THE EFFECT OF SALT TYPE ON OSMOTIC PRESSURE

Full Name: Melike Savas
Department: Mathematics with Applied Statistics

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1.ABSTRACT

Salt starts osmosis by attracting water and enabling it to pass across the membrane toward it. A solute is a material that dissolves in water. When water is applied to a solute, it diffuses, disperses the salt concentration, and forms a solution.

"Water tends to move across a membrane from a solution of low solute concentration to one of high. Or, in other words, since solutions with a high amount of dissolved solute have a lower concentration of water, water will move from a solution of high water concentration to one of lower. This process is known as *osmotic flow."(Lodish, H and others,2000)*

My project suggests that there could be an effect of salt type on osmotic pressure. In this project, my research question is "How do different types of salt affect the osmotic pressure?". I am going to use a repeated Latin square design. I am going to decide what type of lettuce and onions by using simple random sampling. I will replicate it three times. I am going to choose three days for replication by using simple random sampling. For each day I am going to randomize which observations will run first by using simple random sampling.

My response variable in this experiment is the grams of water, treatments are Himalayan pink salt, table salt, and sea salt. I am planning to use two blocks that are lettuce and onion. I am going to replicate it 3 times. Thus, I need to run a total of 27 observations.

I am going to use the ANOVA- glm test. I need to do a comparison to figure out which one is different. I need to use Tukeys or Fisher Tests. When I am applying for these tests I am going to use R and Minitab programs. As a result of this experiment, I am expecting to see that one of the Salt types causing less water to move.

Keywords: ANOVA, Himalayan salt, Kosher salt,Latin-square design, Lettuce, Membrane,Onion, Osmotic Pressure,Salt , Sea salt,Water,

2.INTRODUCTION

Salt, also known as sodium chloride, is made up of about 40% sodium and 60% chloride. A small quantity of sodium is necessary by the human body to conduct nerve impulses, contract and relax muscles, and maintain proper water and mineral balance. For these crucial roles, we need approximately 500 mg of sodium each day.

Osmosis is the diffusion of water across a membrane in response to osmotic pressure caused by an imbalance of molecules on either side of the membrane. Water flows out of cells and into the blood to dilute sodium levels in the blood. Nonetheless, consuming too much sodium can cause high blood pressure, heart disease, and kidney disease. Seizures, coma, and even death may result from the fluid transition and build-up in the brain.

The average American consumes at least 1.5 teaspoons of salt daily, or around 3400 mg of sodium, even more than our bodies need. We can not stop using salt in our meals. But, if we can find out what type of salt has less osmotic pressure, we can find a way to live healthier.

