Plate Reader 2 -- experiment 1

Step 1: data loading and preparation

Add PLATERO set of functions to your working directory:

```
my = version('-release');
if str2double(my(1:4))<2020
addpath(genpath('rprev2020'))
else
addpath(genpath('r2020'))
end
```

Now, load the data resulting from the calibration experiment, written in "`filename`". This data is organized by sheets, where each sheet has one repetition of the measurements.

```
filename = "PlateReader2_exp1.xlsx";
colnames = {'WellID','Well','Concentration','G50','G60','G70','G80','G90','G120'};
[dataPR, indgfp] = readexperiment(filename,"A7:I103",[50:10:90,120],false,colnames,0);
size(dataPR)
```

```
ans = 1×2
        4608           5
```

Divide the dataset into the subset with medium values (`dataPRblk`) and the set with fluorescein values (`dataPRgfp`).

```
datPRblk = dataPR(~indgfp,:);
datPRgfp = dataPR(indgfp,:);
disp(strcat("This data set has ", string(size(datPRblk,1)),...
" BLK observations and ", string(size(datPRgfp,1)), ...
" GFP observations."))
```

```
This data set has 768 BLK observations and 3840 GFP observations.
```

Obtain the partition of the fluorescein dataset into the model building set (70%) and the model validation set (30%). A seed is set to ensure reproducibility of the results. The resulting subsets are stored as the `calibration_PR1.mat` and the `validation_PR1.mat` files.

```
rng(0207)
[datagfp_cal, datagfp_val] = cvsplit(datPRgfp, 0.7);
disp(strcat("The calibration data set has ", string(size(datagfp_cal,1)),...
" observations and the validation data set has ", ...
string(size(datagfp_val,1)), " observations."))
```

```
The calibration data set has 2640 observations and the validation data set has 1200 observations.
```

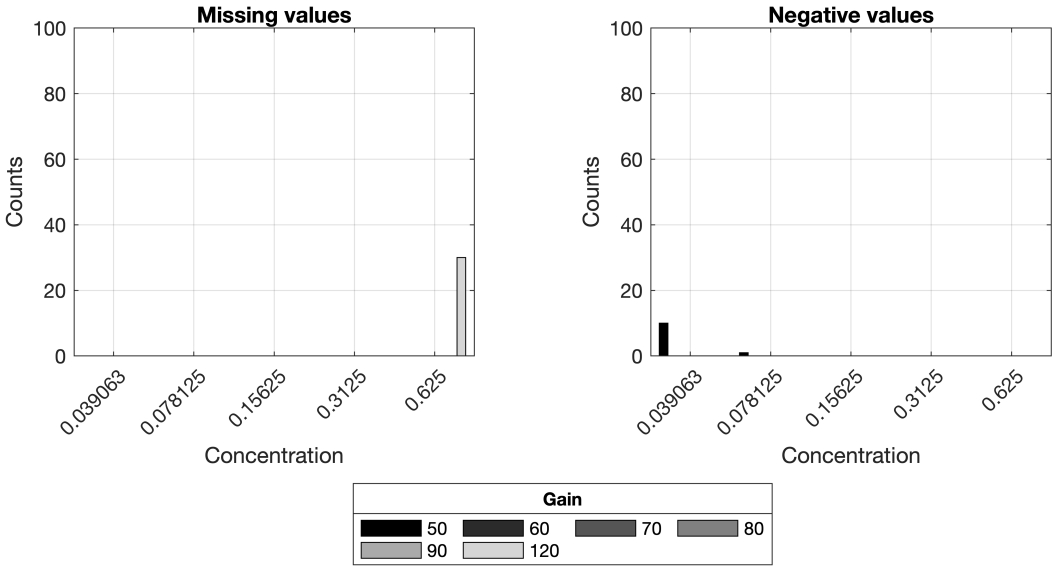
```
data_cal_pr2 = [datPRblk; datagfp_cal];
save('calibration_PR2.mat',"data_cal_pr2")
save('validation_PR2.mat',"datagfp_val")
```

