

Fiber Photometry MATLAB code used in Flanigan et al. 2022

GCaMP Signal Processing (470 nm) for two brain regions using 415 nm as an isosbestic control channel:

```
% for use with arduino generated timestamps, trig 1, 2 roi
```

```
FP.rawdata = Z676data0;
```

```
m=676
```

```
filename = sprintf('SI_end_Analyzed_%d.xls',m)
```

```
picz=sprintf('gcamp_%dzscore',m)
```

```
piccorrection =sprintf( 'gcamp%dcorrection',m)
```

```
picchopped=sprintf( 'gcamp_%dchop',m)
```

```
motioncorrected= sprintf('gcamp_%dmotioncorrect',m)
```

```
regression= sprintf('gcamp_%dregression',m)
```

```
FP.background1=3225
```

```
FP.background2=3065
```

```
%fixes uneven length problem
```

```
if rem(length(FP.rawdata), 2) == 0
```

```
    stop=length(FP.rawdata)
```

```
else
```

```
stop=(length(FP.rawdata)-1)
```

```
end
```

```
%
```

```
% f1 = figure;
```

```

% for i = 1:4
%     subplot(4,1,i)
%     plot(FP.rawdata(:,i))
% end

%f2 deinterleaves and plots each trace individually. written for trig 1
f2 = figure;
subplot(2,3,1)
plot(downsample(FP.rawdata(:,2),2))%takes column 2 data, takes every 2nd data point
starting with the first
title('LHb')
subplot(2,3,4)
plot(downsample(FP.rawdata(2:end,2),2))%same thing but starts with the second number
title('LHb')

subplot(2,3,2)
plot(downsample(FP.rawdata(:,3),2))%same thing but starts with the second number
title('BNST')
subplot(2,3,5)%repeats the same with column 4
plot(downsample(FP.rawdata(2:end,3),2))
title('BNST')

%subplot(2,3,3)
%plot(downsample(FP.rawdata(:,4),2))
%title('OFC')
%subplot(2,3,6)
%plot(downsample(FP.rawdata(2:end,4),2))%same thing but starts with the second number
%title('OFC')

% % starts with 415

