Quantitative phospholipid analysis of zebrafish brains from DJ-1 knockouts, a Parkinson's Disease model

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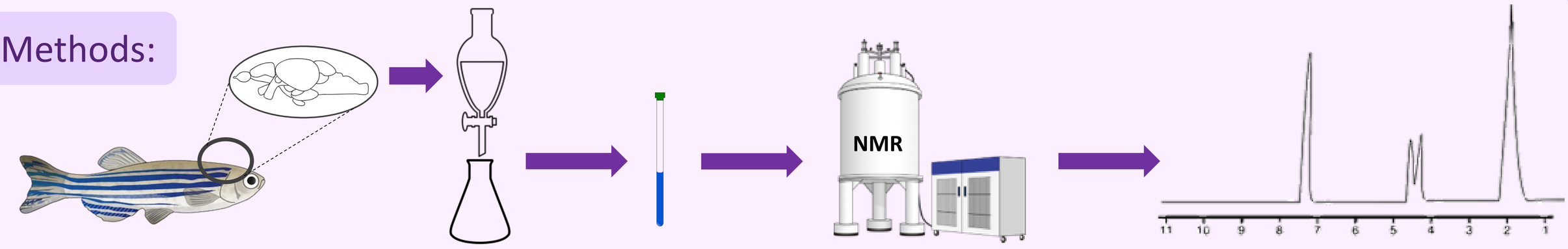
Parkinson's disease is an age-related neurodegenerative disorder associated with both motor and non-motor symptoms. The brain consists of approximately 50% lipids by dry weight. These lipids interact with proteins and other lipids, and are active in many aspects of brain function. Dysfunctional lipid metabolism is linked to inflammation, mitochondrial dysfunction, lipid peroxidation, and oxidative stress, which are also characteristics of Parkinson's disease. **The aim of this project** is to investigate changes in phospholipids related to PD in the brain of wild-type (WT) zebrafish and zebrafish in which the DJ-1/PARK7 gene is deactivated (KO). Lipids will be extracted using a modified Bligh & Dyer method, and their compositions of phospholipids will be analyzed using quantitative ^{31}P NMR.

Methods:



1. Zebrafish brain extraction
2. Two phase lipid extraction
3. Resuspension in CUBO
4. Quantitative ^{31}P NMR
5. Deconvolute and determine lipid relative amounts

Results:

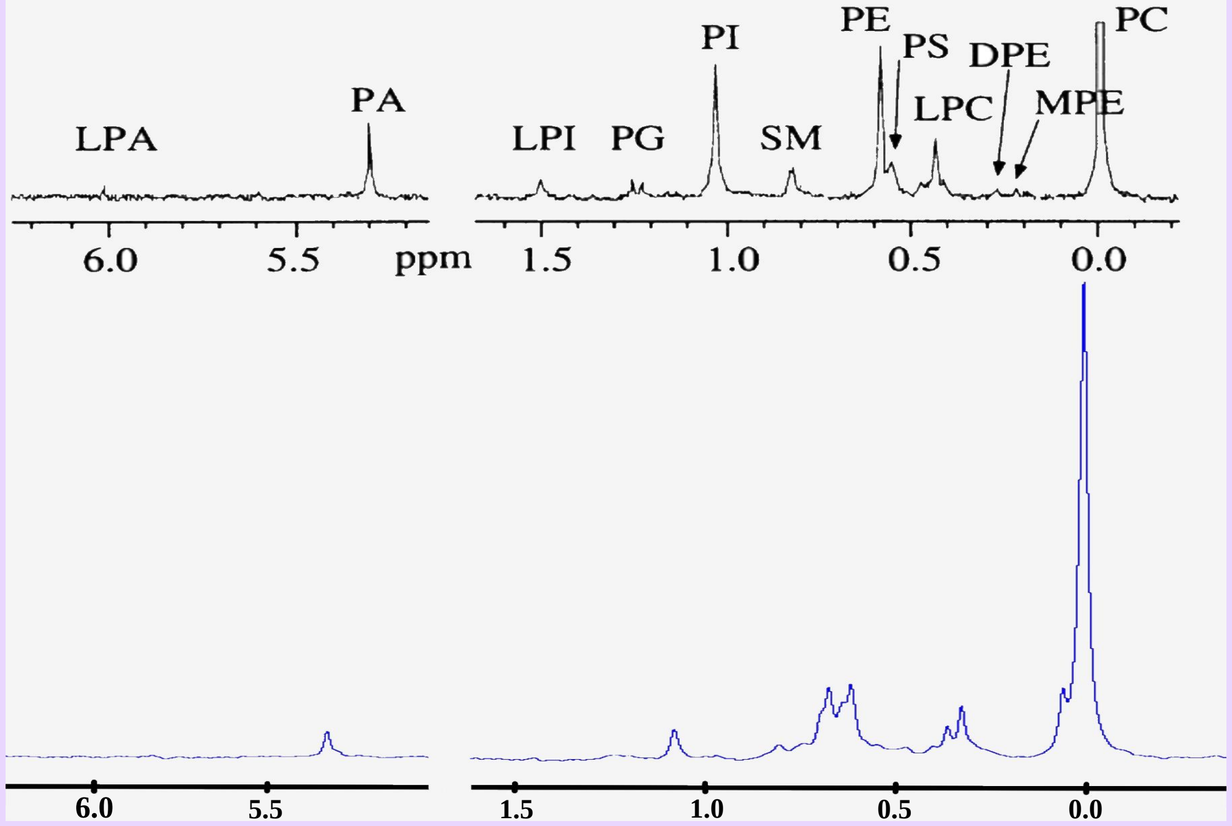


Figure 1: *Result from ^{31}P NMR of a WT zebrafish brain sample (bottom) and NMR spectrum of common phospholipids in CUBO used to determine what lipid corresponds to each peak (top).*

