Statistics for data from Fig 2h Caillaud *et al* PLoS Biology

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13 August 2015

Pre-processing

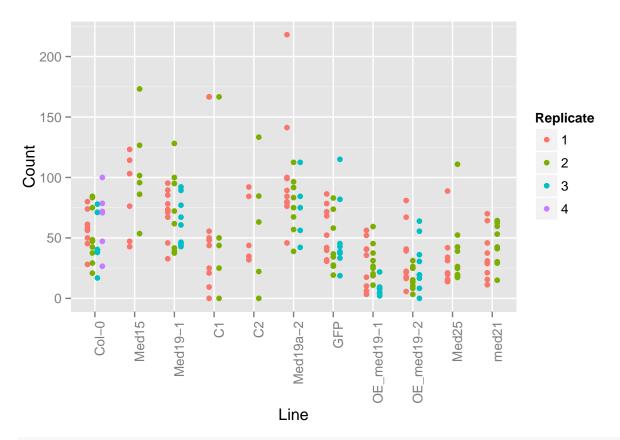
Marie-Cecille Caillaud (MCC) sent me an Excel file of all the haemocytometry measurements she made-file raw/MCC-Dan corrected.xslx. The file annotates figures from the same biological replicates as colours which I can't parse programmatically. I therefore added columns to the sheet stating the replicate number. I also removed spaces in column headers and saved the file as raw/MCC-Dan corrected Reps added.xlsx and exported the sheet with the data to a csv file fig_2h_data_manual.csv which I can operate on programmatically and will use as my input.

Use some Python to get the data file into better shape

```
header = []
results = []
with open('raw/fig_2h_data_manual.csv', 'r') as file:
  for 1 in file:
   l = 1.rstrip('\r\n')
   a = 1.split(',')
   if 1.startswith("Rep"):
      header = a
      for i in range(0,len(header),2):
        rep,line,count = a[i],header[i+1],a[i+1]
        if rep and line and count: ## if we have no empty values
          results.append([rep,line,count])
with open('data/reshaped_data.csv','w') as outfile:
  outfile.write("Replicate,Line,Count\n")
  for r in results:
    outfile.write(",".join(r) + "\n")
```

Load data, reorder as per Figure 2H and do a straightforward plot

ymax not defined: adjusting position using y instead



data

##		Replicate	Line	Count
##	1	1	Co1-0	61.25
##	2	1	Med19-1	78.13
##	3	1	Med15	76.25
##	4	1	Med19a-2	
##	5	1	GFP	30.68
##	6	1	OE_med19-1	
##	7	1	OE med19-2	
##	8	1	med21	64.29
##	9	1	Med25	14.58
##	10	1	C2	43.75
##	11	1	C1	48.08
##	12	1	Col-O	45.31
##	13	1	Med19-1	32.81
##	14	1	Med15	42.76
##	15	1	Med19a-2	79.86
##	16	1	GFP	31.82
##	17	1	OE_med19-1	52.04
##	18	1	OE_med19-2	40.82
##	19	1	med21	11.25
##	20	1	Med25	19.87
##	21	1	C2	84.38
##	22	1	C1	55.56
##	23	1	Col-O	56.25
##	24	1	Med19-1	95.31
##	25	1	Med15	103.13

```
## 26
                   Med19a-2 99.11
               1
## 27
                              78.41
               1
                         GFP
## 28
               1 OE med19-1
                              40.48
## 29
               1 OE_med19-2
                              39.29
## 30
                       med21
                              21.20
## 31
               1
                       Med25 15.74
## 32
               1
                          C2 31.91
                          C1 166.67
## 33
               1
## 34
               1
                       Col-0 57.03
## 35
                    Med19-1 71.14
               1
## 36
               1
                       Med15 47.22
## 37
                   Med19a-2 218.06
               1
## 38
                         GFP
                              39.77
               1
## 39
               1 OE_med19-1
                              56.25
## 40
               1 OE_med19-2
                              22.45
## 41
                       med21
                              45.83
## 42
               1
                       Med25
                              34.03
## 43
                          C2 34.82
                               9.38
## 44
               1
                          C1
## 45
                              28.13
               1
                       Col-O
## 46
               1
                    Med19-1 72.50
## 47
               1
                       Med15 123.21
## 48
                   Med19a-2 84.38
               1
                              86.36
## 49
               1
                         GFP
## 50
               1 OE_med19-1
                              35.71
## 51
               1 OE_med19-2
                              67.14
## 52
                       med21
                              28.85
## 53
               1
                       Med25
                              13.84
## 54
                          C2 92.11
               1
## 55
                          C1
                              21.01
               1
## 56
                       Col-0 58.75
               1
## 57
               1
                    Med19-1 85.42
## 58
                       Med15 114.29
## 59
                   Med19a-2
                             79.17
               1
## 60
                         GFP
                              42.05
               1
## 61
               1 OE_med19-1
                              10.20
               1 OE med19-2
## 62
                               5.71
## 63
               1
                      med21
                              15.63
                              31.25
## 64
               1
                       Med25
## 65
               2
                          C2 133.33
## 66
               1
                          C1
                               0.00
                              50.00
## 67
               1
                       Col-O
## 68
                    Med19-1 73.44
               1
## 69
               2
                       Med15 173.21
## 70
                   Med19a-2
                             89.29
               1
                              71.59
## 71
                         GFP
               1
## 72
               1 OE_med19-1
                              40.82
## 73
               1 OE_med19-2
                              80.95
## 74
                              37.50
               1
                       med21
## 75
               1
                       Med25
                              41.88
                          C2 63.16
## 76
               2
## 77
               1
                          C1 25.00
## 78
                       Col-0 80.00
               1
## 79
                    Med19-1 67.19
```