```

```
FP.calcium_dependent(:,1) = downsample(FP.rawdata(2:stop,1),2); % takes every 3rd
value from column 1 which is time stamps. adjust to start at second or third number
to match what you need
```

```
FP.calcium_dependent(:,2) = downsample(FP.rawdata(2:stop,2),2); % bla
```

```
FP.calcium_dependent(:,3) = downsample(FP.rawdata(2:stop,3),2); % bnst
```

```
%FP.calcium_dependent(:,4) = downsample(FP.rawdata(2:stop,3),2); % ofc
```

```
FP.calcium_dependent(:,4) = (FP.calcium_dependent(:,1)-
FP.calcium_dependent(1,1))./60000
```

```
FP.isos(:,1) = downsample(FP.rawdata(1:stop,1),2); % takes every 3rd value from
column 1 which is time stamps. adjust to start at second or third number to match
what you need
```

```
FP.isos(:,2) = downsample(FP.rawdata(1:stop,2),2); % bla
```

```
FP.isos(:,3) = downsample(FP.rawdata(1:stop,3),2); % bnst
```

```
%FP.isos(:,4) = downsample(FP.rawdata(1:stop,3),2); % ofc
```

```
FP.isos(:,4) = (FP.calcium_dependent(:,1)-FP.calcium_dependent(1,1))./60000
```

```
%starts wtih 470
```

```
FP.calcium_dependent(:,1) = downsample(FP.rawdata(1:stop,1),2); % takes every 3rd
value from column 1 which is time stamps. adjust to start at second or third number
to match what you need
```

```
FP.calcium_dependent(:,2) = downsample(FP.rawdata(1:stop,2),2); % bla
```

```
FP.calcium_dependent(:,3) = downsample(FP.rawdata(1:stop,3),2); % bnst
```

```
%FP.calcium_dependent(:,4) = downsample(FP.rawdata(1:stop,4),2); % ofc
```

```
FP.calcium_dependent(:,4) = (FP.calcium_dependent(:,1)-
FP.calcium_dependent(1,1))./60000
```

```
FP.isos(:,1) = downsample(FP.rawdata(2:stop,1),2); % takes every 3rd value from
column 1 which is time stamps. adjust to start at second or third number to match
what you need
```

```
FP.isos(:,2) = downsample(FP.rawdata(2:stop,2),2); % bla
```

```
FP.isos(:,3) = downsample(FP.rawdata(2:stop,3),2); % bnst
```

```
%FP.isos(:,4) = downsample(FP.rawdata(2:stop,4),2); % ofc
```

```
FP.isos(:,4) = (FP.calcium_dependent(:,1)-FP.calcium_dependent(1,1))./60000
```

```
%stop=find(FP.calcium_dependent(:,4)>=1.6); %chop off end of trial if needed, time is  
in minutes
```

```
%stop=find(FP.rawdata(:,1)>=(FP.timestamps(10,1)+150000))
```

```
f3 = figure; %plots data against time
```

```
for i = 1:2 %green first. top will be LHb, bottom is bnst
```

```
    subplot(2,1,i)
```

```
    plot(FP.calcium_dependent(:,4),FP.calcium_dependent(:,i+1),'b')
```

```
    hold on
```

```
    plot(FP.calcium_dependent(:,4), FP.isos(:,i+1),'m')
```

```
    xlabel('Time (min)','fontsize',14)
```

```
    ylabel('Signal Intensity (AU)','fontsize',14)
```

```
    title('470','fontsize',16)
```

```
end
```

```
%just subtracts the background for each column from the raw data
```

```
FP.backgroundsubtracted(:,1)= FP.calcium_dependent(:,1); %keeps time in first col
```

```
FP.backgroundsubtracted(:,2)= (FP.calcium_dependent(:,2)-FP.background1);% subtracts  
branch 1 background from branch 1 data
```

```
FP.backgroundsubtracted(:,3)= (FP.calcium_dependent(:,3)-FP.background2);% right
```

```
%FP.backgroundsubtracted(:,4)= (FP.calcium_dependent(:,4)-FP.background2);
```

```
FP.backgroundsubtracted(:,4)= FP.calcium_dependent(:,4);% keeps modified time in 4th  
col
```

```
FP.backgroundsubtractedisos(:,1)= FP.isos(:,1); %keeps time in first col
```

```
FP.backgroundsubtractedisos(:,2)= (FP.isos(:,2)-FP.background1);% subtracts branch 1  
background from branch 1 data
```

```
FP.backgroundsubtractedisos(:,3)= (FP.isos(:,3)-FP.background2);% right
```

```
%FP.backgroundsubtractedisos(:,4)= (FP.isos(:,4)-FP.background2);
```

```
FP.backgroundsubtractedisos(:,4)= FP.isos(:,4);% keeps modified time in 4th col
```

```

%filer specs

Fstop=5

Fsample=20

order=10

%design filter based on parameters specified above

butterfive = designfilt('lowpassfir', 'FilterOrder', order, 'HalfPowerFrequency',
Fstop, 'SampleRate', Fsample, 'DesignMethod', 'butter');

filt=butterfive;

FP.filtered(:,1)= FP.backgroundsubtracted(:,1);
FP.filtered(:,2)= filtfilt(filt,FP.backgroundsubtracted(:,2));
FP.filtered(:,3)= filtfilt(filt,FP.backgroundsubtracted(:,3));
FP.filtered(:,4)= FP.backgroundsubtracted(:,4);

FP.filteredisos(:,1)= FP.backgroundsubtractedisos(:,1);
FP.filteredisos(:,2)= filtfilt(filt,FP.backgroundsubtractedisos(:,2));
FP.filteredisos(:,3)= filtfilt(filt,FP.backgroundsubtractedisos(:,3));
FP.filteredisos(:,4)= FP.backgroundsubtracted(:,4);

start=find(FP.calcium_dependent(:,4)>=0.25); %chop off beginning of trial if needed,
time is in minutes

f4 = figure; %fit signal to bioexp and subtract and normalize470 data
subplot(4,2,1);
temp_fit = fit(FP.filtered(start:end,4),FP.filtered(start:end,2),'exp2');
plot(temp_fit,FP.filtered(start:end,4),FP.filtered(start:end,2))
title('Uncorrected LHb 470','fontsize',16)
ylabel('F (au)','fontsize',14)