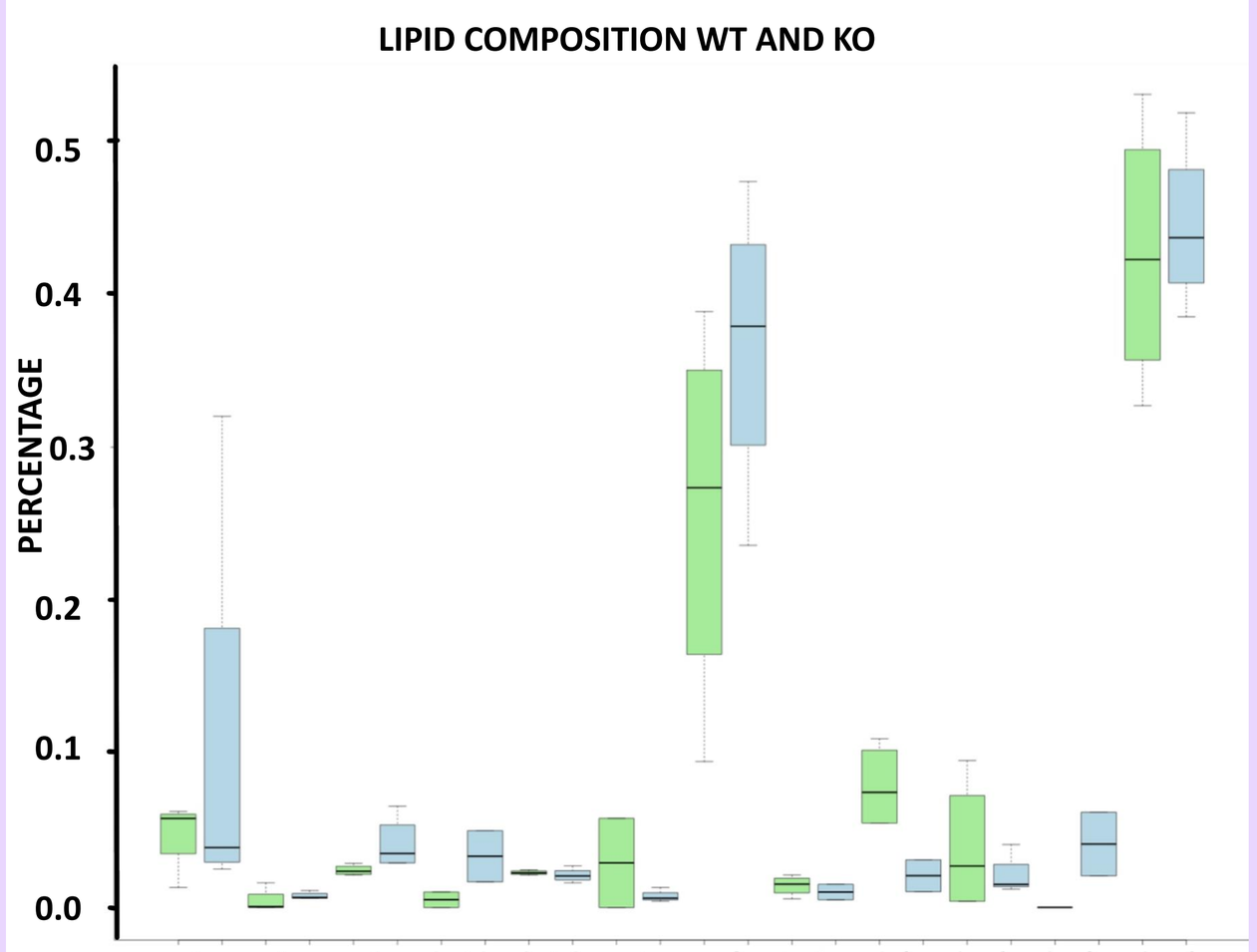


Figure 2: *Lipid composition of WT and KO zebrafish brain samples. Boxplot for WT and KO are paired for the same lipid to compare the different level of the lipid. Blue illustrates WT samples and green illustrates KO. Thick line indicates median values, box limits indicates 25th and 75th percentile, and whiskers shows min and max percentile. No statistical difference was found between groups (P > 0.05, pairwise student's t-test)*

Discussion:

The results from the experiment show the composition divided by phospholipid classes. The relative amounts of each lipid classes could differ between the KO and WT. No significant differences ($p > 0.05$, student's t-test) between WT and KO was observed for any of the detected phospholipids.

The study is limited by ^{31}P NMR not picking up signals from non-phosphorus lipids such as cholesterol. Moreover, the fatty acid chains and the oxidative state of the lipids is not detected either.

Future prospects:

Lipid samples from all extractions were set aside for LC-MS and GC-MS, analyses which could provide more details on more lipid molecules.

References:

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