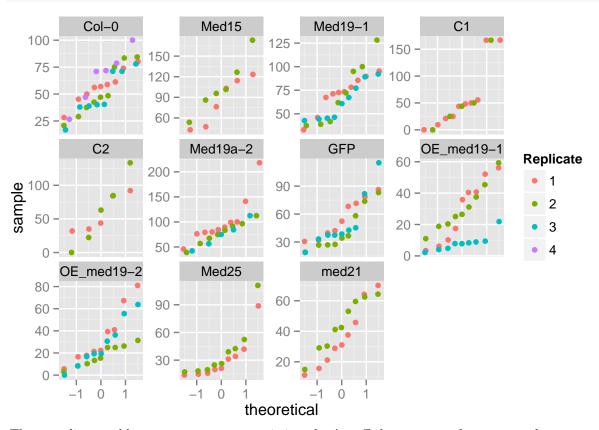
```
## 80
               2
                       Med15 53.57
## 81
                   Med19a-2 76.25
               1
## 82
                              68.18
               1
                         GFP
## 83
               1 OE_med19-1
                              17.53
## 84
               1 OE_med19-2
                              21.43
## 85
               1
                       med21
                              30.98
## 86
               1
                       Med25
                              21.05
## 87
                          C2 84.62
               2
                              43.75
## 88
               1
                          C1
## 89
               1
                       Col-0 73.96
## 90
               1
                    Med19-1 89.58
## 91
               2
                              95.70
                       Med15
## 92
               1
                   Med19a-2 141.25
## 93
               1
                         GFP
                              52.27
               1 OE_med19-1
## 94
                               3.37
## 95
               1 OE_med19-2
                              17.86
## 96
               1
                       med21
                              70.00
## 97
                       Med25 88.75
               1
## 98
                          C2 22.22
               2
## 99
               1
                          C1 166.67
## 100
               2
                       Col-0 37.50
## 101
               1
                    Med19-1 45.83
## 102
               2
                       Med15 126.60
## 103
               1
                    Med19a-2
                              45.83
## 104
               2
                         GFP
                              58.13
## 105
               2 OE med19-1
                              26.56
## 106
               2 OE_med19-2
                              10.42
## 107
               2
                       med21
                              53.13
## 108
               2
                             18.23
                       Med25
## 109
               2
                          C2
                               0.00
## 110
                              50.00
               1
                          C1
## 111
               2
                       Col-O
                              48.44
## 112
               2
                    Med19-1
                              61.76
## 113
               2
                       Med15 101.56
               2
## 114
                              38.89
                   Med19a-2
               2
                              27.50
## 115
                         GFP
               2 OE med19-1
## 116
                              37.50
## 117
               2 OE_med19-2
                               3.33
## 118
               2
                       med21
                              30.26
## 119
               2
                       Med25
                              24.84
## 120
                               0.00
               2
                          C1
## 121
               2
                              83.33
                       Col-O
## 122
               2
                    Med19-1
                              41.67
## 123
               2
                       Med15
                              86.11
## 124
               2
                    Med19a-2
                              96.43
## 125
               2
                         GFP
                              36.75
## 126
               2 OE_med19-1
                              45.31
## 127
               2 OE_med19-2
                               8.33
## 128
               2
                              42.50
                       med21
               2
## 129
                       Med25
                              19.68
               2
                              25.00
## 130
                          C1
               2
## 131
                       Col-0 84.38
## 132
               2
                    Med19-1 37.50
## 133
                   Med19a-2 112.50
```

```
## 134
                        GFP 73.75
## 135
               2 OE_med19-1
                              20.31
## 136
               2 OE med19-2
                              25.00
## 137
                      med21
                              64.29
## 138
               2
                              42.53
                      Med25
## 139
               2
                          C1 43.75
## 140
               2
                      Col-O
                             42.50
               2
                              72.22
## 141
                    Med19-1
                              67.35
## 142
               2
                   Med19a-2
## 143
               2
                         GFP
                              83.00
## 144
               2 OE_med19-1
                              59.38
## 145
               2 OE_med19-2
                              13.33
## 146
               2
                      med21
                              62.50
               2
## 147
                      Med25
                            17.30
## 148
               2
                          C1 166.67
## 149
               2
                      Col-O
                              20.83
## 150
               2
                    Med19-1
                              95.00
## 151
                   Med19a-2
                              75.00
## 152
               2
                         GFP
                              26.88
               2 OE med19-1
## 153
                              18.75
## 154
               2 OE_med19-2
                              25.00
## 155
                      med21
                              15.00
## 156
               2
                      Med25
                             39.06
               2
                              50.00
## 157
                          C1
## 158
               2
                      Col-0 75.00
## 159
               2