```

```

subplot(4,2,2);
temp_fit2 = fit(FP.filtered(start:end,4),FP.filtered(start:end,3),'exp2');
plot(temp_fit2,FP.filtered(start:end,4),FP.filtered(start:end,3))
title('Uncorrected BNST 470','fontsize',16)
ylabel('F (au)','fontsize',14)

subplot(4,2,5);
temp_fit3 = fit(FP.filteredisos(start:end,4),FP.filteredisos(start:end,2),'exp2');
plot(temp_fit3,FP.filteredisos(start:end,4),FP.filteredisos(start:end,2))
title('Uncorrected LHb isos','fontsize',16)
ylabel('F (au)','fontsize',14)

subplot(4,2,6);
temp_fit4 = fit(FP.filteredisos(start:end,4),FP.filteredisos(start:end,3),'exp2');
plot(temp_fit4,FP.filteredisos(start:end,4),FP.filteredisos(start:end,3))
title('Uncorrected BNST isos','fontsize',16)
ylabel('F (au)','fontsize',14)

subplot(4,2,3);
plot(FP.filtered(start:end,4),100*(FP.filtered(start:end,2)-
temp_fit(FP.filtered(start:end,4)))./temp_fit(FP.filtered(start:end,4)))
title('Corrected LHb 470','fontsize',16)
xlabel('Time (m)','fontsize',14)
ylabel('dF/F %','fontsize',14)

subplot(4,2,4);
plot(FP.filtered(start:end,4),100*(FP.filtered(start:end,3)-
temp_fit2(FP.filtered(start:end,4)))./temp_fit2(FP.filtered(start:end,4)))
title('Corrected BNST 470','fontsize',16)
xlabel('Time (m)','fontsize',14)

```

```
ylabel('dF/F %', 'fontsize', 14)
```

```
subplot(4,2,7);
```

```
plot(FP.filteredisos(start:end,4),100*(FP.filteredisos(start:end,2)-  
temp_fit3(FP.filteredisos(start:end,4)))./temp_fit3(FP.filteredisos(start:end,4)))
```

```
title('Corrected LHb 415', 'fontsize', 16)
```

```
xlabel('Time (m)', 'fontsize', 14)
```

```
ylabel('dF/F %', 'fontsize', 14)
```

```
subplot(4,2,8);
```

```
plot(FP.filteredisos(start:end,4),100*(FP.filteredisos(start:end,3)-  
temp_fit4(FP.filteredisos(start:end,4)))./temp_fit4(FP.filteredisos(start:end,4)))
```

```
title('Corrected BNST 415', 'fontsize', 16)
```

```
xlabel('Time (m)', 'fontsize', 14)
```

```
ylabel('dF/F %', 'fontsize', 14)
```

```
savefig(piccorrection)
```

```
%gives background subtracted data- the fit of background subtracted
```

```
%data/fit. this is df/f
```

```
FP.fitsubtract(:,1) = (FP.filtered(start:end,1));
```

```
FP.fitsubtract(:,2) = (FP.filtered(start:end,2)-  
temp_fit(FP.filtered(start:end,4)))./temp_fit(FP.filtered(start:end,4));
```

```
FP.fitsubtract(:,3) = (FP.filtered(start:end,3)-  
temp_fit2(FP.filtered(start:end,4)))./temp_fit2(FP.filtered(start:end,4));
```

```
%FP.fitsubtract(:,4) = (FP.filtered(start:end,4)-  
temp_fit2(FP.filtered(start:end,5)))./temp_fit3(FP.filtered(start:end,5));
```

```
FP.fitsubtract(:,4) = FP.calcium_dependent(start:end,4);
```

```
FP.fitsubtractisos(:,1) = (FP.filteredisos(start:end,1));
```

```
FP.fitsubtractisos(:,2) = (FP.filteredisos(start:end,2)-  
temp_fit3(FP.filteredisos(start:end,4)))./temp_fit3(FP.filteredisos(start:end,4));
```

```
FP.fitsubtractisos(:,3) = (FP.filteredisos(start:end,3)-  
temp_fit4(FP.filteredisos(start:end,4)))./temp_fit4(FP.filteredisos(start:end,4));
```

```

%FP.fitsubtractisos(:,4) = (FP.filteredisos(start:end,4)-
temp_fit4(FP.filteredisos(start:end,5))./temp_fit4(FP.filteredisos(start:end,5)));

FP.fitsubtractisos(:,4) = FP.calcium_dependent(start:end,4);

```

```

FP.zscore(:,1)= FP.fitsubtract(:,1);
FP.zscore(:,2)=zscore(FP.fitsubtract(:,2));
FP.zscore(:,3)=zscore(FP.fitsubtract(:,3));
%FP.zscore(:,4)=zscore(FP.fitsubtract(:,4));
FP.zscore(:,4)=FP.fitsubtract(:,4);