Step 2: Model Building step

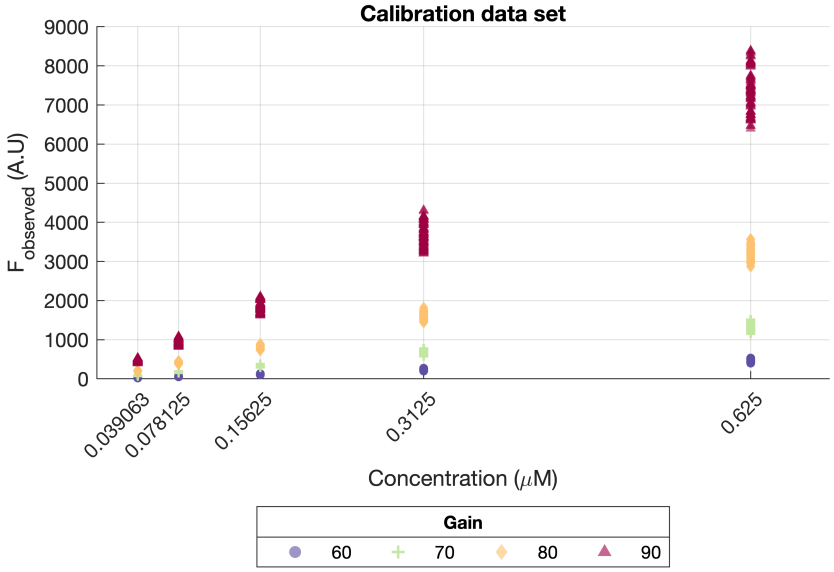
Load the calibration subset, fit the model and store the coefficients.

```
data_cal_pr2 = load("calibration_PR2.mat").data_cal_pr2;
[blk_data, flu_data_PR2e1] = explore_data(data_cal_pr2, 0);
```

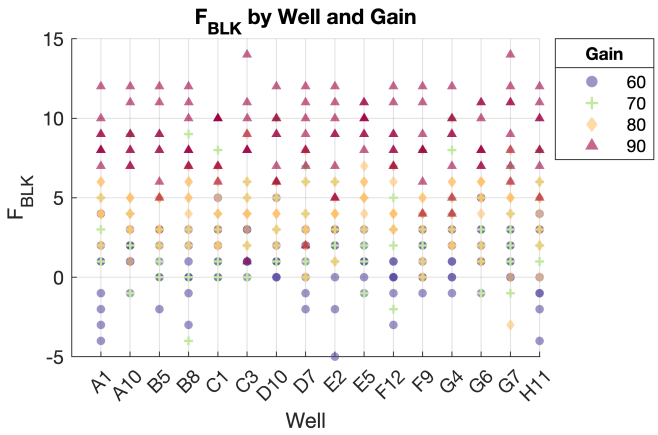
```
Explorative plot of missing data for each concentration level:
```

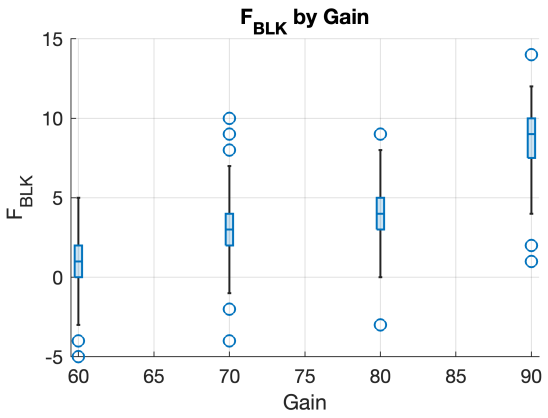



```
Concentration(s)0.625 had missing values
Concentration(s)0.0391, 0.0781 had negative values
Gain(s)120 had missing values
Gain(s)50 had negative values
Explorative plot of the raw F_observed Fluorescein data:
```




```
Explorative plot of the raw F_BLK data:
```






```
[flu_data_PR2e1, modelPR2e1, calmetrics_PR2e1] = fit_platero_model(blk_data, flu_data_
    "PR_2e1");
```

Fit f_G and plot corrected data (F_reporter):
ANOVA on coefficient b_1 for all levels of concentration

Analysis of Variance

Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F
Concentration	0.00102	4	0.00026	0.33	0.8598
Well(Concentration)	0.01214	50	0.00024	0.31	1
Error	0.30114	385	0.00078		
Total	0.3143	439			