                    Med19-1 128.13
## 160
               2
                   Med19a-2
                              83.33
## 161
               2
                         GFP
                              26.67
               2 OE_med19-1
## 162
                              31.25
## 163
               2 OE_med19-2
                              31.25
## 164
               2
                              29.17
                      med21
## 165
               2
                      Med25 52.34
## 166
               2
                      Col-0 29.17
## 167
               2
                    Med19-1 100.00
               2
## 168
                   Med19a-2 56.94
               2
                              19.25
## 169
                         GFP
## 170
               2 OE med19-1
                              10.94
## 171
               2 OE_med19-2
                              26.39
## 172
               2
                      med21
                              59.62
## 173
               2
                      Med25
                             26.32
## 174
                      Col-0 47.22
## 175
               2
                    Med19-1
                              38.89
## 176
               2
                   Med19a-2
                              91.67
## 177
               2
                         GFP
                              34.25
## 178
               2 OE_med19-1
                              25.00
## 179
               2 OE_med19-2
                              15.28
## 180
               2
                      med21 41.07
               2
## 181
                      Med25 110.94
## 182
               3
                      Col-0 16.88
## 183
               3
                    Med19-1
                             44.32
               3
                             75.00
## 184
                   Med19a-2
               3
## 185
                         GFP
                              42.86
               3 OE_med19-1
## 186
                               7.81
## 187
               3 OE med19-2
                               0.00
```

```
## 188
               3
                       Col-0 71.15
## 189
               3
                     Med19-1
                              89.29
## 190
               3
                    Med19a-2
                              56.25
## 191
               3
                         GFP
                              18.75
## 192
               3 OE_med19-1
                               21.88
## 193
               3 OE med19-2
                              36.11
## 194
               3
                       Co1-0
                              71.15
## 195
               3
                     Med19-1
                              60.71
## 196
               3
                    Med19a-2 112.50
## 197
                3
                              33.33
                         GFP
## 198
               3 OE_med19-1
                                7.81
                3 OE_med19-2
## 199
                              30.56
## 200
                       Col-O
                              38.89
                3
## 201
               3
                     Med19-1
                              45.31
## 202
               3
                    Med19a-2
                              84.38
## 203
                3
                         GFP
                               45.31
## 204
               3 OE_med19-1
                                8.75
               3 OE med19-2
## 205
                              19.44
## 206
               3
                       Col-O
                              40.00
               3
## 207
                     Med19-1
                              67.19
## 208
               3
                    Med19a-2
                              42.19
## 209
               3
                         GFP
                              81.94
## 210
               3 OE_med19-1
                                4.17
## 211
                3 OE med19-2
                              63.89
## 212
                3
                       Col-0
                              38.00
## 213
               3
                     Med19-1
                              42.86
## 214
                3
                         GFP
                              38.64
## 215
                3 OE_med19-1
                                8.33
                3 OE_med19-2
## 216
                              16.67
## 217
                3
                       Col-O
                              77.94
## 218
                3
                     Med19-1
                              92.19
## 219
               3
                         GFP
                              37.50
## 220
                3 OE_med19-1
                                4.86
## 221
               3 OE_med19-2
                              55.56
## 222
               3
                       Col-0
                              40.48
## 223
               3
                     Med19-1
                              77.08
## 224
               3
                         GFP 115.00
## 225
               3 OE_med19-1
                                9.38
## 226
               3 OE med19-2
                              19.44
## 227
                4
                       Col-0 100.00
## 228
               3
                     Med19-1
                              46.43
## 229
               3
                         GFP
                              37.50
## 230
                3 OE_med19-1
                                2.08
## 231
                3 OE_med19-2
                                8.33
## 232
                       Co1-0
                              26.56
                4
## 233
                       Col-O
                              47.22
                4
## 234
                4
                       Col-O
                              78.57
## 235
                4
                       Col-O
                              70.83
## 236
                              71.88
                       Col-O
```

