NENS230 – Final Project

Nicolette Meyer, December 2017

Nano Secondary Ion Mass Spectrometer is a powerful analytical technique, that provides nanoscale maps of elemental and isotopic composition. It combines high resolution, sensitivity and spatial resolution to simultaneously detect 7 masses from a small sample volume (Guerquin-Kern et al., 2005). In microcosm experiments in environmental microbiology, the addition of substrates containing an isotope label (e.g., ^{13}C, ^{15}N, ^{18}O, ^{2}H, ^{34}S, etc.) can be used to track the anabolic activity of Archaea and Bacteria (Dekas and Orphan, 2011). Furthermore, nanoSIMS measurements can be combined with microscopy using fluorescence in situ hybridization (FISH) to link phylogeny with activity. During FISH, cells fluoresce if binding of specific oligonucleotide probes to regions of the 16S rRNA subunit of the ribosome occurs (Pernthaler et al., 2002). Thus, a subset of cells that are first optically imaged using epifluorescent microscopy, and subsequently targeted with the nanoSIMS, can be used to correlate 16S rRNA phylogenetic identity with the anabolic uptake of certain substrates. In environmental microbiology, the combination of these two techniques has led to the discovery of e.g., anaerobic methane oxidizers in consortia with sulfate reducers at methane seeps (Orphan et al., 2001), the sharing of fixed nitrogen products by diazotrophic cyanobacteria to their symbionts (Behrens et al., 2008), and the detection of nitrogen fixation in marine sediments (Dekas et al., 2009).

However, it has been shown that the FISH process causes a decrease in the ratio of the minor to major isotope ($^{13}C/^{12}C$, $^{15}N/^{14}N$) due to the introduction of unlabeled chemicals and the loss of labelled biomass (Musat et al., 2014; Woebken et al., 2015). This will cause an underestimation of the anabolic rates calculated from nanoSIMS data. Thus, I aim to expand on the existing data by quantifying the decrease in the $^{13}C/^{12}C$, $^{15}N/^{14}N$ and $^{18}O/^{16}O$ isotope label during FISH for *Methanosarcina acetivorans* (Archaea, methanogen found in diverse environments), *Sulfolobus acidocaldarius* (Archaea, thermophile from hotsprings), and *Pseudomonas aeruginosa* (Bacteria, found in diverse environments). The test data used in this assignment was obtained from FISH-treated *M. acetivorans* cells incubated in labelled minimal media containing 50 at-% ^{13}C, 50 at-% ^{15}N and 5 at-% ^{18}O.

Due to the high hourly cost of running the nanoSIMS, rapid data processing during the analysis is essential to aid decision-making and maximize time efficiency on the instrument. The Dekas lab has begun to use Look@NanoSIMS to process nanoSIMS raw data. Look@NanoSIMS is a free software, distributed as a Matlab code, that converts .im raw data files to data tables and figure outputs (Polerecky et al., 2012). However, the default generated outputs are insufficient for our purposes. Thus, I have written a matlab script (*FinalProject_NM_031217.m*) that processes the raw data, and generates a table and a figure output for each element (carbon, nitrogen and oxygen) with 5 subplots (Figure 1). The figures summarize data from one acquisition area represented by a single .im file. The test data used contains 50 ROIs of *M. acetivorans* cells, analyzed over 29 planes, with the raw data tables (.dac and .dat files) from Look@NanoSIMS provided with the submission. All code has been written for this assignment and has not been borrowed or inherited.

References

Behrens, S., Lösekann, T., Pett-Ridge, J., Weber, P.K., Ng, W.O., Stevenson, B.S., Hutcheon, I.D., Relman, D.A. and Spormann, A.M., 2008. Linking microbial phylogeny to metabolic activity at the single-cell level by using