```

```

FP.zscorei(:,1)= FP.fitsubtractisos(:,1);
FP.zscorei(:,2)=zscore(FP.fitsubtractisos(:,2));
FP.zscorei(:,3)=zscore(FP.fitsubtractisos(:,3));
%FP.zscorei(:,4)=zscore(FP.fitsubtractisos(:,4));
FP.zscorei(:,4)=FP.fitsubtractisos(:,4);

```

```

f7=figure %plots z scored data against time
subplot(2,1,1)
plot(FP.zscore(:,4),FP.zscore(:,2),'b')
hold on
plot(FP.zscorei(:,4),FP.zscorei(:,2),'m')
title('Z-scored LHB','fontsize',16)
xlabel('Time (m)','fontsize',14)
ylabel('Z-score','fontsize',14)

```

```

subplot(2,1,2)
plot(FP.zscore(:,4),FP.zscore(:,3),'b')
hold on
plot(FP.zscorei(:,4),FP.zscorei(:,3),'m')
title('Z-scored L','fontsize',16)
xlabel('Time (m)','fontsize',14)
ylabel('Z-score','fontsize',14)

```



```

savefig(picz)

% using non negative robust linear regression---DO NOT USE THIS PART IF
% USING A NON GCAMP SENSOR

fitdataR = fit(FP.zscorei(:,2),FP.zscore(:,2),fitttype('poly1'),'Robust','on');
fitdataL = fit(FP.zscorei(:,3),FP.zscore(:,3),fitttype('poly1'),'Robust','on');
% Plot fit
figure
subplot(2,1,1)
hold on
plot(FP.zscorei(:,2),FP.zscore(:,2),'k.')
plot(fitdataR,'b')
title('Lin reg R')
hold off

subplot(2,1,2)
hold on
plot(FP.zscorei(:,3),FP.zscore(:,3),'k.')
plot(fitdataL,'b')
title('lin reg L')
hold off
savefig(regression)

%fit isos to ca signal
isosfitted(:,1)=fitdataR(FP.zscorei(:,2));
isosfitted(:,2)=fitdataL(FP.zscorei(:,3));

% Plot aligned signals
figure
subplot(2,1,1)
plot(FP.zscore(:,4),isosfitted(:,1),'m')

```

```

hold on
plot(FP.zscore(:,4),FP.zscore(:,2),'b')
title('fitted isos and signal R')
hold off

subplot(2,1,2)
plot(FP.zscore(:,4),isosfitted(:,2),'m')
hold on
plot(FP.zscore(:,4),FP.zscore(:,3),'b')
title('fitted isos and signal L')
hold off

FP.corrected(:,1)=FP.zscore(:,1)
FP.corrected(:,2)=(FP.zscore(:,2)-isosfitted(:,1))
FP.corrected(:,3)=(FP.zscore(:,3)-isosfitted(:,2))
%FP.corrected(:,4)=(FP.zscore(:,2)-isosfitted(:,1))
FP.corrected(:,4)=FP.zscore(:,4)

figure
subplot(2,1,1)
plot(FP.corrected(:,4),FP.corrected(:,2),'k')
title('Motion Corrected signal R')
xlabel('Time')
ylabel('z-score')

hold on

subplot(2,1,2)
plot(FP.corrected(:,4),FP.corrected(:,3),'k')
title('Motion Corrected signal L')
xlabel('Time')

