TN249-10 - Lipofuscin and Euphausiid Aging
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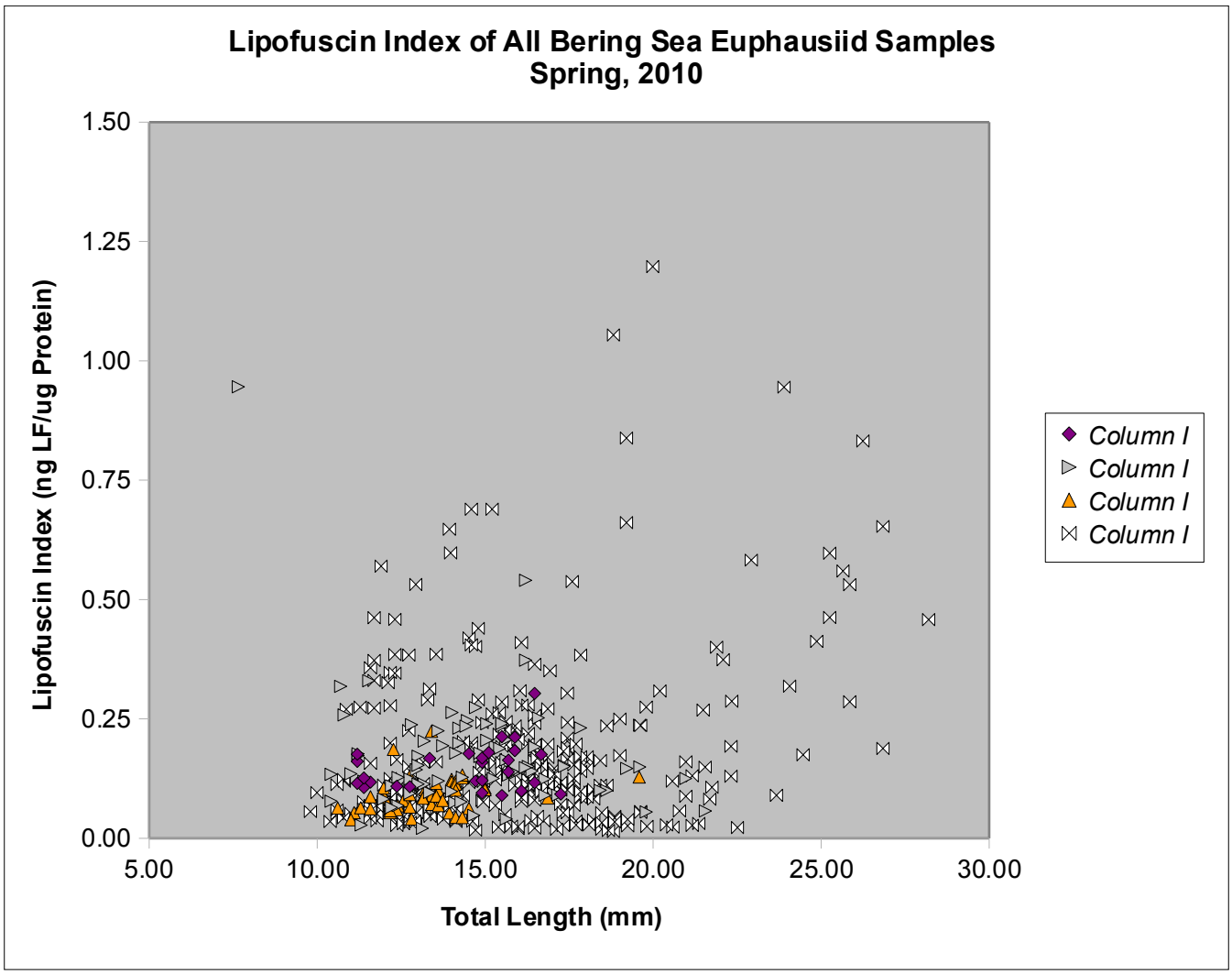
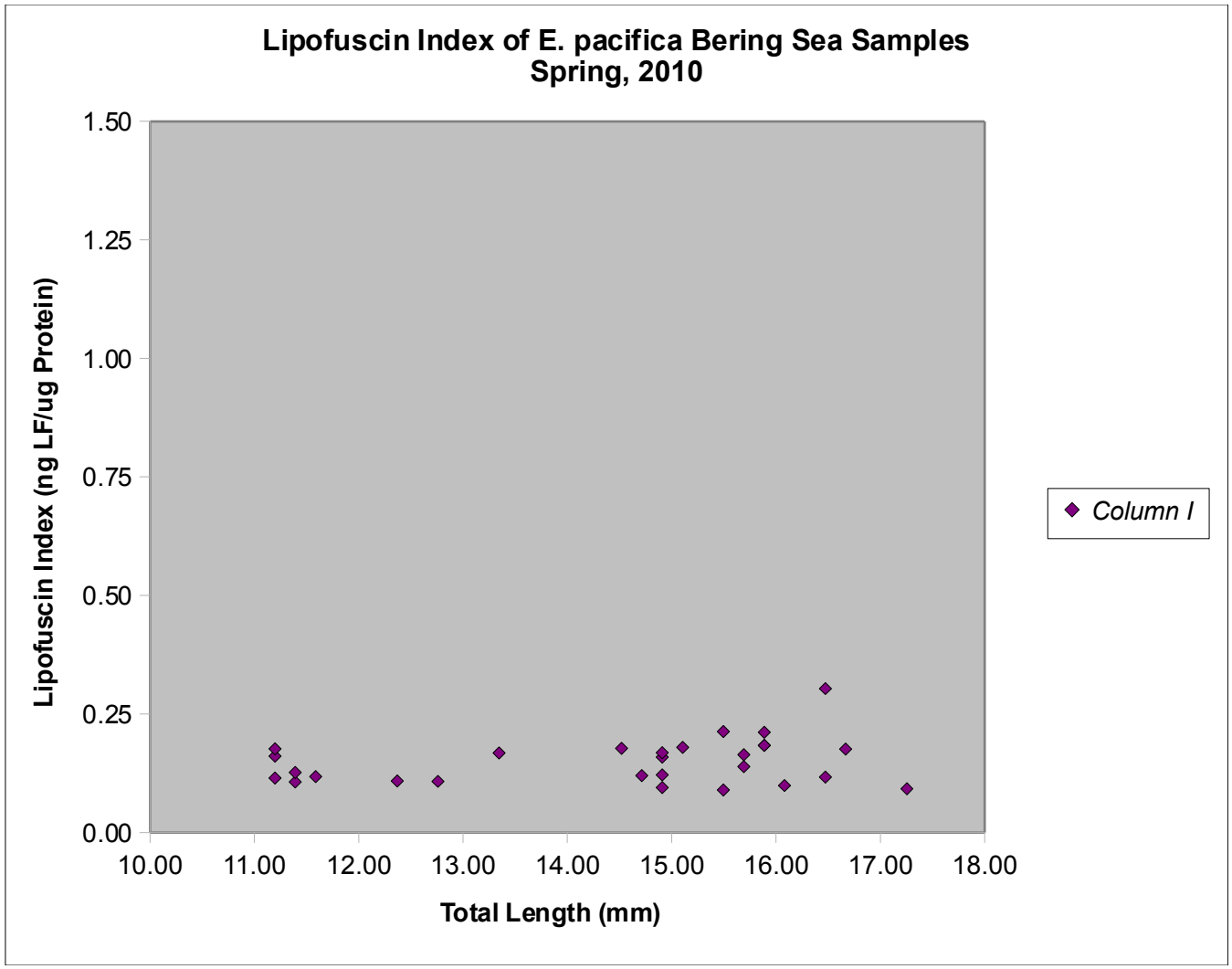
Introduction