enhanced element labeling-catalyzed reporter deposition fluorescence in situ hybridization (EL-FISH) and NanoSIMS. *Applied and environmental microbiology*, *74*(10), pp.3143-3150.
Dekas, A.E., Poretsky, R.S. and Orphan, V.J., 2009. Deep-sea archaea fix and share nitrogen in methane-consuming microbial consortia. *Science*, *326*(5951), pp.422-426.
Dekas, A.E. and Orphan, V.J., 2011. Identification of diazotrophic microorganisms in marine sediment via fluorescence in situ hybridization coupled to nanoscale secondary ion mass spectrometry (FISH-NanoSIMS). *Methods Enzymol*, *486*, pp.281-305.
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Musat, N., Stryhanyuk, H., Bombach, P., Adrian, L., Audinot, J.N. and Richnow, H.H., 2014. The effect of FISH and CARD-FISH on the isotopic composition of 13 C-and 15 N-labeled Pseudomonas putida cells measured by nanoSIMS. *Systematic and applied microbiology*, *37*(4), pp.267-276.
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Pernthaler, A., Pernthaler, J. and Amann, R., 2002. Fluorescence in situ hybridization and catalyzed reporter deposition for the identification of marine bacteria. *Applied and Environmental Microbiology*, *68*(6), pp.3094-3101.
Polerecky, L., Adam, B., Milucka, J., Musat, N., Vagner, T. and Kuypers, M.M., 2012. Look@ NanoSIMS–a tool for the analysis of nanoSIMS data in environmental microbiology. *Environmental microbiology*, *14*(4), pp.1009-1023.
Woebken, D., Burow, L.C., Behnam, F., Mayali, X., Schintlmeister, A., Fleming, E.D., Prufert-Bebout, L., Singer, S.W., Cortés, A.L., Hoehler, T.M. and Pett-Ridge, J., 2015. Revisiting N2 fixation in Guerrero Negro intertidal microbial mats with a functional single-cell approach. *The ISME journal*, *9*(2), pp.485-496.

