

3-20-18 Calculations using 28-day ddPCR data

Purpose: Gail calculated outliers and normalized the data to the UniSp5 spike-in. I observed there was no correlation between target expression and UniSp5 measures, suggesting that whatever is affecting the measurement of the spike-in is likely not affecting measurements in the target miRNA. Therefore I am re-doing the analysis on raw target data.

Tests:

The identification of outliers suggests that the data is not normally distributed. I will use a Shapiro-Wilk Normality Test for each group for each target and test for normality. The webtool located here was used: <http://sdittami.altervista.org/shapirotest/ShapiroTest.html>

Raw data for miR-182 is below and used to calculate normality

miR-182	
6K DEHP	C
0.915	2.25
7.2	11.65
4.05	1.015
3.25	0.66
7.25	2.1
8.6	
6.95	
3.6	

Control group:

Results:

n = 5

Mean = 3.535

SD = 4.587474795571088

W = 0.6865050686367097

Threshold (p=0.01) = 0.6859999895095825 --> HO accepted

Threshold (p=0.05) = 0.7620000243186951 --> HO rejected

Threshold (p=0.10) = 0.8059999942779541 --> HO rejected

--> Your data is not normally distributed p<0.05

From <<http://sdittami.altervista.org/shapirotest/ShapiroTest.html>>

6K group:

Results:

n = 8
Mean = 5.226875000000001
SD = 2.6421162209486546
W = 0.920491999472594
Threshold (p=0.01) = 0.7490000128746033 --> HO accepted
Threshold (p=0.05) = 0.8180000185966492 --> HO accepted
Threshold (p=0.10) = 0.8510000109672546 --> HO accepted
--> Your data seems normal

From <<http://sdittami.altervista.org/shapirotest/ShapiroTest.html>>

Results for miR-182: They indicate the 6K treatment group is not normally distributed. Therefore a non-parametric test must be used to examine the data.

A Mann-Whitney U Test was used to compare control to the treatment groups for miR-182. The webtool is located here: <http://www.socscistatistics.com/tests/mannwhitney/Default2.aspx>

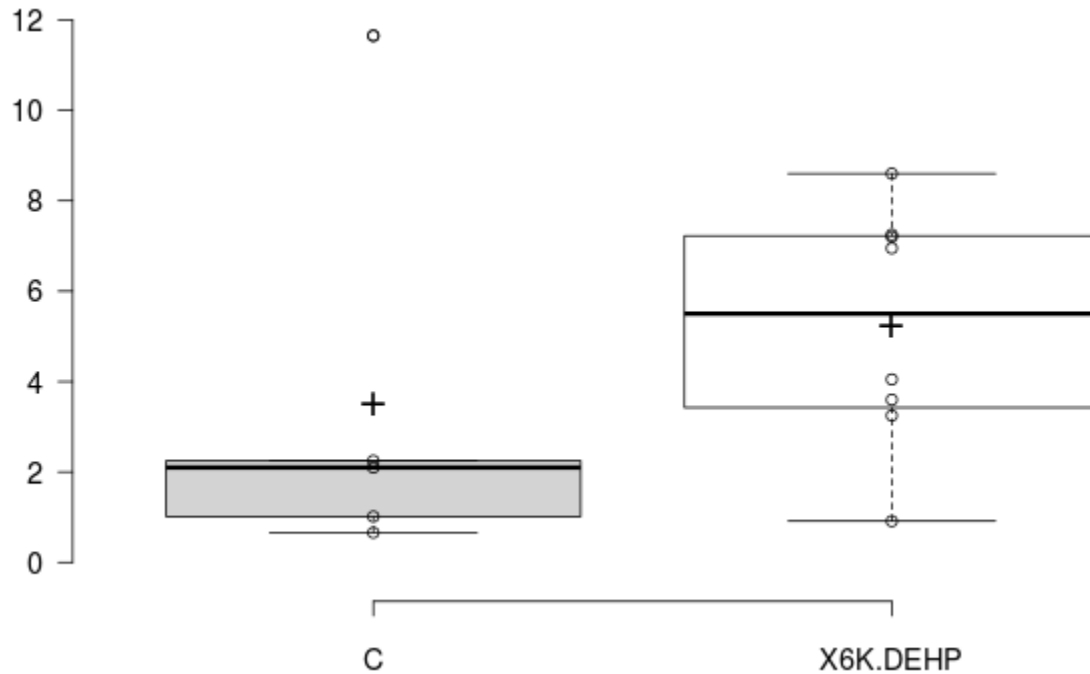
Control vs. 6K

The U-value is 11. The critical value of U at $p < .05$ is 6. Therefore, the result is *not* significant at $p < .05$.

The Z-Score is 1.24427. The p -value is .21498. The result is *not* significant at $p < .05$.
Note: The approximation to the form of the normal distribution becomes less robust at sample sizes smaller than 10, so caution is appropriate here in making use the Z-value calculation.