```

```
ylabel('z-score')
savefig(motioncorrected)
```

GRAB-5HT Signal Processing (470 nm) for two brain regions:

```
% for use with arduino generated timestamps, trig 1, 2 roi
```

```
FP.rawdata = Z947data0;
```

```
FP.timestamps =Z947timestamps;
```

```
m=947
```

```
filename = sprintf('etohDrinking_Analyzed_%d.xls',m)
```

```
picz=sprintf('gcamp_%dzscore',m)
```

```
piccorrection =sprintf( 'gcamp%dcorrection',m)
```

```
picchopped=sprintf( 'gcamp_%dchop',m)
```

```
motioncorrected= sprintf('gcamp_%dmotioncorrect',m)
```

```
regression= sprintf('gcamp_%dregression',m)
```

```
FP.background1=3225
```

```
FP.background2=3065
```

```
%fixes uneven length problem
```

```
if rem(length(FP.rawdata), 2) == 0
```

```
    stop=length(FP.rawdata)
```

```
else
```

```
stop=(length(FP.rawdata)-1)
```

```
end
```

```

%stop=find(FP.rawdata(:,1)>=(FP.timestamps(10,1)+150000))
%
% f1 = figure;
% for i = 1:4
%     subplot(4,1,i)
%     plot(FP.rawdata(:,i))
% end

%f2 deinterleaves and plots each trace individually. written for trig 1
f2 = figure;
subplot(2,3,1)
plot(downsample(FP.rawdata(:,2),2))%takes column 2 data, takes every 2nd data point
starting with the first
title('LHb')
subplot(2,3,4)
plot(downsample(FP.rawdata(2:end,2),2))%same thing but starts with the second number
title('LHb')

subplot(2,3,2)
plot(downsample(FP.rawdata(:,3),2))%same thing but starts with the second number
title('BNST')
subplot(2,3,5)%repeats the same with column 4
plot(downsample(FP.rawdata(2:end,3),2))
title('BNST')

%subplot(2,3,3)
%plot(downsample(FP.rawdata(:,4),2))
%title('OFC')
%subplot(2,3,6)
%plot(downsample(FP.rawdata(2:end,4),2))%same thing but starts with the second number
%title('OFC')

```

```
% % starts wtih 415
```

```
FP.calcium_dependent(:,1) = downsample(FP.rawdata(2:stop,1),2); % takes every 3rd  
value from column 1 which is time stamps. adjust to start at second or third number  
to match what you need
```

```
FP.calcium_dependent(:,2) = downsample(FP.rawdata(2:stop,2),2); % bla
```

```
FP.calcium_dependent(:,3) = downsample(FP.rawdata(2:stop,3),2); % bnst
```

```
%FP.calcium_dependent(:,4) = downsample(FP.rawdata(2:stop,3),2); % ofc
```

```
FP.calcium_dependent(:,4) = (FP.calcium_dependent(:,1)-  
FP.calcium_dependent(1,1))./60000
```

```
FP.isos(:,1) = downsample(FP.rawdata(1:stop,1),2); % takes every 3rd value from  
column 1 which is time stamps. adjust to start at second or third number to match  
what you need
```

```
FP.isos(:,2) = downsample(FP.rawdata(1:stop,2),2); % bla
```

```
FP.isos(:,3) = downsample(FP.rawdata(1:stop,3),2); % bnst
```

```
%FP.isos(:,4) = downsample(FP.rawdata(1:stop,3),2); % ofc
```

```
FP.isos(:,4) = (FP.calcium_dependent(:,1)-FP.calcium_dependent(1,1))./60000
```

```
%starts wtih 470
```

```
FP.calcium_dependent(:,1) = downsample(FP.rawdata(1:stop,1),2); % takes every 3rd  
value from column 1 which is time stamps. adjust to start at second or third number  
to match what you need
```

```
FP.calcium_dependent(:,2) = downsample(FP.rawdata(1:stop,2),2); % bla
```

```
FP.calcium_dependent(:,3) = downsample(FP.rawdata(1:stop,3),2); % bnst
```

```
%FP.calcium_dependent(:,4) = downsample(FP.rawdata(1:stop,4),2); % ofc
```

```
FP.calcium_dependent(:,4) = (FP.calcium_dependent(:,1)-  
FP.calcium_dependent(1,1))./60000
```

```
FP.isos(:,1) = downsample(FP.rawdata(2:stop,1),2); % takes every 3rd value from  
column 1 which is time stamps. adjust to start at second or third number to match  
what you need
```

```
FP.isos(:,2) = downsample(FP.rawdata(2:stop,2),2); % bla
```

```

FP.isos(:,3) = downsample(FP.rawdata(2:stop,3),2); % bnst
%FP.isos(:,4) = downsample(FP.rawdata(2:stop,4),2); % ofc
FP.isos(:,4) = (FP.calcium_dependent(:,1)-FP.calcium_dependent(1,1))./60000

f3 = figure; %plots data agaist time
for i = 1:2 %green first. top will be LHb, bottom is bnst
    subplot(2,1,i)
    plot(FP.calcium_dependent(:,4),FP.calcium_dependent(:,i+1),'b')
    hold on
    plot(FP.calcium_dependent(:,4), FP.isos(:,i+1),'m')
    xlabel('Time (min)','fontsize',14)
    ylabel('Signal Intensity (AU)','fontsize',14)
    title('470','fontsize',16)
end

%just subtracts the background for each column from the raw data
FP.backgroundsubtracted(:,1)= FP.calcium_dependent(:,1); %keeps time in first col
FP.backgroundsubtracted(:,2)= (FP.calcium_dependent(:,2)-FP.background1);% subtracts
branch 1 background from branch 1 data
FP.backgroundsubtracted(:,3)= (FP.calcium_dependent(:,3)-FP.background2);% right
%FP.backgroundsubtracted(:,4)= (FP.calcium_dependent(:,4)-FP.background2);
FP.backgroundsubtracted(:,4)= FP.calcium_dependent(:,4);% keeps modified time in 4th
col

FP.backgroundsubtractedisos(:,1)= FP.isos(:,1); %keeps time in first col
FP.backgroundsubtractedisos(:,2)= (FP.isos(:,2)-FP.background1);% subtracts branch 1
background from branch 1 data
FP.backgroundsubtractedisos(:,3)= (FP.isos(:,3)-FP.background2);% right
%FP.backgroundsubtractedisos(:,4)= (FP.isos(:,4)-FP.background2);
FP.backgroundsubtractedisos(:,4)= FP.isos(:,4);% keeps modified time in 4th col