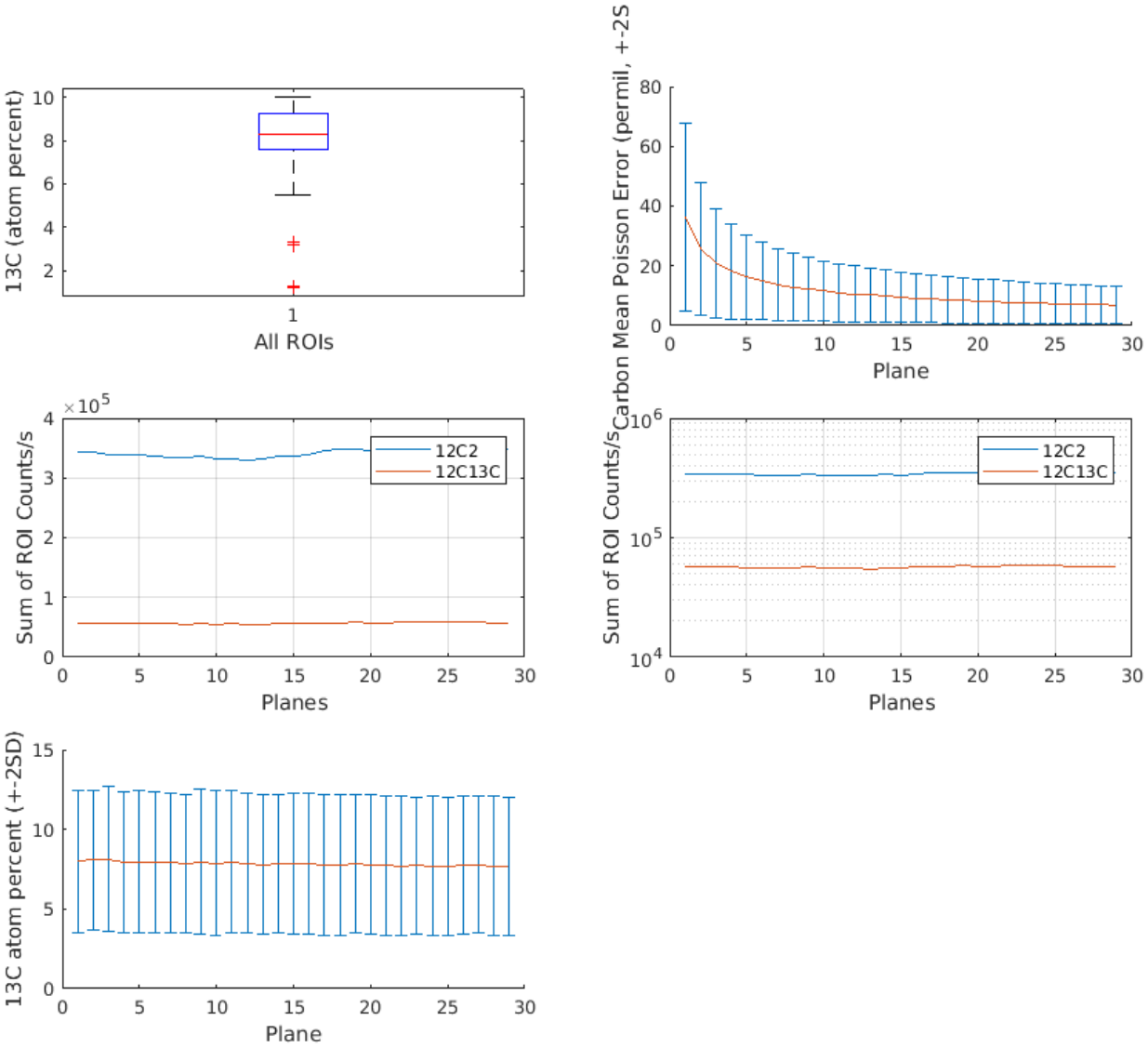


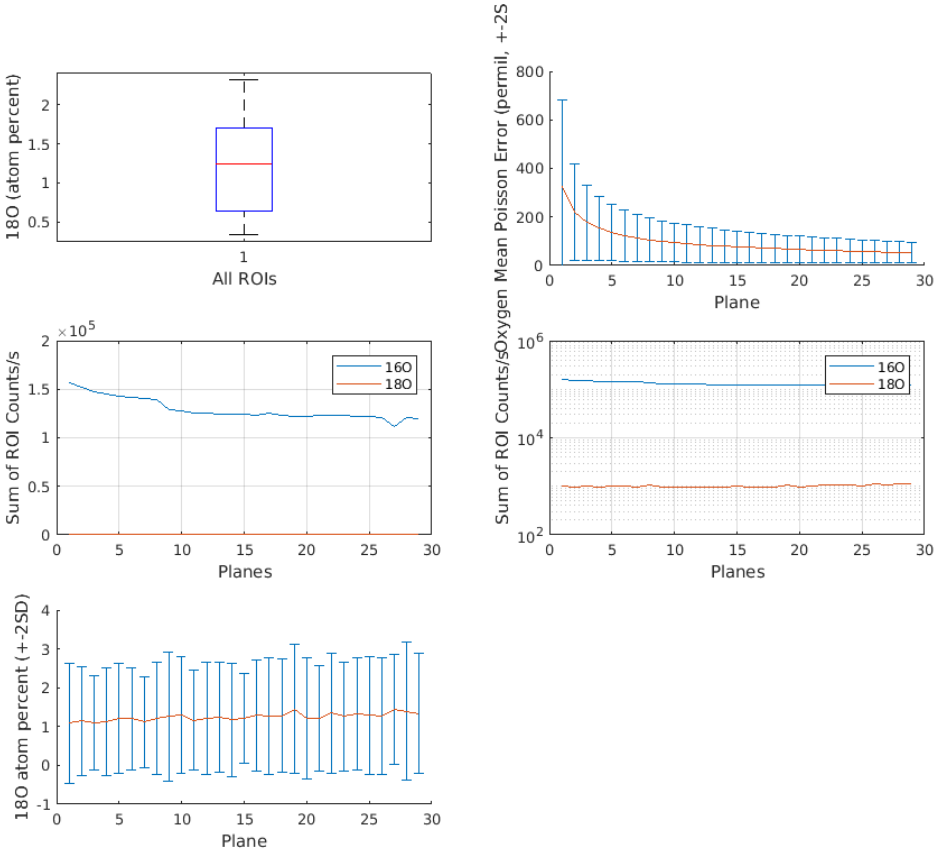
Figure 1: a) box plot showing the spread in the ^{13}C at-% for all ROI's. The carbon isotope ratio for each ROI represents the mean of the ^{13}C at-%over 29 planes. b) line plot showing the decrease in the Poisson error as more data was calculated during the analysis. The Poisson error for each ROI and each plane was calculated from the cumulative sum of the $^{12}C^{13}C$ isotope's counts/s from the first plane to the current plane. The line represents the mean of the ROI's Poisson error. The error bars represent 2 standard deviations about the mean. c) the sum of all the ROI counts/s for $^{12}C_2$ and $^{12}C^{13}C$ for each plane. d) the sum of all the ROI counts/s for $^{12}C_2$ and $^{12}C^{13}C$ for each plane on a log plot. e) the mean ^{13}C at-% for all ROI's for each plane. The error bars represent 2 standard deviations about the mean.