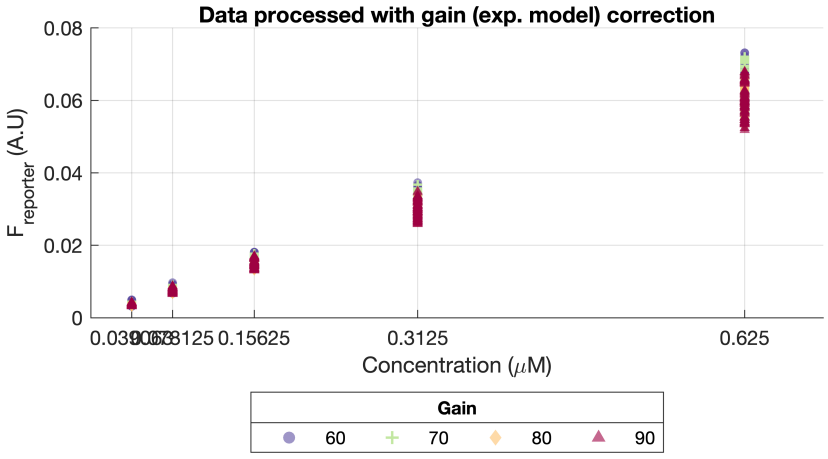
Constrained (Type III) sums of squares.

ANOVA on coefficient b_2 for all levels of concentration

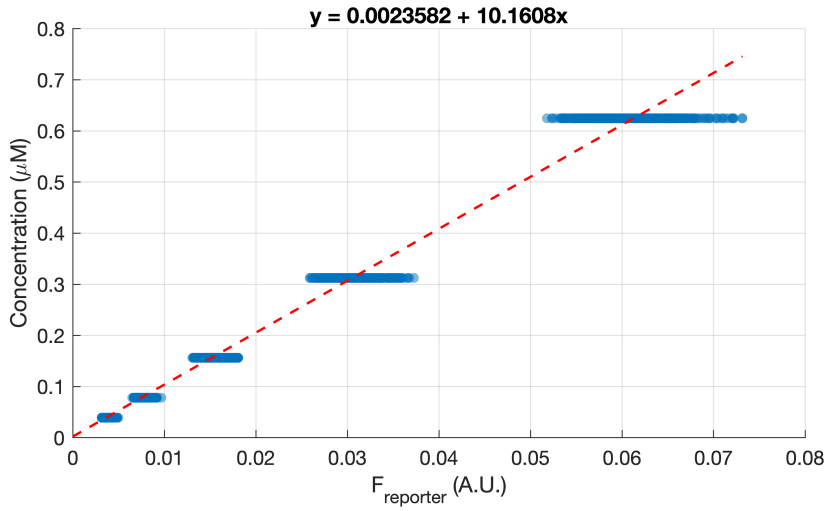
Analysis of Variance

Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F
Concentration	4.0355e-08	4	1.00888e-08	0.34	0.8477
Well(Concentration)	4.78955e-07	50	9.57909e-09	0.33	1
Error	1.12713e-05	385	2.92761e-08		
Total	1.17906e-05	439			

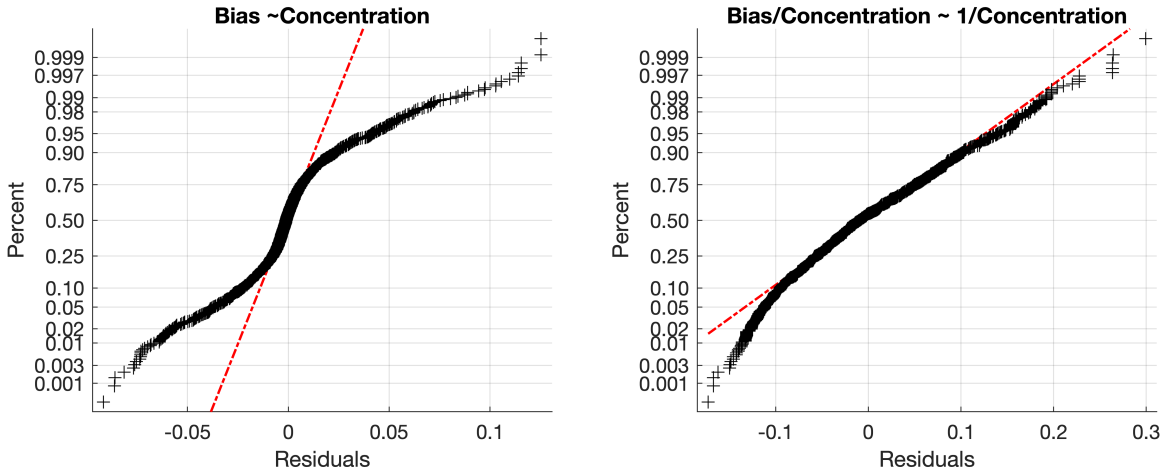
Constrained (Type III) sums of squares.