From <<http://www.socscistatistics.com/tests/mannwhitney/Default2.aspx>>

No Significance is therefore observed with 6K DEHP treatment group for miR-182 measurements in the serum. Boxplot of data is below using the webtool <http://shiny.chemgrid.org/boxplotr/>



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Box plot statistics

	C	X6K.DEHP
Upper whisker	2.25	8.60
3rd quartile	2.25	7.22
Median	2.10	5.50
1st quartile	1.01	3.42
Lower whisker	0.66	0.92
Nr. of data points	5.00	8.00
Mean	3.54	5.23

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<<180320_mir378a_DEHP_28day_serum.pdf>>

Raw data for miR-378a is below and used to calculate normality

C	DEHP
12.8	4.95
333.5	134.65
11.8	56.4
9.85	16.25
62.4	199
	180
	180.5
	71

Control group:

Results:

n = 5
Mean = 86.07000000000001
SD = 140.06774075425076
W = 0.6584026622326289
Threshold for (p=0.01) = 0.6859999895095825 --> HO rejected
Threshold (p=0.05) = 0.7620000243186951 --> HO rejected
Threshold (p=0.10) = 0.8059999942779541 --> HO rejected
--> Your data is not normally distributed p<0.01

From <<http://sdittami.altervista.org/shapirotest/ShapiroTest.html>>

6K group:

Results:

n = 8
Mean = 105.34375
SD = 77.86797635145932
W = 0.8915006114706987
Threshold (p=0.01) = 0.7490000128746033 --> HO accepted
Threshold (p=0.05) = 0.8180000185966492 --> HO accepted
Threshold (p=0.10) = 0.8510000109672546 --> HO accepted
--> Your data seems normal

From <<http://sdittami.altervista.org/shapirotest/ShapiroTest.html>>

Results for miR-378a: They indicate the 6K treatment group is not normally distributed. Therefore a non-parametric test must be used to examine the data.

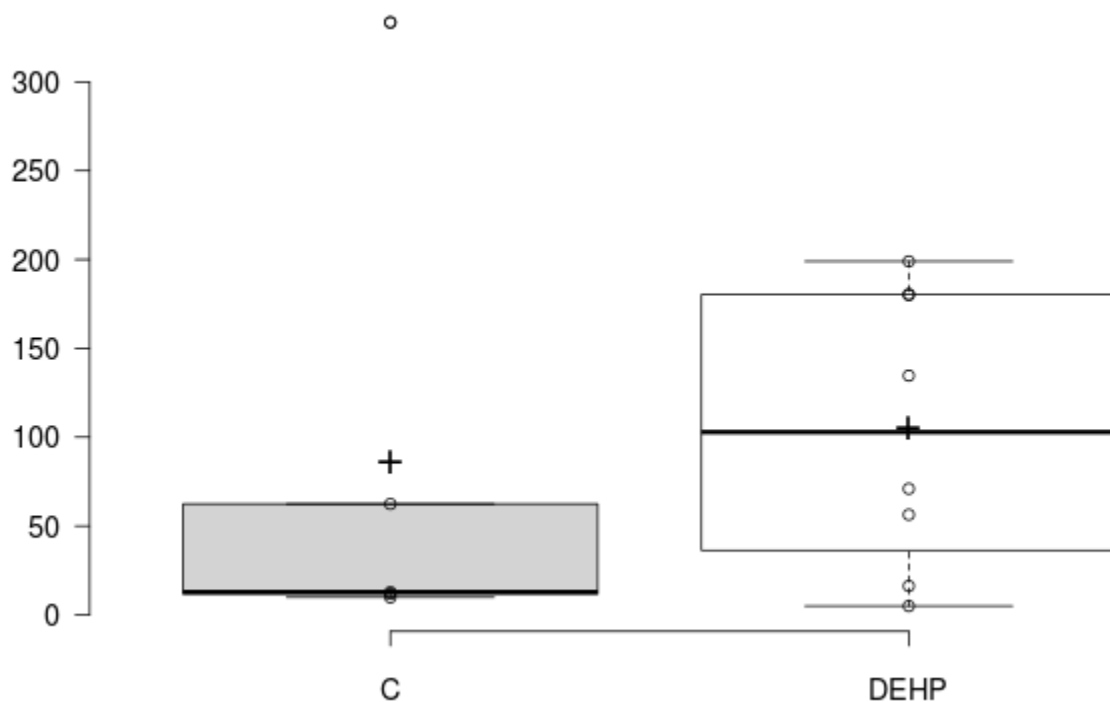
A Mann-Whitney U Test was used to compare control to the treatment groups for miR-378a. The webtool is located here: <http://www.socscistatistics.com/tests/mannwhitney/Default2.aspx>

Control vs. 6K

The *U*-value is 14. The critical value of *U* at $p < .05$ is 6. Therefore, the result is *not* significant at $p < .05$.

The *Z*-Score is 0.80512. The *p*-value is .41794. The result is *not* significant at $p < .05$.
Note: The approximation to the form of the normal distribution becomes less robust at sample sizes smaller than 10, so caution is appropriate here in making use the Z-value calculation.

From <<http://www.socscistatistics.com/tests/mannwhitney/Default2.aspx>>



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Box plot statistics

	C	DEHP
Upper whisker	62.40	199.00
3rd quartile	62.40	180.25
Median	12.80	102.83
1st quartile	11.80	36.33
Lower whisker	9.85	4.95
Nr. of data points	5.00	8.00
Mean	86.07	105.34

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