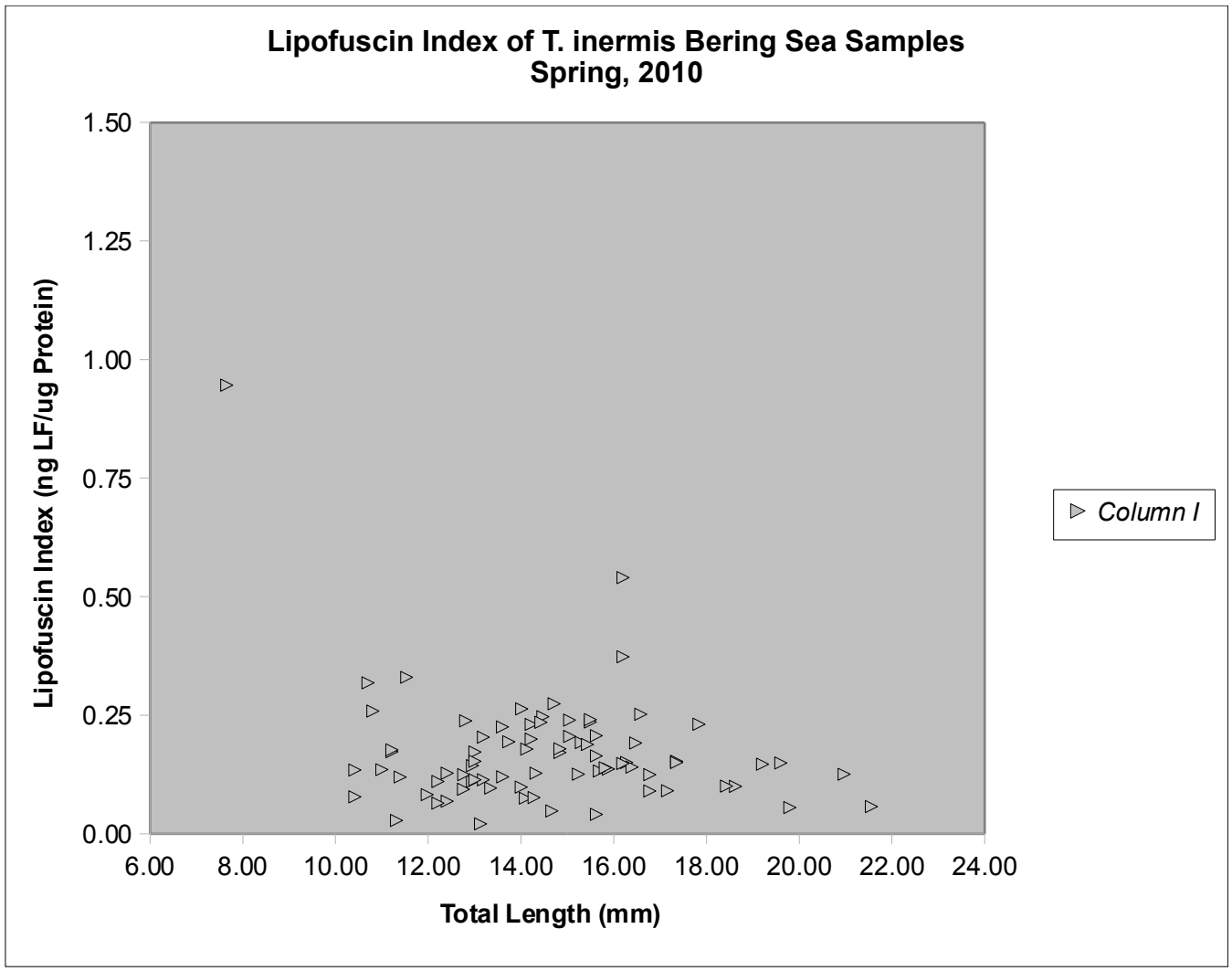
Age determination in crustaceans relies on quantification of a fluorescent "age pigment," lipofuscin, that accumulates in eye stalks of krill over time. Lipofuscin includes a mixture of protein oxidation products formed in neural tissues. The extacted products are quantified using a fluorescence detector. The concentration of lipofuscin is normalized to the protein content from the same tissues. The ratio of lipofuscin to protein concentration is the Lipofuscin Index and is calculated for each individual organisms. In order for the index to reflect chronological age, a calibration is required using euphausiids of known age. A comparison of the calculated ages to the specific lengths for each krill provides the ages for those krill analyzed during each cruise.