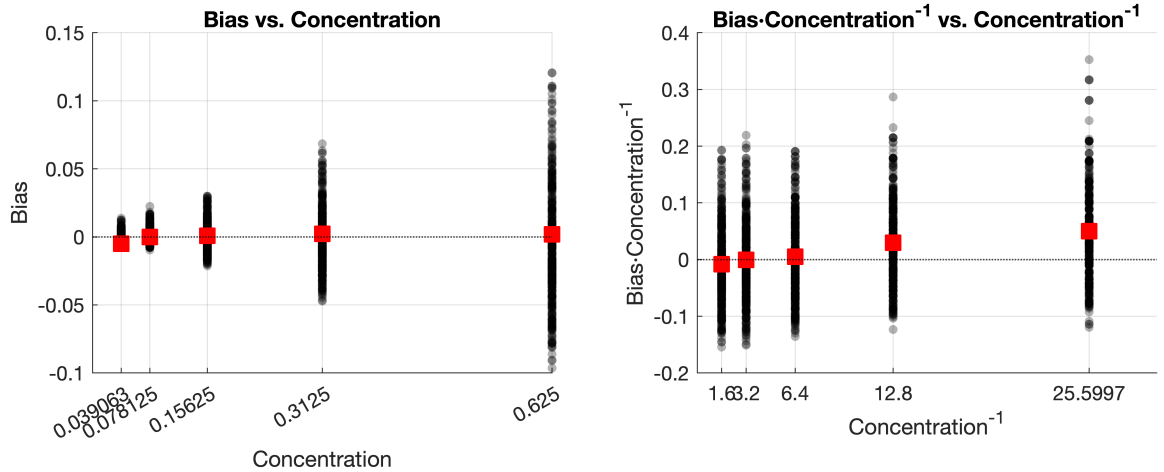


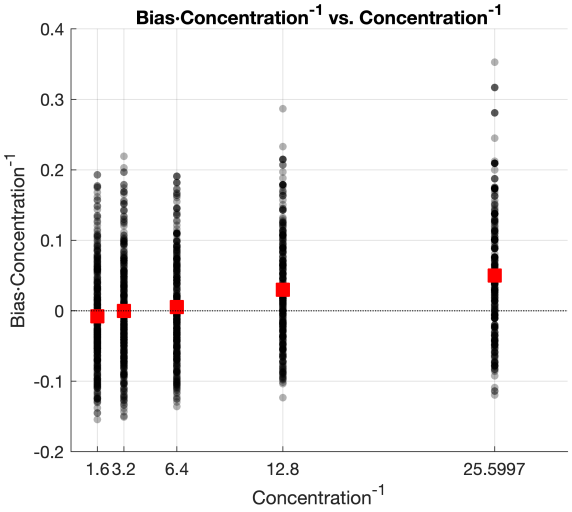
Fit f_UC and plot estimated concentration data (C):

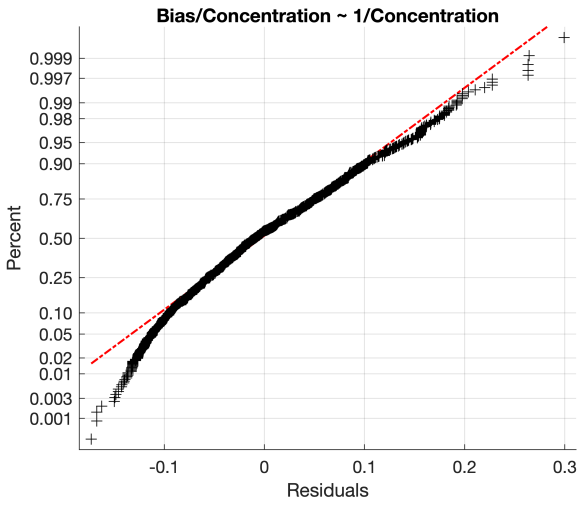


Analize the bias in the predictions and estimate the uncertainty in the predictions (s_Bias):