```

```

%filer specs

Fstop=5

Fsample=20

order=10

%design filter based on parameters specified above

butterfive = designfilt('lowpassiir', 'FilterOrder', order, 'HalfPowerFrequency',
Fstop, 'SampleRate', Fsample, 'DesignMethod', 'butter');

filt=butterfive;

FP.filtered(:,1)= FP.backgroundsubtracted(:,1);
FP.filtered(:,2)= filtfilt(filt,FP.backgroundsubtracted(:,2));
FP.filtered(:,3)= filtfilt(filt,FP.backgroundsubtracted(:,3));
FP.filtered(:,4)= FP.backgroundsubtracted(:,4);

FP.filteredisos(:,1)= FP.backgroundsubtractedisos(:,1);
FP.filteredisos(:,2)= filtfilt(filt,FP.backgroundsubtractedisos(:,2));
FP.filteredisos(:,3)= filtfilt(filt,FP.backgroundsubtractedisos(:,3));
FP.filteredisos(:,4)= FP.backgroundsubtracted(:,4);

start=find(FP.calcium_dependent(:,4)>=0.75); %chop off beginning of trial if needed,
time is in minutes

f4 = figure; %fit signal to bioexp and subtract and normalize470 data
subplot(4,2,1);
temp_fit = fit(FP.filtered(start:end,4),FP.filtered(start:end,2),'exp2');
plot(temp_fit,FP.filtered(start:end,4),FP.filtered(start:end,2))
title('Uncorrected LHb 470','fontsize',16)
ylabel('F (au)','fontsize',14)

```

```

subplot(4,2,2);
temp_fit2 = fit(FP.filtered(start:end,4),FP.filtered(start:end,3),'exp2');
plot(temp_fit2,FP.filtered(start:end,4),FP.filtered(start:end,3))
title('Uncorrected BNST 470','fontsize',16)
ylabel('F (au)','fontsize',14)

subplot(4,2,5);
temp_fit3 = fit(FP.filteredisos(start:end,4),FP.filteredisos(start:end,2),'exp2');
plot(temp_fit3,FP.filteredisos(start:end,4),FP.filteredisos(start:end,2))
title('Uncorrected LHb isos','fontsize',16)
ylabel('F (au)','fontsize',14)

subplot(4,2,6);
temp_fit4 = fit(FP.filteredisos(start:end,4),FP.filteredisos(start:end,3),'exp2');
plot(temp_fit4,FP.filteredisos(start:end,4),FP.filteredisos(start:end,3))
title('Uncorrected BNST isos','fontsize',16)
ylabel('F (au)','fontsize',14)

subplot(4,2,3);
plot(FP.filtered(start:end,4),100*(FP.filtered(start:end,2)-
temp_fit(FP.filtered(start:end,4)))./temp_fit(FP.filtered(start:end,4)))
title('Corrected LHb 470','fontsize',16)
xlabel('Time (m)','fontsize',14)
ylabel('dF/F %','fontsize',14)

subplot(4,2,4);
plot(FP.filtered(start:end,4),100*(FP.filtered(start:end,3)-
temp_fit2(FP.filtered(start:end,4)))./temp_fit2(FP.filtered(start:end,4)))
title('Corrected BNST 470','fontsize',16)
xlabel('Time (m)','fontsize',14)