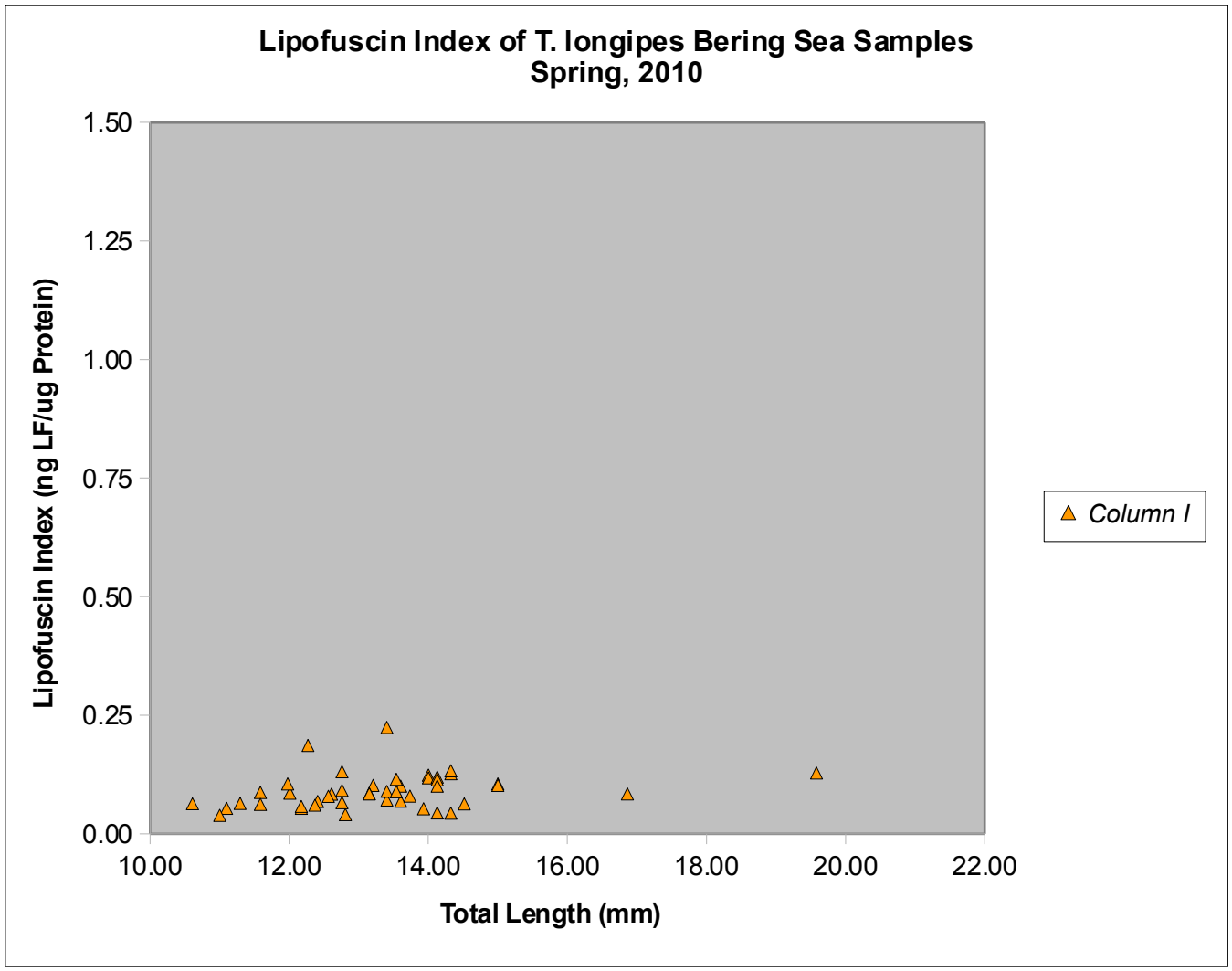
This data is preliminary until an age calibration curve is completed following shore-based rearing.