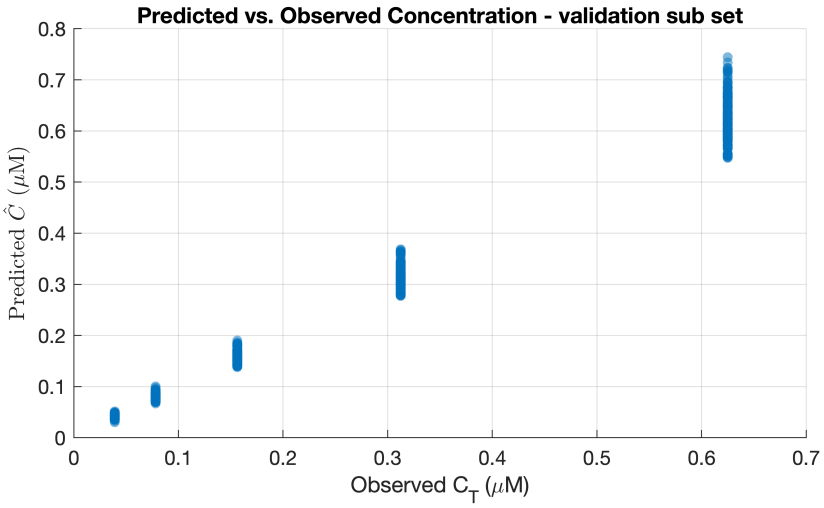
Compute error metrics for the Model Building step:

F_BLK (G = 60)	F_BLK (G = 70)	F_BLK (G = 80)	F_BLK (G = 90)	b1	b2	
1	3	4	9	0.18389	-0.00059598	0.00

Step 3: Model Validation step

```
datagfp_val = load("validation_PR2.mat").datagfp_val;
uG = unique(flu_data_PR2e1.Gain);
uC = unique(flu_data_PR2e1.Concentration);
data_val_pr2e1 = datagfp_val(ismember(datagfp_val.Gain, uG),:);
G = unique(data_val_pr2e1.Gain);

% Assign the correspoding F_BLK values to each observation F_obs
data_val_pr2e1.Fblk = repmat(modelPR2e1{:,1:4}', size(data_val_pr2e1,1)/length(G),1);

% Run the model on the validation set
[data_val_pr2e1, valmetrics_inrange, vprocv] = use_platero_model(data_val_pr2e1, ...
    modelPR2e1,"PR_2e1");
```

A 0 % of the observations was missing.
Plot the Validation dataset transformed to concentration units:



R&R Analysis:

R & R Analysis on measurements for C = 0.039063

Analysis of Variance

Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F
Reprod (Gain), C = (0.039063)	0.00007	3	2.2289e-05	2.13	0.0986
Replicates, C = (0.039063)	0.00006	4	1.52638e-05	1.46	0.2172
Error	0.00159	152	1.04565e-05		
Total	0.00172	159			

Constrained (Type III) sums of squares.

R & R Analysis on measurements for C = 0.078125

Analysis of Variance

Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F
Reprod (Gain), C = (0.078125)	0.00039	3	0.00013	3.75	0.0124
Replicates, C = (0.078125)	0.00036	4	0.00009	2.6	0.0382
Error	0.00521	152	0.00003		
Total	0.00595	159			

Constrained (Type III) sums of squares.

R & R Analysis on measurements for C = 0.15625

Analysis of Variance

Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F
Reprod (Gain), C = (0.15625)	0.00172	3	0.00057	5.08	0.0022
Replicates, C = (0.15625)	0.00042	4	0.0001	0.93	0.4504
Error	0.01719	152	0.00011		
Total	0.01933	159			

Constrained (Type III) sums of squares.

R & R Analysis on measurements for C = 0.3125

Analysis of Variance

Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F
Reprod (Gain), C = (0.3125)	0.00713	3	0.00238	5.41	0.0014
Replicates, C = (0.3125)	0.00087	4	0.00022	0.49	0.7395
Error	0.06675	152	0.00044		
Total	0.07476	159			