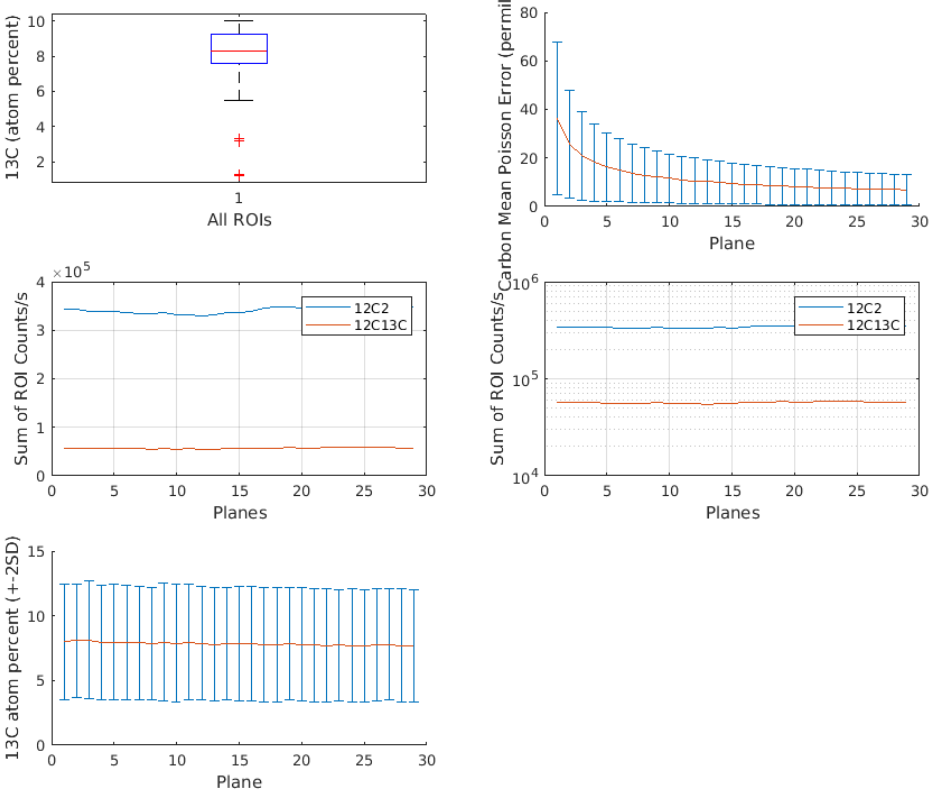
```
%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%Final Project%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%Processing real time nanoSIMS data%%%
%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%

%By: Nicolette Meyer
%December 2017

%This script will help process nanoSIMS data .dac and .dat file
%outputs from Look@NanoSIMS. It processes data from 1 acquisition
 area.
%It is to be used for 16O, 18O, 12C2, 12C13C, 12C14N and 12C15N data.
%All the .dac .dat files required are found in the 'dat' folder. This
%script is to be used for highly enriched samples, as the outputs are
 in
%atom-% instead of delta ratios.

%Data files required:
%'16O.dac'
%'16O-z.dat'
%'18O.dac'
%'18O-z.dat'
%'(18O-(16O+18O))-100.dac'
%'(18O-(16O+18O))-100-z.dat'
%'12C2.dac'
%'12C2-z.dat'
%'12C13C.dac'
%'12C13C-z.dat'
%'(12C13C-(12C2-2+12C13C))-100.dac'
%'(12C13C-(12C2-2+12C13C))-100-z.dat'
%'12C14N.dac'
%'12C14N-z.dat'
%'12C15N.dac'
%'12C15N-z.dat'
%'(12C15N-(12C14N+12C15N))-100.dac'
%'(12C15N-(12C14N+12C15N))-100-z.dat'

%Figure output (1 figure each for oxygen, carbon and nitrogen)
%1. Boxplot showing the spread of isotope ratios for all the ROIs
%2. Change in mean ROI Poisson Error with increasing plane numbers. To
 be used
%to determine the minimum planes required to optimize Poisson
%Error to acquisition time
%3. Sum of all the ROI's counts/s for the major and minor isotope with
%different plane numbers
%4. Sum of all the ROI's counts/s for the major and minor isotope with
%different plane numbers on a log plot
%5. The mean ROI isotope ratios with different plane numbers. To be
 used to
%determine if there is a change in isotope ratios over the analysis
 period
```