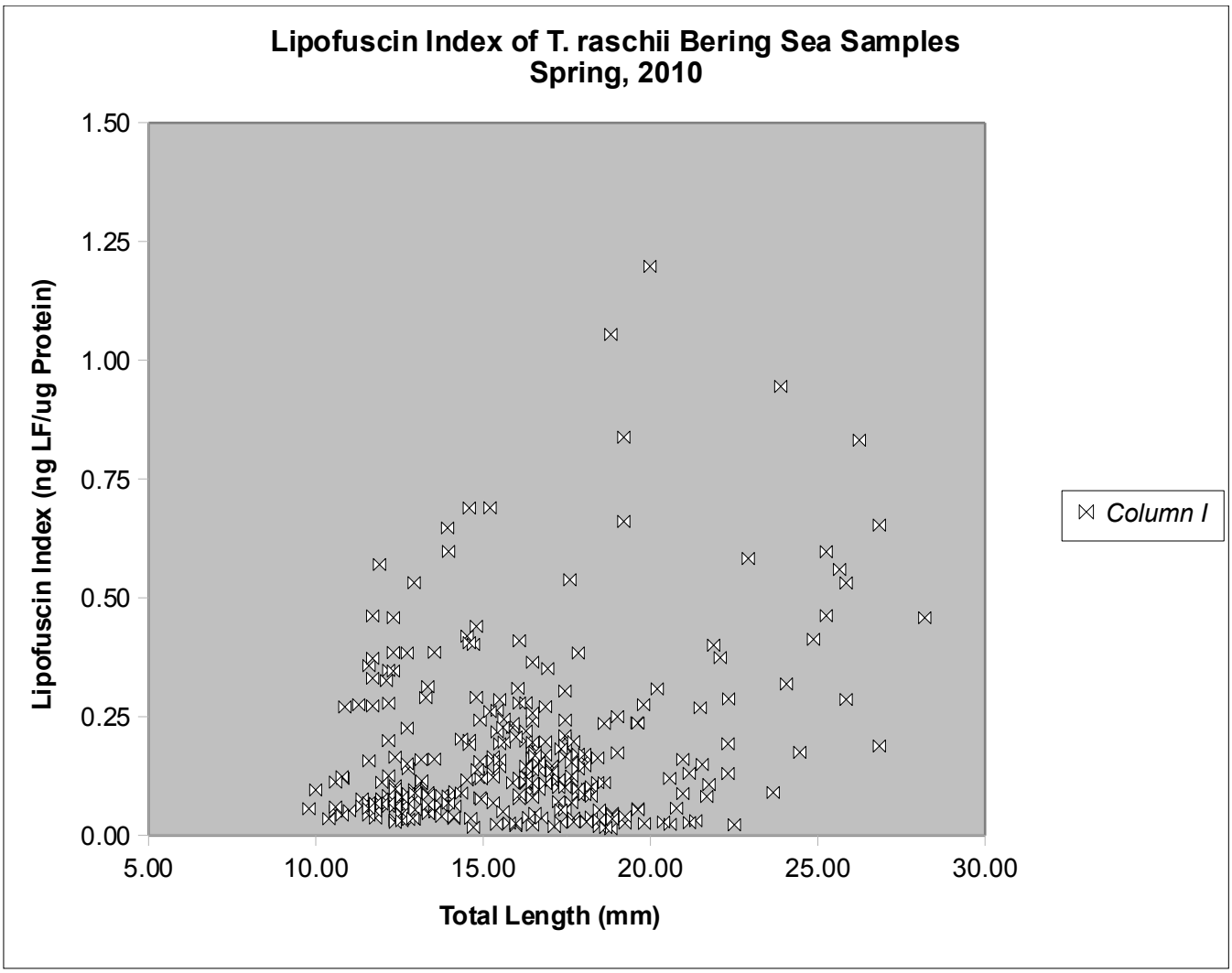
Data Plots:




Lipofuscin Index of E. pacifica Bering Sea Samples
Spring, 2010
Lipofuscin Index (ng LF/ug Protein)
1.50
1.25
1.00
0.75
0.50
0.25
0.00
10.00
11.00
12.00
13.00
14.00
15.00
16.00
17.00
18.00
Total Length (mm)
Column I


Lipofuscin Index of T. inermis Bering Sea Samples
Spring, 2010
Lipofuscin Index (ng LF/ug Protein)
1.50
1.25
1.00
0.75
0.50
0.25
0.00
6.00
8.00
10.00
12.00
14.00
16.00
18.00
20.00
22.00
24.00
Total Length (mm)
Column I


Lipofuscin Index of T. longipes Bering Sea Samples
Spring, 2010
Lipofuscin Index (ng LF/ug Protein)
1.50
1.25
1.00
0.75
0.50
0.25
0.00
10.00
12.00
14.00
16.00
18.00
20.00
22.00
Total Length (mm)
Column I



Calibration Curves

Lipofuscin Proxy

No standards exist for the mix of lipofuscin (LF) "age pigments" that accumulate in crustacean tissues. Quinine sulfate dihydrate serves as a pure compound wth similar fluoresence properties as lipofuscin. Bottles of pre-weighed quinine sulfate dihydrate (QSD) were diluted with a known volume of 0.1N sulfuric acid in deionized water.Both the QSD proxy and the lipofuscin oxidation products were quantified using an Agilent HPLC Fluorescence detector. (QSD Ex: 350nm/Em: 450nm; LF Ex: 355nm/ Em: 525nm)
Sample calibration curves can be found in the data file.

Total Protein

The protein Bovine Serum Albumin (BSA) served as the proxy for total protein concentration found in crustacean neural tissues. Protein content was quantified through fluorescence detection (Ex 285nm/ Em340nm) of aromatic amino acids. The calibration curve generated is then used to quantify the amount of protein in extracts. The method of standard additions is used to generate calibrant solutions from one milliliter ampules of 2mg/mL BSA. (Ex: 285nm/Em: 340nm)
Sample calibration curves can be found in the data file.