The data look ok, a few outliers in Med19a-2 and C1 that could affect summary statistics. Let's do some qqplots and see how they lie.

#qnorm is default distribution - we are testing for a normal distribution
ggplot(data, aes(sample=Count)) + geom_jitter(stat="qq", aes(colour=Replicate)) + facet_wrap(~ Line,



Those outliers could mess up summary statistics, they're off the curve, we have no good reason to ditch them though. I suppose they mean that occasionally the method used (spore counting) throws up some very extreme numbers. Overall these plots are ok, the variation seems normally distributed on the whole.

Let's have a look at summary statistics:

```
library(plyr)
summary <- ddply(data, "Line", summarise, mean=mean(Count), median=median(Count), diff=abs(mean(Count) - mesummary</pre>
```

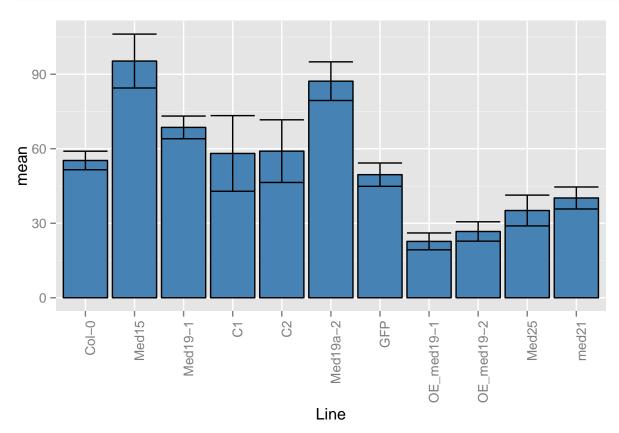
```
##
            Line
                     mean median
                                        diff
                                              std_dev
                                                         std_err
           Col-0 55.26875 53.125
## 1
                                   2.1437500 21.11359
                                                        3.732392
## 2
           Med15 95.30083 98.630
                                   3.3291667 37.59931 10.853987
## 3
         Med19-1 68.58889 71.140
                                   2.5511111 23.73107
                                                        4.567046
## 4
              C1 58.10267 43.750 14.3526667 58.98184 15.229045
              C2 59.03000 53.455
## 5
                                   5.5750000 39.89419
## 6
        Med19a-2 87.20130 83.330
                                   3.8713043 37.32340
                                                        7.782467
##
             GFP 49.56074 39.770
                                   9.7907407 24.40441
                                                        4.696630
      OE_med19-1 22.69333 18.750
                                   3.9433333 17.61271
##
  8
                                                        3.389568
## 9
      OE_med19-2 26.68185 21.430
                                   5.2518519 20.25470
                                                        3.898019
## 10
           Med25 35.12389 25.580
                                   9.5438889 26.30013
                                                        6.199001
## 11
           med21 40.17056 39.285
                                   0.8855556 18.79839
                                                        4.430822
```

The summary stats seem fine overall, similar SD and SE and not much drift of the median from the mean, the concern again is Med19a-2 and C1 with the high standard deviation and mean dragged up by that couple of points.

Does a bar chart imply a higher effect than we see generally?

Let's make a bar graph with error bars on that first scatter to see how using a standard bar chart might be misleading our thinking.





The barchart is definitely suggesting a higher overall effect than we see from the individual replicates in the scatter plot for Med19a-2 and C1 My conclusion here is that although the mean is calculated correctly, it's just that the mean is a slightly misleading number to boil our data down to in this case. Also that very slight increase in standard error isn't giving us a clue as to that messy single outlier. Taken together the mean and SE plotted like this convince of us a bigger effect in general so the plot style isn't helpful.