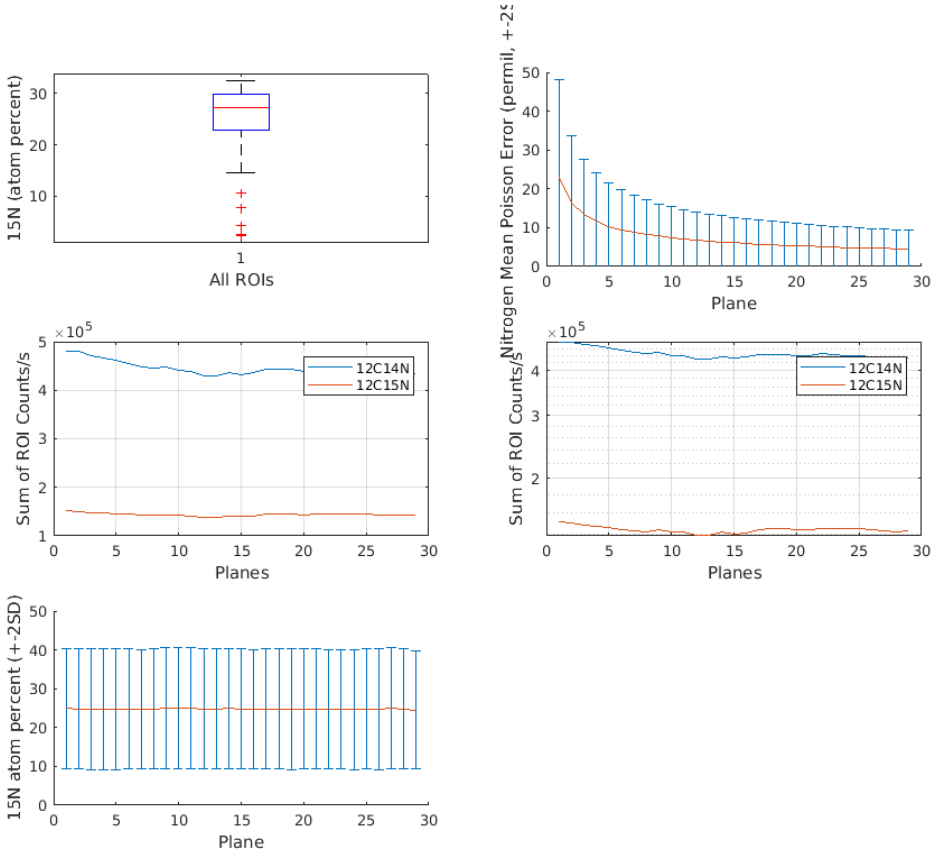
```
%-----------------------------------------------------------------
%Loading the data into separate variables
%Oxygen data
majorO = importdata('16O.dac'); %Mean counts/s of 16O data for each
 ROI
majorOz = importdata('16O-z.dat'); %Counts/s of 16O data for each ROI
 and for each plane
minorO = importdata('18O.dac'); %Mean counts/s of 18O data for each
 ROI
minorOz = importdata('18O-z.dat'); %Counts/s of 18O data for each ROI
 and for each plane
ORatio = importdata('(18O-(16O+18O))-100.dac'); %Mean 18O atom percent
 data for each ROI
ORatioz = importdata('(18O-(16O+18O))-100-z.dat'); %Mean 18O atom
 percent data for each ROI and each plane

%Carbon data
majorC = importdata('12C2.dac');
majorCz = importdata('12C2-z.dat');
minorC = importdata('12C13C.dac');
minorCz = importdata('12C13C-z.dat');
CRatio = importdata('(12C13C-(12C2-2+12C13C))-100.dac');
CRatioz = importdata('(12C13C-(12C2-2+12C13C))-100-z.dat');

%Nitrogen data
majorN = importdata('12C14N.dac');
majorNz = importdata('12C14N-z.dat');
minorN = importdata('12C15N.dac');
minorNz = importdata('12C15N-z.dat');
NRatio = importdata('(12C15N-(12C14N+12C15N))-100.dac');
NRatioz = importdata('(12C15N-(12C14N+12C15N))-100-z.dat');


%-----------------------------------------------------------------
%concatenating data into single structures
%Variable of all the mean 18O, 13C and 15N isotope ratios for all the
 ROIs
ROImeanAllData = [ORatio.data(:,4), CRatio.data(:,4),
 NRatio.data(:,4)];
AllMinorzData = {minorOz.data, minorCz.data, minorNz.data};
AllMajorzData = {majorOz.data, majorCz.data, majorNz.data};
AllIsotopezData = {ORatioz.data, CRatioz.data, NRatioz.data};
AllIsotopeData = {ORatio.data, CRatio.data, NRatio.data};

%Pre-assigning the summary table
SummaryOfResults = zeros(3,4);

%A for loop to create subplots for oxygen, carbon and nitrogen data
%on 3 different figures
for index = 1:3 %index where 1 = oxygen, 2 = carbon, and 3 = nitrogen
    figure(index);
        set(gcf, 'Position', [1000, 1000, 1000, 1000]); %increasing
 the size of the figure generated
```

```
        %1. Box plot of all the ROIs isotope ratios
        subplot(3,2,1);
            ROI_boxplot(ROImeanAllData(:,index), index); %function
 made for this assignment, called 'ROI_boxplot.m'

        %2. Line plot showing the change in Poisson error over time
        subplot(3,2,2);
            PoissonError( AllMinorzData{1,index}, index); %function
 made for this assignment, called 'PoissonError.m'

        %3. and 4. Two line plots showing the change in counts/s over
 to course of the analysis
        subplot(3,2,3);
            CountsOverTime_linear( AllMajorzData{1,index},
 AllMinorzData{1,index}, index ); %function made for this assignment,
 called 'CountsOverTime_linear.m'
        subplot(3,2,4);
            CountsOverTime_log( AllMajorzData{1,index},
 AllMinorzData{1,index}, index ); %function made for this assignment,
 called 'CountsOverTime_log.m'

        %5. Line plot showing the change in the isotope ratios over to
 course of the analysis
        subplot(3,2,5);
            RatiosOverTime( AllIsotopezData{1,index},
 index ); %function made for this assignment, called
 'RatiosOverTime_linear.m'

    %Summary table
    row = summaryTable(AllIsotopeData{1,index}, index );
    SummaryOfResults(index,:) = row;
end %end of for loop

%Converting the summary table into a table with headers
isotope = [1;2;3];
SummaryOfResults = [isotope, SummaryOfResults];
SummaryOfResults = num2cell(SummaryOfResults);
header = {'1=O,2=C,3=N','Mean_ROI_Isotope-
Ratio','Std_ROI_Isotope_Ratio','Mean_ROI_Poiss_Ei', 'Mean_ROI_Poiss_
%Ei'};
SummaryOfResults = [header; SummaryOfResults]


SummaryOfResults =

  4×5 cell array

  Columns 1 through 3

    '1=O,2=C,3=N'    'Mean_ROI_Isotope-…'    'Std_ROI_Isotope_R…'
    [          1]    [             1.2425]    [             0.5747]
    [          2]    [             7.8466]    [             2.1732]
    [          3]    [            24.8152]    [             7.7205]
```

```
Columns 4 through 5

    'Mean_ROI_Poiss_Ei'     'Mean_ROI_Poiss_%Ei'
    [              0.0988]    [              7.5335]
    [              0.0731]    [              1.0034]
    [              0.1519]    [              0.6781]
```