```



```

ylabel('dF/F %', 'fontsize', 14)

subplot(4,2,7);
plot(FP.filteredisos(start:end,4),100*(FP.filteredisos(start:end,2)-
temp_fit3(FP.filteredisos(start:end,4)))./temp_fit3(FP.filteredisos(start:end,4)))
title('Corrected LHb 415', 'fontsize', 16)
xlabel('Time (m)', 'fontsize', 14)
ylabel('dF/F %', 'fontsize', 14)

subplot(4,2,8);
plot(FP.filteredisos(start:end,4),100*(FP.filteredisos(start:end,3)-
temp_fit4(FP.filteredisos(start:end,4)))./temp_fit4(FP.filteredisos(start:end,4)))
title('Corrected BNST 415', 'fontsize', 16)
xlabel('Time (m)', 'fontsize', 14)
ylabel('dF/F %', 'fontsize', 14)
savefig(piccorrection)

%gives background subtracted data- the fit of background subtracted
%data/fit. this is df/f
FP.fitsubtract(:,1) = (FP.filtered(start:end,1));
FP.fitsubtract(:,2) = (FP.filtered(start:end,2)-
temp_fit(FP.filtered(start:end,4)))./temp_fit(FP.filtered(start:end,4));
FP.fitsubtract(:,3) = (FP.filtered(start:end,3)-
temp_fit2(FP.filtered(start:end,4)))./temp_fit2(FP.filtered(start:end,4));
%FP.fitsubtract(:,4) = (FP.filtered(start:end,4)-
temp_fit2(FP.filtered(start:end,5)))./temp_fit3(FP.filtered(start:end,5));
FP.fitsubtract(:,4) = FP.calcium_dependent(start:end,4);

FP.fitsubtractisos(:,1) = (FP.filteredisos(start:end,1));
FP.fitsubtractisos(:,2) = (FP.filteredisos(start:end,2)-
temp_fit3(FP.filteredisos(start:end,4)))./temp_fit3(FP.filteredisos(start:end,4));
FP.fitsubtractisos(:,3) = (FP.filteredisos(start:end,3)-
temp_fit4(FP.filteredisos(start:end,4)))./temp_fit4(FP.filteredisos(start:end,4));

```

```

%FP.fitsubtractisos(:,4) = (FP.filteredisos(start:end,4)-
temp_fit4(FP.filteredisos(start:end,5))./temp_fit4(FP.filteredisos(start:end,5)));

FP.fitsubtractisos(:,4) = FP.calcium_dependent(start:end,4);

```

```

FP.zscore(:,1)= FP.fitsubtract(:,1);
FP.zscore(:,2)=zscore(FP.fitsubtract(:,2));
FP.zscore(:,3)=zscore(FP.fitsubtract(:,3));
%FP.zscore(:,4)=zscore(FP.fitsubtract(:,4));
FP.zscore(:,4)=FP.fitsubtract(:,4);

```

```

FP.zscorei(:,1)= FP.fitsubtractisos(:,1);
FP.zscorei(:,2)=zscore(FP.fitsubtractisos(:,2));
FP.zscorei(:,3)=zscore(FP.fitsubtractisos(:,3));
%FP.zscorei(:,4)=zscore(FP.fitsubtractisos(:,4));
FP.zscorei(:,4)=FP.fitsubtractisos(:,4);

```

```

f7=figure %plots z scored data against time
subplot(2,1,1)
plot(FP.zscore(:,4),FP.zscore(:,2), 'b')
hold on
plot(FP.zscorei(:,4),FP.zscorei(:,2), 'm')
title('Z-scored LHB', 'fontsize', 16)
xlabel('Time (m)', 'fontsize', 14)
ylabel('Z-score', 'fontsize', 14)

```

```

subplot(2,1,2)
plot(FP.zscore(:,4),FP.zscore(:,3), 'b')
hold on
plot(FP.zscorei(:,4),FP.zscorei(:,3), 'm')
title('Z-scored L', 'fontsize', 16)
xlabel('Time (m)', 'fontsize', 14)

```

```
ylabel('Z-score', 'fontsize', 14)  
savefig(picz)
```