Significance Tests

I need to boil down the data to the biological replicates.

library(reshape)

```
##
## Attaching package: 'reshape'
##
## The following objects are masked from 'package:plyr':
##
## rename, round_any
```

```
bioreps <- cast(data, Line~Replicate, mean)</pre>
```

Using Count as value column. Use the value argument to cast to override this choice

```
bioreps <- melt(bioreps)
bioreps</pre>
```

```
##
               Line
                          value Replicate
## X1
              Col-O
                      56.742222
                                         1
              Med15 84.476667
## X1.1
                                         1
## X1.2
            Med19-1
                     71.135000
                                         1
## X1.3
                  C1 58.612000
                                         1
## X1.4
                  C2 57.394000
                                         1
## X1.5
           Med19a-2 101.320000
                                         1
## X1.6
                 GFP
                      55.681111
                                         1
## X1.7 OE med19-1
                      29.183333
                                         1
## X1.8 OE_med19-2
                      34.675556
                                         1
## X1.9
              Med25
                      31.221111
                                         1
## X1.10
              med21
                      36.170000
                                         1
## X2
              Col-O
                      52.041111
                                         2
                                         2
## X2.1
              Med15 106.125000
## X2.2
            Med19-1
                                         2
                     71.896250
                                         2
## X2.3
                  C1
                     57.084000
## X2.4
                  C2
                      60.666000
                                         2
## X2.5
                      77.763750
                                         2
           Med19a-2
                                         2
## X2.6
                 GFP
                      42.908889
                                         2
## X2.7 OE med19-1 30.555556
## X2.8 OE_med19-2
                      17.592222
                                         2
## X2.9
              Med25
                      39.026667
                                         2
## X2.10
              med21
                      44.171111
                                         2
                                         3
## X3
              Col-O
                      49.311250
## X3.1
                                         3
              Med15
                            {\tt NaN}
                                         3
## X3.2
            Med19-1
                      62.820000
## X3.3
                                         3
                  C1
                            NaN
## X3.4
                  C2
                            NaN
                                         3
## X3.5
           Med19a-2
                      74.064000
                                         3
## X3.6
                 GFP
                      50.092222
                                         3
## X3.7 OE_med19-1
                                         3
                       8.341111
## X3.8 OE_med19-2
                                         3
                      27.777778
## X3.9
              Med25
                                         3
                            NaN
## X3.10
                                         3
              med21
                            NaN
## X4
                                         4
              Col-O
                      65.843333
## X4.1
                                         4
              Med15
                            NaN
## X4.2
            Med19-1
                                         4
                            NaN
                                         4
## X4.3
                  C1
                            NaN
## X4.4
                                         4
                  C2
                            {\tt NaN}
## X4.5
           Med19a-2
                            NaN
                                         4
## X4.6
                 GFP
                            NaN
                                         4
## X4.7
                                         4
        OE_med19-1
                            {\tt NaN}
                                         4
## X4.8 OE_med19-2
                            {\tt NaN}
## X4.9
              Med25
                            NaN
                                         4
## X4.10
              med21
                            NaN
                                         4
```