13C (atom percent)
All ROIs
Carbon Mean Poisson Error (permil, +-2S
Plane
Sum of ROI Counts/s
Planes
12C2
12C13C
13C atom percent (+-2SD)
Plane



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```
% Called by FinalProject_NM_031217.m to make a summary table showing
 the
% mean ROI isotope ratios
%
% USAGE:
%       figh = ROI_boxplot(data, isotope)
%
% INPUTS:
%       data        A double array of dimension (number of
 ROI's)*(number of elements analysed = 3)
%                   containing all the ROI means
%       isotope     1 = oxygen, 2 = carbon, 3 = nitrogen
%
% OUTPUTS:
%       figh         Figure handle of resulting figure.
%
% Created by Nicolette Meyer on 02 December 2017


function figh = ROI_boxplot(data, isotope)
minorlabel = ["18O", "13C", "15N"];
elementlabel = ["Oxygen", "Carbon", "Nitrogen"];
figh = figure(gcf);
    boxplot(data);
    xlabel('All ROIs');
    ylabel(sprintf('%s (atom percent)', minorlabel(isotope)));
end
```

```
Not enough input arguments.

Error in ROI_boxplot (line 22)
    boxplot(data);
```

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```
% Called by FinalProject_NM_031217.m to create a line plot showing the
% change in mean Poisson Error with increasing planes. The plot has
 error bars
% that represent 2 standard deviations of the Poisson Error.
%
% USAGE:
%       figh = PoissonError( minorisotope, isotope )
%
% INPUTS:
%       minorisotope        An array of the minor isotope's counts/s
 from the
%                           the depth profile .dat data table. The
 table is
%                           generated by Look@NanoSIMS and is found in
 the 'dat' folder
%       isotope             1 = oxygen, 2 = carbon, 3 = nitrogen
%
% OUTPUTS:
%       figh                Figure handle of resulting figure.
%
% Created by Nicolette Meyer on 02 December 2017

function figh = PoissonError( minorisotope, isotope )
%Getting the dimensions of the minorisotope array
[nrow ncol] = size(minorisotope);

%Removing the columns of errors from the minorisotope array
minorisotope2 = minorisotope(:, (2:2:ncol));

%Making a new array of cumulative counts/s for each ROI over the
 planes
cumulative = zeros(nrow,(ncol-1)/2);
cumulative(1,:) = minorisotope2(1,:);
for plane = 2:nrow
    for ROI = 1:((ncol-1)/2)
        cumulative(plane,ROI) =
 cumulative((plane-1),ROI)+minorisotope2(plane,ROI);
    end
end

%Calculating the poisson error for the cumulative counts/s
PEtable = ((1./cumulative).^0.5).*1000;

%Getting the mean poisson error for all the ROIs for each plane
summaryPE = zeros(nrow,3);
summaryPE(:,1) = minorisotope(:,1);
for plane = 1:nrow
    summaryPE(plane,2) = mean(PEtable(plane,:));
    summaryPE(plane,3) = 2.*std(PEtable(plane,:));
end
```

```
%Making a line plot of the change in poisson error over the # of
 planes
elementlabel = ["Oxygen", "Carbon", "Nitrogen"];
figh = figure(gcf);
    hold on;
    errorbar(summaryPE(:,1), summaryPE(:,2), summaryPE(:,3)), 'black';
    %The plot has error bars that represent 2SD of the Poisson Error
    plot(summaryPE(:,1), summaryPE(:,2));
    xlabel('Plane');
    ylabel(sprintf('%s Mean Poisson Error (permil, +-2SD)',
 elementlabel(isotope)));
    yl = ylim; %obtaining the y-axis' limits
    yl = [0, yl(2)]; %setting the y-axis' lower limit to 0
    ylim(yl);
    hold off;
end
```

```
Not enough input arguments.

Error in PoissonError (line 21)
[nrow ncol] = size(minorisotope);
```