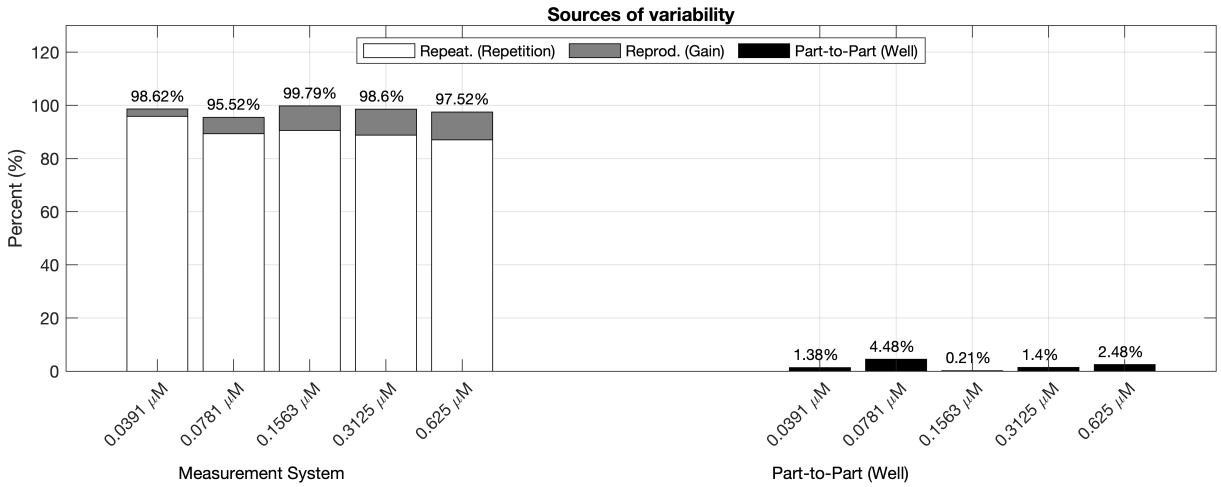
Constrained (Type III) sums of squares.

R & R Analysis on measurements for C = 0.625

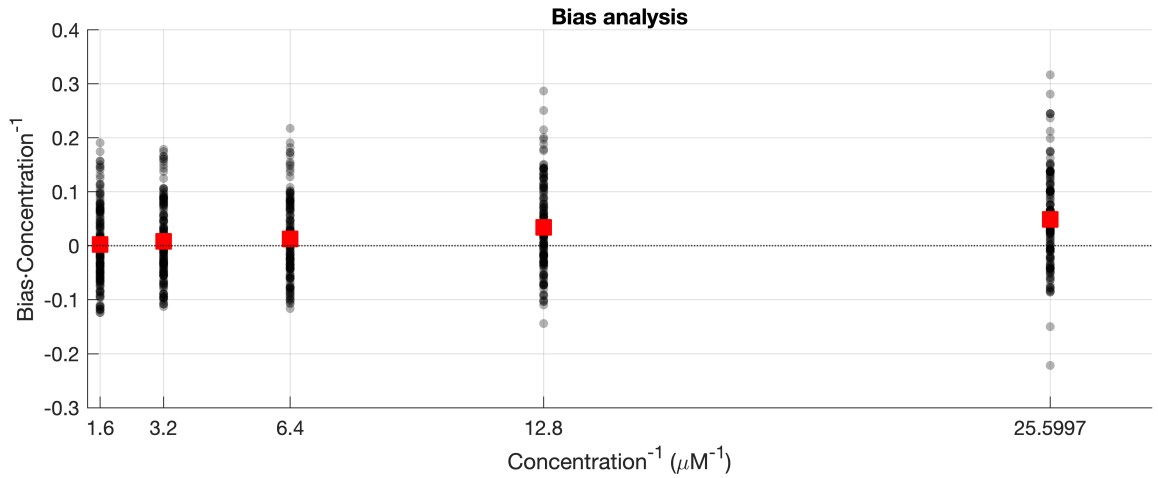
Analysis of Variance

Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F
Reprod (Gain), C = (0.625)	0.03055	3	0.01018	5.81	0.0009
Replicates, C = (0.625)	0.0134	4	0.00335	1.91	0.1112
Error	0.26629	152	0.00175		
Total	0.31024	159			

Constrained (Type III) sums of squares.