I'll do an ANOVA and Tukey's HSD for multiple comparisons.

```
### ANOVA and Tukey's HSD on all pairwise - though really only interested in VS Col-O the control
fit <- aov(lm(value ~ Line,data=bioreps))
TukeyHSD(fit)</pre>
```

```
Tukey multiple comparisons of means
##
##
       95% family-wise confidence level
##
## Fit: aov(formula = lm(value ~ Line, data = bioreps))
## $Line
                                diff
##
                                             lwr
                                                          upr
                                                                  p adj
## Med15-Col-0
                           39.316354
                                       10.505345
                                                  68.1273629 0.0033725
## Med19-1-Col-0
                          12.632604
                                      -12.776317
                                                  38.0415256 0.7560958
## C1-Col-0
                                      -26.947488
                                                  30.6745296 1.0000000
                           1.863521
## C2-Co1-0
                           3.045521
                                      -25.765488
                                                  31.8565296 0.9999983
## Med19a-2-Col-0
                          28.398104
                                        2.989183
                                                  53.8070256 0.0213201
## GFP-Col-0
                          -6.423738
                                     -31.832660
                                                  18.9851830 0.9961024
## OE med19-1-Col-0
                                      -58.700067
                                                  -7.8822244 0.0050706
                          -33.291146
## OE med19-2-Col-0
                         -29.302627
                                      -54.711549
                                                  -3.8937059 0.0163874
## Med25-Col-0
                         -20.860590
                                      -49.671599
                                                   7.9504185 0.2940977
                                                  12.9970851 0.6466693
## med21-Col-0
                         -15.813924
                                      -44.624932
## Med19-1-Med15
                          -26.683750
                                      -57.053220
                                                   3.6857198 0.1149884
## C1-Med15
                         -37.452833
                                      -70.720921
                                                  -4.1847460 0.0200774
## C2-Med15
                         -36.270833
                                     -69.538921
                                                  -3.0027460 0.0260686
## Med19a-2-Med15
                         -10.918250
                                      -41.287720
                                                  19.4512198 0.9539136
## GFP-Med15
                          -45.740093
                                      -76.109562 -15.3706228 0.0011758
## OE_med19-1-Med15
                         -72.607500 -102.976970 -42.2380302 0.0000029
## OE_med19-2-Med15
                          -68.618981
                                      -98.988451 -38.2495117 0.0000066
## Med25-Med15
                          -60.176944
                                      -93.445032 -26.9088571 0.0001323
## med21-Med15
                         -55.130278
                                      -88.398365 -21.8621905 0.0003896
## C1-Med19-1
                         -10.769083
                                      -41.138553
                                                  19.6003864 0.9576648
## C2-Med19-1
                          -9.587083
                                      -39.956553
                                                  20.7823864 0.9801671
## Med19a-2-Med19-1
                          15.765500
                                      -11.397780
                                                  42.9287795 0.5771317
## GFP-Med19-1
                         -19.056343
                                      -46.219622
                                                   8.1069370 0.3317637
## OE med19-1-Med19-1
                         -45.923750
                                      -73.087030 -18.7604705 0.0003061
## OE_med19-2-Med19-1
                          -41.935231
                                      -69.098511 -14.7719519 0.0008904
## Med25-Med19-1
                          -33.493194
                                      -63.862664
                                                  -3.1237247 0.0237690
## med21-Med19-1
                         -28.446528
                                     -58.815998
                                                   1.9229420 0.0778077
## C2-C1
                           1.182000
                                      -32.086087
                                                  34.4500873 1.0000000
## Med19a-2-C1
                                       -3.834886
                                                  56.9040531 0.1187604
                           26.534583
## GFP-C1
                           -8.287259
                                      -38.656729
                                                  22.0822105 0.9930153
## OE_med19-1-C1
                         -35.154667
                                      -65.524136
                                                  -4.7851969 0.0158700
## OE_med19-2-C1
                         -31.166148
                                      -61.535618
                                                  -0.7966784 0.0414666
## Med25-C1
                          -22.724111
                                      -55.992198
                                                  10.5439762 0.3649176
## med21-C1
                          -17.677444
                                      -50.945532
                                                  15.5906429 0.6847841
## Med19a-2-C2
                          25.352583
                                       -5.016886
                                                  55.7220531 0.1526233
## GFP-C2
                          -9.469259
                                      -39.838729
                                                  20.9002105 0.9817785
## OE_med19-1-C2
                          -36.336667
                                      -66.706136
                                                  -5.9671969 0.0118798
## OE med19-2-C2
                         -32.348148
                                      -62.717618
                                                  -1.9786784 0.0313070
## Med25-C2
                         -23.906111
                                      -57.174198
                                                   9.3619762 0.3029590
## med21-C2
                         -18.859444
                                      -52.127532
                                                  14.4086429 0.6069765
## GFP-Med19a-2
                         -34.821843
                                     -61.985122
                                                 -7.6585630 0.0062678
```

```
## OE med19-1-Med19a-2
                         -61.689250 -88.852530 -34.5259705 0.0000061
## OE_med19-2-Med19a-2
                         -57.700731 -84.864011 -30.5374519 0.0000157
## Med25-Med19a-2
                         -49.258694
                                    -79.628164 -18.8892247 0.0005027
## med21-Med19a-2
                                    -74.581498 -13.8425580 0.0017075
                         -44.212028
## OE med19-1-GFP
                         -26.867407
                                     -54.030687
                                                  0.2958721 0.0540066
## OE med19-2-GFP
                         -22.878889 -50.042168
                                                  4.2843907 0.1455538
## Med25-GFP
                         -14.436852
                                    -44.806322 15.9326179 0.7982744
## med21-GFP
                          -9.390185
                                     -39.759655
                                                 20.9792846 0.9828033
## OE_med19-2-OE_med19-1
                           3.988519
                                     -23.174761
                                                 31.1517981 0.9999631
## Med25-OE_med19-1
                          12.430556
                                    -17.938914
                                                 42.8000253 0.9027929
## med21-0E_med19-1
                          17.477222
                                     -12.892248
                                                 47.8466920 0.5879787
## Med25-OE_med19-2
                           8.442037
                                     -21.927433
                                                 38.8115068 0.9919835
## med21-0E_med19-2
                          13.488704
                                     -16.880766
                                                 43.8581735 0.8525396
                                     -28.221421
## med21-Med25
                           5.046667
                                                 38.3147540 0.9999503
```

A long table, but it's showing the overexpressers <code>OE_med19-1</code> and <code>OE_med19-2</code> are different from the <code>Col-O</code> control, as is the one with the noted high outliers <code>Med19a-2</code> and also <code>Med15</code>.