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```
% Called by FinalProject_NM_031217.m to create a figure that shows the
% sum of the counts/s over the course of the analysis (sum ROI counts/
s vs. plane
% number). The y axis scale is linear.
%
% USAGE:
%       figh = CountsOverTime_linear( majorisotope, minorisotope,
 isotope )
%
% INPUTS:
%       major isotope      Data structure with the ROI counts/s for
 each
%                          plane for the major isotope. The structure
%                          should have the same number of columns and
 rows
%                          as the depth raw data output from
 Look@NanoSIMS
%       minor isotope      Data structure with the ROI counts/s for
 each
%                          plane for the minor isotope. The structure
%                          should have the same number of columns and
 rows
%                          as the depth raw data output from
 Look@NanoSIMS
%       isotope            1 = oxygen, 2 = carbon, 3 = nitrogen
%
% OUTPUTS:
%       figh               Figure handle of resulting figure.
%
% Created by Nicolette Meyer on 03 December 2017



function figh = CountsOverTime_linear( majorisotope, minorisotope,
 isotope )
%Getting the dimensions of the minorisotope array
[nrow ncol] = size(minorisotope); %The dimension of the major isotope
 array should be the same

%Minor Isotope
%Removing the columns of errors from the minorisotope array
minorisotope2 = minorisotope(:, (2:2:ncol));

%Making a table of the sum of the counts/s for each plane
SumMinorCounts = zeros(nrow,1);

for plane = 1:nrow
    SumMinorCounts(plane,1) = sum(minorisotope2(plane,:));
end


%Major Isotope
```

```
%Removing the columns of errors from the majorisotope array
majorisotope2 = majorisotope(:, (2:2:ncol));

%Making a table of the sum of the counts/s for each plane
SumMajorCounts = zeros(nrow,1);

for plane = 1:nrow
    SumMajorCounts(plane,1) = sum(majorisotope2(plane,:));
end


%Making a figure of the change in sum(counts/s) vs. plane
minorlabel = ["18O", "12C13C", "12C15N"];
majorlabel = ["16O", "12C2", "12C14N"];

%Linear scale
figh = figure(gcf);
   plot(SumMajorCounts);
   hold on;
   plot(SumMinorCounts);
   legend(majorlabel(isotope),minorlabel(isotope));
   xlabel('Planes');
   ylabel('Sum of ROI Counts/s');
   grid on;
   hold off;
end
```

```
Not enough input arguments.

Error in CountsOverTime_linear (line 28)
[nrow ncol] = size(minorisotope); %The dimension of the major isotope
 array should be the same
```

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```
% Called by FinalProject_NM_031217.m to create a figure that shows the
% sum of the counts/s over the course of the analysis (sum ROI counts/
s vs. plane
% number). The y axis scale is log base 10.
%
% USAGE:
%       figh = CountsOverTime_log( majorisotope, minorisotope,
 isotope )
%
% INPUTS:
%       major isotope      Data structure with the ROI counts/s for
 each
%                          plane for the major isotope. The structure
%                          should have the same number of columns and
 rows
%                          as the depth raw data output from
 Look@NanoSIMS
%       minor isotope      Data structure with the ROI counts/s for
 each
%                          plane for the minor isotope. The structure
%                          should have the same number of columns and
 rows
%                          as the depth raw data output from
 Look@NanoSIMS
%       isotope            1 = oxygen, 2 = carbon, 3 = nitrogen
%
% OUTPUTS:
%       figh               Figure handle of resulting figure.
%
% Created by Nicolette Meyer on 03 December 2017

function [figh] = CountsOverTime_log( majorisotope, minorisotope,
 isotope )
%Getting the dimensions of the minorisotope array
[nrow ncol] = size(minorisotope); %The dimension of the major isotope
 array should be the same

%Minor Isotope
%Removing the columns of errors from the minorisotope array
minorisotope2 = minorisotope(:, (2:2:ncol));

%Making a table of the sum of the counts/s for each plane
SumMinorCounts = zeros(nrow,1);

for plane = 1:nrow
    SumMinorCounts(plane,1) = sum(minorisotope2(plane,:));
end


%Major Isotope
%Removing the columns of errors from the majorisotope array
majorisotope2 = majorisotope(:, (2:2:ncol));
```

```
%Making a table of the sum of the counts/s for each plane
SumMajorCounts = zeros(nrow,1);

for plane = 1:nrow
    SumMajorCounts(plane,1) = sum(majorisotope2(plane,:));
end


%Making a figure of the change in sum(counts/s) vs. plane
minorlabel = ["18O", "12C13C", "12C15N"];
majorlabel = ["16O", "12C2", "12C14N"];


%Log scale
   semilogy(SumMajorCounts);
   hold on;
   semilogy(SumMinorCounts);
   legend(majorlabel(isotope),minorlabel(isotope));
   xlabel('Planes');
   ylabel('Sum of ROI Counts/s');
   grid on;
   hold off;
end
```

```
Not enough input arguments.

Error in CountsOverTime_log (line 26)
[nrow ncol] = size(minorisotope); %The dimension of the major isotope
 array should be the same
```