B&L Analysis:



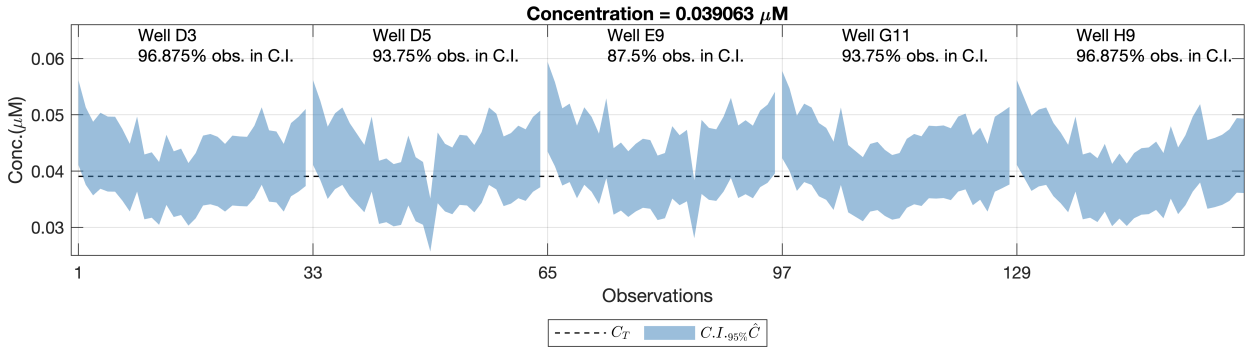
Linear regression model:
y ~ 1 + x1

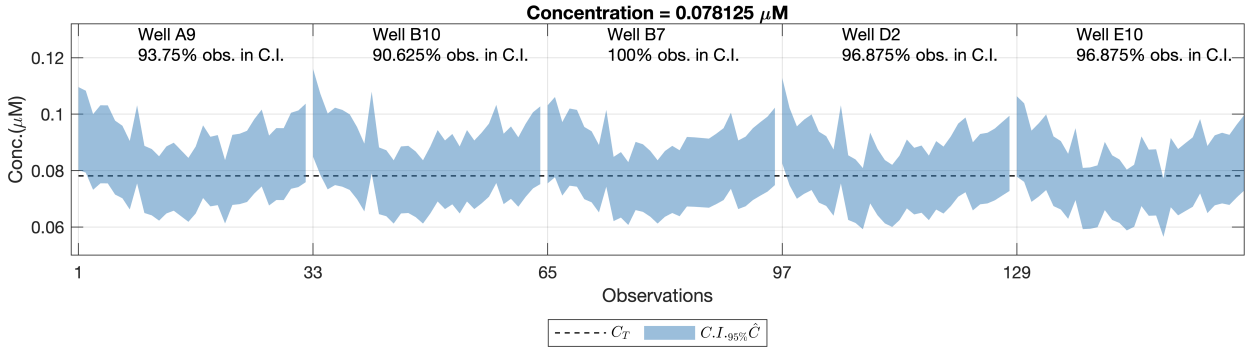
Estimated Coefficients:

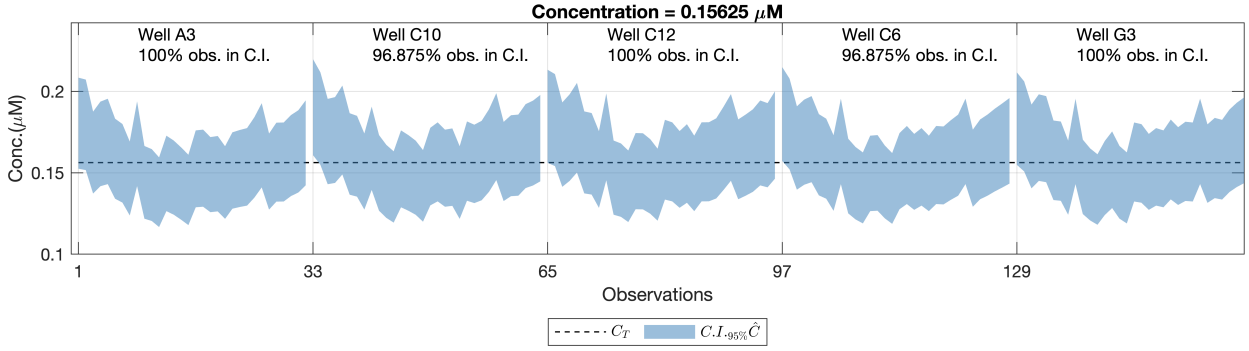
	Estimate	SE	tStat	pValue
(Intercept)	0.0019598	0.0040028	0.4896	0.62455
x1	0.0019459	0.00030294	6.4233	2.2893e-10