P-Hacking

Let's see how removing those high (>=150) outliers affects the p-values, see if any signficance we have is coming from one or two atypical data.

```
under_150 <- data[data$value < 150, ]
bioreps_under150 <- cast(data, Line~Replicate, mean)</pre>
```

Using Count as value column. Use the value argument to cast to override this choice

```
bioreps_under150 <- melt(bioreps_under150)
fit <- aov(lm(value ~ Line,data=bioreps_under150))
TukeyHSD(fit)</pre>
```

```
##
     Tukey multiple comparisons of means
##
       95% family-wise confidence level
##
## Fit: aov(formula = lm(value ~ Line, data = bioreps_under150))
##
## $Line
##
                                diff
                                             lwr
                                                         upr
                                                                  p adj
## Med15-Col-0
                          39.316354
                                       10.505345
                                                  68.1273629 0.0033725
## Med19-1-Col-0
                          12.632604
                                      -12.776317
                                                  38.0415256 0.7560958
## C1-Co1-0
                           1.863521
                                     -26.947488
                                                  30.6745296 1.0000000
## C2-Co1-0
                           3.045521
                                      -25.765488
                                                  31.8565296 0.9999983
                                                  53.8070256 0.0213201
## Med19a-2-Col-0
                          28.398104
                                        2.989183
## GFP-Col-0
                          -6.423738
                                                  18.9851830 0.9961024
                                     -31.832660
## OE_med19-1-Col-0
                         -33.291146
                                     -58.700067
                                                  -7.8822244 0.0050706
## OE med19-2-Col-0
                                      -54.711549
                         -29.302627
                                                  -3.8937059 0.0163874
## Med25-Col-0
                         -20.860590
                                     -49.671599
                                                   7.9504185 0.2940977
## med21-Col-0
                                      -44.624932
                                                  12.9970851 0.6466693
                         -15.813924
## Med19-1-Med15
                         -26.683750
                                     -57.053220
                                                   3.6857198 0.1149884
## C1-Med15
                         -37.452833
                                     -70.720921
                                                  -4.1847460 0.0200774
## C2-Med15
                         -36.270833
                                     -69.538921 -3.0027460 0.0260686
```

```
## Med19a-2-Med15
                         -10.918250
                                     -41.287720 19.4512198 0.9539136
## GFP-Med15
                         -45.740093
                                     -76.109562 -15.3706228 0.0011758
## OE med19-1-Med15
                         -72.607500 -102.976970 -42.2380302 0.0000029
## OE_med19-2-Med15
                                     -98.988451 -38.2495117 0.0000066
                         -68.618981
## Med25-Med15
                         -60.176944
                                     -93.445032 -26.9088571 0.0001323
## med21-Med15
                         -55.130278
                                     -88.398365 -21.8621905 0.0003896
## C1-Med19-1
                         -10.769083
                                     -41.138553
                                                  19.6003864 0.9576648
## C2-Med19-1
                          -9.587083
                                     -39.956553
                                                  20.7823864 0.9801671
## Med19a-2-Med19-1
                                     -11.397780
                                                  42.9287795 0.5771317
                          15.765500
## GFP-Med19-1
                         -19.056343
                                     -46.219622
                                                   8.1069370 0.3317637
## OE_med19-1-Med19-1
                         -45.923750
                                     -73.087030 -18.7604705 0.0003061
## OE_med19-2-Med19-1
                         -41.935231
                                     -69.098511 -14.7719519 0.0008904
## Med25-Med19-1
                         -33.493194
                                     -63.862664
                                                  -3.1237247 0.0237690
                         -28.446528
## med21-Med19-1
                                     -58.815998
                                                   1.9229420 0.0778077
## C2-C1
                           1.182000
                                     -32.086087
                                                  34.4500873 1.0000000
## Med19a-2-C1
                          26.534583
                                      -3.834886
                                                  56.9040531 0.1187604
## GFP-C1
                                     -38.656729
                                                  22.0822105 0.9930153
                          -8.287259
## OE med19-1-C1
                         -35.154667
                                      -65.524136
                                                  -4.7851969 0.0158700
## OE_med19-2-C1
                         -31.166148
                                     -61.535618
                                                  -0.7966784 0.0414666
## Med25-C1
                         -22.724111
                                      -55.992198
                                                  10.5439762 0.3649176
## med21-C1
                         -17.677444
                                     -50.945532
                                                  15.5906429 0.6847841
## Med19a-2-C2
                                      -5.016886
                          25.352583
                                                  55.7220531 0.1526233
## GFP-C2
                                     -39.838729
                                                  20.9002105 0.9817785
                          -9.469259
## OE med19-1-C2
                         -36.336667
                                     -66.706136
                                                  -5.9671969 0.0118798
## OE med19-2-C2
                         -32.348148
                                     -62.717618
                                                 -1.9786784 0.0313070
## Med25-C2
                         -23.906111
                                     -57.174198
                                                   9.3619762 0.3029590
## med21-C2
                                     -52.127532
                         -18.859444
                                                  14.4086429 0.6069765
## GFP-Med19a-2
                         -34.821843
                                     -61.985122
                                                  -7.6585630 0.0062678
## OE_med19-1-Med19a-2
                         -61.689250
                                     -88.852530 -34.5259705 0.0000061
## OE_med19-2-Med19a-2
                         -57.700731
                                     -84.864011 -30.5374519 0.0000157
## Med25-Med19a-2
                         -49.258694
                                     -79.628164 -18.8892247 0.0005027
## med21-Med19a-2
                         -44.212028
                                     -74.581498 -13.8425580 0.0017075
## OE_med19-1-GFP
                         -26.867407
                                     -54.030687
                                                   0.2958721 0.0540066
## OE_med19-2-GFP
                         -22.878889
                                     -50.042168
                                                   4.2843907 0.1455538
## Med25-GFP
                         -14.436852
                                     -44.806322
                                                  15.9326179 0.7982744
## med21-GFP
                          -9.390185
                                     -39.759655
                                                  20.9792846 0.9828033
## OE med19-2-OE med19-1
                           3.988519
                                     -23.174761
                                                  31.1517981 0.9999631
## Med25-OE_med19-1
                                     -17.938914
                                                  42.8000253 0.9027929
                          12.430556
## med21-0E_med19-1
                          17.477222
                                     -12.892248
                                                  47.8466920 0.5879787
## Med25-OE_med19-2
                           8.442037
                                     -21.927433
                                                  38.8115068 0.9919835
## med21-0E med19-2
                          13.488704
                                     -16.880766
                                                  43.8581735 0.8525396
## med21-Med25
                           5.046667
                                     -28.221421
                                                  38.3147540 0.9999503
```