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```
% Called by FinalProject_NM_031217.m to create a figure that shows the
% change in isotope ratios over the course of the analysis (isotope
 ratio vs. plane
% number).The plot shows error bars that represent 2 standard
 deviations of the isotope ratio.
%
% USAGE:
%       figh = RatiosOverTime( isotoperatioz, isotope )
%
% INPUTS:
%       isotoperatioz      Data structure with the ROI isotope ratios
 for each plane. The
%                          structure should have the same number of
 columns and rows
%                          as the depth raw data output from
 Look@NanoSIMS
%       isotope            1 = oxygen, 2 = carbon, 3 = nitrogen
%
% OUTPUTS:
%       figh               Figure handle of resulting figure.
%
% Created by Nicolette Meyer on 03 December 2017


function [figh] = RatiosOverTime( isotoperatioz, isotope )
%Getting the dimensions of the input array
[nrow ncol] = size(isotoperatioz); %The dimension of the major isotope
 array should be the same

%Removing the columns of errors from the array
isotoperatioz2 = isotoperatioz(:, (2:2:ncol));

%Making a table of the mean and SDs of the isotope ratios for each
 plane
SummaryRatiosz = zeros(nrow,3);
SummaryRatiosz(:,1) = isotoperatioz(:, 1);

for plane = 1:nrow
    SummaryRatiosz(plane,2) = mean(isotoperatioz2(plane,:));
    SummaryRatiosz(plane,3) = 2.*std(isotoperatioz2(plane,:));
end

%Making a line plot of the change in isotope ratios vs. plane (time)
minorlabel = ["18O", "13C", "15N"];
figh = figure(gcf);
    hold on;
    errorbar(SummaryRatiosz(:,1), SummaryRatiosz(:,2),
 SummaryRatiosz(:,3)), 'black';
    %The plot has error bars that represent 2SD of the Poisson Error
    plot(SummaryRatiosz(:,1), SummaryRatiosz(:,2));
    xlabel('Plane');
    ylabel(sprintf('%s atom percent (+-2SD)', minorlabel(isotope)));
```

```
    hold off;
end
```

```
Not enough input arguments.

Error in RatiosOverTime (line 22)
[nrow ncol] = size(isotoperatioz); %The dimension of the major isotope
 array should be the same
```

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```
% Called by FinalProject_NM_031217.m to make a summary table showing
 the
% mean, standard deviation and Poisson Error of the ROI isotope ratios
%
% USAGE:
%       summary = summaryTable(data, isotope)
%
% INPUTS:
%       data        Data structure with the ROI mean isotope ratios.
 The
%                   structure should have the same number of columns
 and rows
%                   as the raw data output from Look@NanoSIMS
%       isotope     1 = oxygen, 2 = carbon, 3 = nitrogen
%
% OUTPUTS:
%       summary     Summary table of the isotope ratios' means,
 standard
%                   deviation and mean Poisson Error
%
% Created by Nicolette Meyer on 03 December 2017


function summary = summaryTable( data, isotope )
summary = zeros(1,4); %Pre-allocation
summary(1,1) = mean(data(:,4)); %Mean ROI isotope ratio
summary(1,2) = std(data(:,4)); %Std ROI isotope ratio
summary(1,3) = mean(data(:,5)); %Mean of the Poiss_Ei
summary(1,4) = mean(data(:,6)); %Mean of the Poiss_%Ei
end
```

```
Not enough input arguments.

Error in summaryTable (line 22)
summary(1,1) = mean(data(:,4)); %Mean ROI isotope ratio
```

Published with MATLAB® R2017a