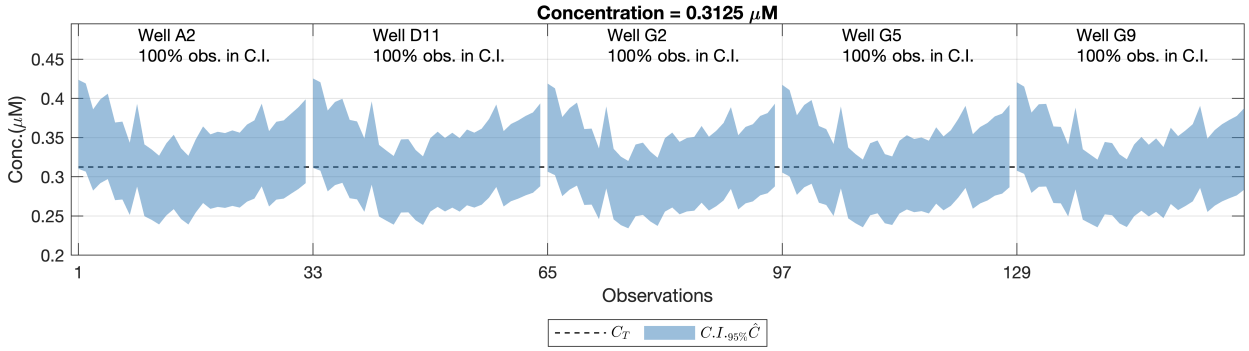
Number of observations: 800, Error degrees of freedom: 798
Root Mean Squared Error: 0.0748
R-squared: 0.0492, Adjusted R-Squared: 0.048
F-statistic vs. constant model: 41.3, p-value = 2.29e-10

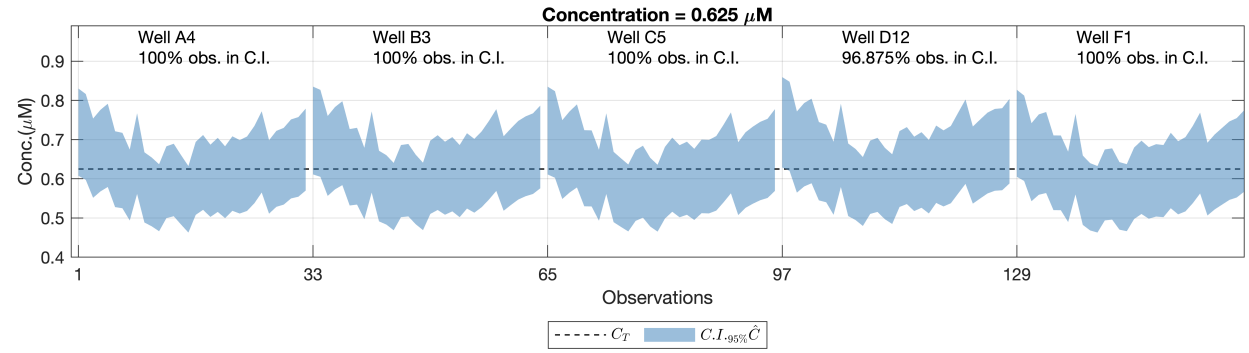
```
------------------------------------------------------------------------
Contribution of model terms to the total bias variability:
Bias Model - linear term (%): 0.196 %
Bias Model - bias term (%): 4.4521 %

Confidence Intervals and Error metrics:
       pctgeci: 97.5000
           mse: 5.1973e-04
        relerr: [800×1 double]
    minrelerror: 8.3939e-06
    maxrelerror: 0.3169
```












```
% Comparison between calibration-set and validation-set metrics
% load(strcat(dirdatasave,'cal_results.mat'))
perftable = table([calmetrics_PR2e1.mse;valmetrics_inrange.mse],...
[calmetrics_PR2e1.minrelerror;valmetrics_inrange.minrelerror]*100,...
[calmetrics_PR2e1.maxrelerror;valmetrics_inrange.maxrelerror]*100,...
'RowNames',{'Calibration', 'Validation (within range)'},...
'VariableNames',{'MSE','Min.Rel.Error (%)','Max.Rel.Error (%)'});
display(perftable)
```

perftable = 2×3 table

	MSE	Min.Rel.Error (%)	Max.Rel.Error (%)
1 Calibration	5.5309e-04	0.0045	35.2767
2 Validation (within range)	5.1973e-04	0.0008	31.6867