Looks good! The same Lines come up as significant - the outliers aren't messing with the overall significance result.

More P-Hacking - ditching data originally in Figure 2H!

According to MCC and JJ then the lines of interest are really the med19-1, Med19a-2, OE_med19-1 and OE_med19-2. Let's do the same tests for the restricted set and see if it substantially affects the result.

```
of_interest <- bioreps_under150[bioreps_under150$Line %in% c("Col-0", "Med19-1", "Med19a-2", "OE_med19-fit <- aov(lm(value ~ Line,data=of_interest))
```

TukeyHSD(fit)

```
##
     Tukey multiple comparisons of means
##
       95% family-wise confidence level
##
## Fit: aov(formula = lm(value ~ Line, data = of_interest))
##
## $Line
##
                               diff
                                           lwr
                                                       upr
                                                               p adj
## Med19-1-Col-0
                          12.632604 -12.105257
                                                 37.370466 0.4979780
## Med19a-2-Col-0
                                                53.135966 0.0228532
                          28.398104
                                      3.660243
## OE_med19-1-Col-0
                         -33.291146 -58.029007
                                                -8.553284 0.0081105
## OE med19-2-Col-0
                         -29.302627 -54.040489
                                                -4.564766 0.0188384
## Med19a-2-Med19-1
                          15.765500 -10.680386 42.211386 0.3584632
## OE_med19-1-Med19-1
                         -45.923750 -72.369636 -19.477864 0.0011733
## OE med19-2-Med19-1
                         -41.935231 -68.381118 -15.489345 0.0024215
## OE_med19-1-Med19a-2
                         -61.689250 -88.135136 -35.243364 0.0000889
## OE_med19-2-Med19a-2
                         -57.700731 -84.146618 -31.254845 0.0001638
## OE_med19-2-OE_med19-1
                           3.988519 -22.457368 30.434405 0.9869299
```

The result is not substantially different from before, the same lines show up as significantly different, that is Med19a-2, OE_med19-1, OE_med19-2 and Med15 are significantly different from the Col-0 control. Med19-1 is not

Conclusion

The Med19-2 and Med15 lines get significantly more spores than the Col-0 wild-type and the two over-expressors of Med19 show significantly fewer spores than Col-0. There is no evidence for difference from the wild-type and other lines.

```
## Saving 6.5 x 4.5 in image
## ymax not defined: adjusting position using y instead
```