathways relevant to the oncogenic process. Furthermore, it is expected to evaluate the effect of mannose administration on the development of tumors in vivo in experimental animals, correlating the results with those verified for cell cultures, as well as with the expression of the PMI enzyme. From a therapeutic point of view, the results obtained will also allow the elucidation of fundamental molecular aspects that may support the increment of clinical intervention strategies using this hexose (mannose) associated with therapies.
already used for the treatment of human melanoma.

**Start date:** 01-02-2022

**End date:** 06-02-2025

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Proteomics and degradomics applied to the study of the effects of mannose in mutant and non-mutant B-RAF melanoma cell lines

Data Collection

What data will you collect or create?

Experimental approaches will generate 2 main types of data:
(1) Data regarding the profile of proteins from melanoma cells (B-RAF mutant or not, cell lines A375 and WM1366, respectively) in culture conditions with (a) glucose 25 mM, (b) glucose + mannose 25 mM (final). Sucha data will be collected after shotgun proteomics experiment derived from these two culture conditions;
(2) Degradomics data derived from the culture conditions described above but subjected to N-terminal protein enrichment technique using Terminal Amine Isobaric Labeling of Substrates (TAILS);

The mass spectrometric data format will be made available under the original vendor (RAW) format as well as will be converted to the universal data format 'mzML'. Assembled contig data will be made available in "fasta" format. Proteomics and data will be submitted to public repositories such as the ProteomeXchange Consortium (http://www.proteomexchange.org/) In addition, for bioinformatic analysis purposes, data from the above repository might also be interrogated.

Data will be locally stored, in a Microsoft Windows server, located at the Functional Proteomics Laboratory, Federal University of São Paulo, São José dos Campos, Brazil. All the RAW files will be submitted to the ProteomeXchange public repository (http://www.proteomexchange.org/).

How will the data be collected or created?

The main objective of this project is to evaluate the effects of mannose administration on the proteome of human malignant melanoma cells (B-Raf mutant or not), with emphasis on processes regulated by the activity of proteases.

Proteomics data will be named after the experimental conditions used in this work, as well as the corresponding biological replicate, namely: Man_R1 (mannose, biological replicate 1), Glc_R1 (glucose, biological replicate 1), Glc_Man_R1 (glucose + manose, biological replicate 1).

Before submitting data to any public repository, each LC-MS/MS run will be evaluated based on the protein identification/quantitation which must display consistency among each biological replicate (i.e, Pearson correlation coefficient greater or equal than 0.7 for protein quantitative values).

Documentation and Metadata

What documentation and metadata will accompany the data?

The information on the dataset generated in this study (metadata) will be compiled in a spreadsheet, including information on RAW file size, software (and version) used for Peptide-to-Spectrum assignment, search parameters, and database size. This will be done in order to enable the reproducibility of our proteomics data analyses by any researcher worldwide.

Ethics and Legal Compliance

How will you manage any ethical issues?
This section is not applicable to our study.

How will you manage copyright and Intellectual Property Rights (IP/PR) issues?

Data will be stored locally, as described above, and will be made publicly available as soon as the publication is out (this is the only situation where data sharing might be postponed).

Storage and Backup

How will the data be stored and backed up during the research?

In addition to the public repositories mentioned above, data will be stored locally in a server located at the Functional Proteomics Laboratory, at the Institute of Science and Technology of the Federal University of São Paulo, at São José dos Campos, Brazil. A backup will be done on a weekly basis, using external hard drives.

How will you manage access and security?

Data in public repositories are available to the general public. Ideally, our local storage will mirror the public repositories.

Selection and Preservation

Which data are of long-term value and should be retained, shared, and/or preserved?

All data will be retained and used throughout the development of this and other research projects at the laboratory.

What is the long-term preservation plan for the dataset?

At this time (December 2021), there is no budget limitation for time and effort to preserve data, as we have plenty of space on our server. However, as the project starts and data will come, it will be necessary a minimum of financial support for ordinary maintenance. However, as mentioned before, all data will be made public through public repositories of ‘omics’ data.

Data Sharing

How will you share the data?

All data will be made public through public repositories of ‘omics’ data (i.e proteomeXchange)

Are any restrictions on data sharing required?

No
Responsibilities and Resources

Who will be responsible for data management?

As of today, all data handling in our server is managed by graduate students and myself. Additionally, IT personnel from the University are available for assisting in any issue.

What resources will you require to deliver your plan?

In fact, financial support is of paramount importance for the ordinary activities of data handling and hardware updating. The IT personnel of the University is constantly assisting us in any issue that may occur with our server or with data handling.