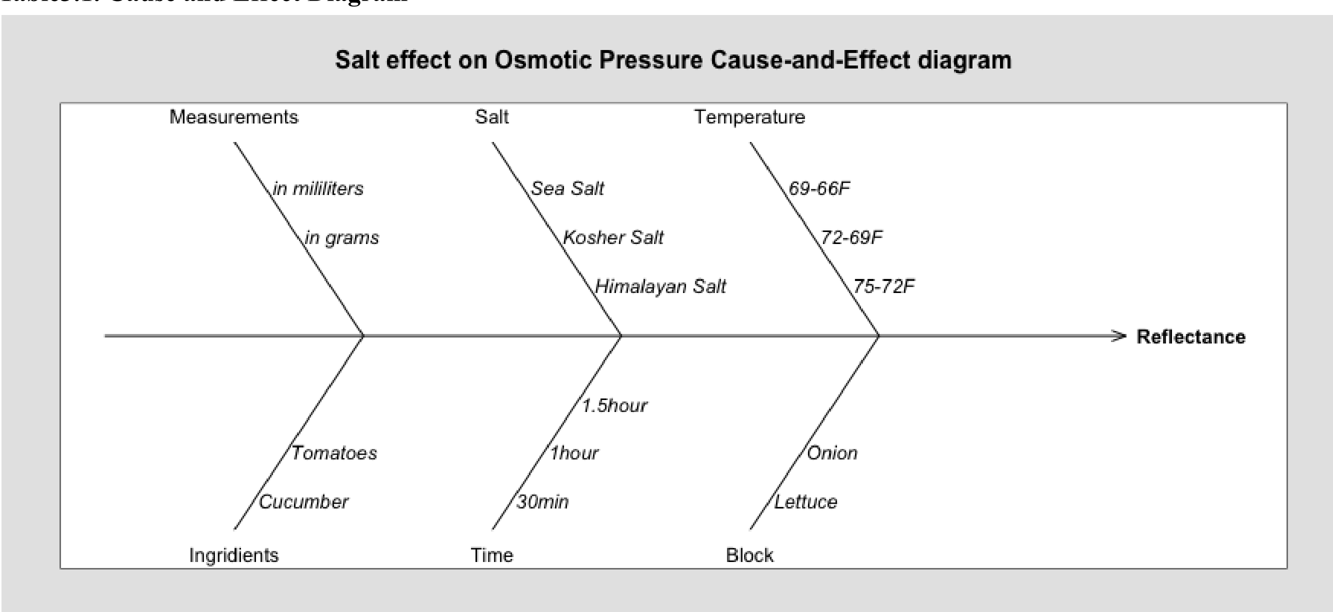
3.EXPLORATORY DATA ANALYSIS

In my experiment, my response variable is grams of water. My blocking variables are “lettuce” and “onion”. My treatment is “ types of salt”. I do not have covariates. My measurements are going to be in grams.

I am using the “Digital Milligram Pocket Scale 50 x 0.001g, Mini Jewelry Gold Lab Carat Powder Weigh Scales with Calibration Weights Tweezers, Weighing Pans, LCD Display” scale for my measurements.

For the blocking variables, I am going to use 3 blocks for “lettuce” and 3 blocks for “onion”. I have 6 types of onions to choose three from them; those are shallots, yellow onions, sweet onions, red onions, white onions, scallions. I also have six types of lettuce to choose three of them; that are Green Leaf lettuce, Red Leaf lettuce, Coral Lettuce, Romaine lettuce, Iceberg lettuce, and Boston lettuce. My treatment is a type of salt. I will pick three types of them from the six kinds of salt that are Himalayan salt, Sea Salt, Celtic salt, Kosher Salt, Refined salt, Black Hawaiian Salt.

Table3.1. Cause and Effect Diagram



My design of the experiment is using the fishbone diagram above(Table.1). I will fix everything except blocking variables and treatment. I will use 100gr of lettuce, 30gr of onion, and 8gr of salt for each observation.

First of all, I searched for types of lettuces, onions, and salts online. Then I decided what types of lettuces, onions, and salts could be used in my experiment. Rolling dice is useful for each part of my randomization (choosing types of lettuce, onion, and salt) since I have six options for each. I gave numbers 1 through 6 to each type of lettuce. I rolled a fair dice until I got three different numbers. Then I decided to use types of lettuces that have those numbers. I applied this process for choosing onions, and salts as well. I ended up with 'Romaine lettuce, Iceberg lettuce, and Red Leaf lettuce' for lettuce types, 'Red onion, Yellow onion, and White onion' for onion types, and 'Himalayan salt, Sea salt, Kosher Salt' for salt types.

After deciding what types of lettuces, onions, and salts I am going to use, I decided how to layout them in a Latin square design table. Treatments are assigned at random within rows and columns, with each treatment once per row and once per column. There are three numbers of rows, columns, and treatments. (Table.2)

Table3.2.Latin Square Design

	LETTUCE Romaine Lettuce	Iceberg Lettuce	Red Leaf Lettuce
ONIONS Yellow Onion	Kosher Salt	Sea Salt	Himalayan Salt
White Onion	Himalayan Salt	Kosher Salt	Sea Salt
Red Onion	Sea Salt	Himalayan Salt	Kosher Salt

I used R for sample size simulation. I have 3 rows and columns, and 3 treatment levels. My sample size will be 9 for each replication. I am using repeated Latin Square design. I am going to have 9 samples for each replication. My total sample size is going to be 27 since I have 3 replications.

For my replications I need three different days, I gave numbers 1 through 7 to days, and then I used a random number generator to decide on what days I am going to run my observations. I need three replications, so I chose three different days. I am going to run my observations on " Monday", "Friday", and "Sunday". For each day I randomly choose which observation will be done first. The order of each observation is written at the bottom of the table for each day on table3.3

Table.3.3.Order randomization for days

on Monday	on Friday	on Sunday
RL IL LL	RL IL LL	RL IL LL

<table>
<tr>
<td><table><tr><td>YO</td><td>1.K</td><td>2.S</td><td>3.H</td></tr><tr><td>WO</td><td>4.H</td><td>5.K</td><td>6.S</td></tr><tr><td>RO</td><td>7.S</td><td>8.H</td><td>9.K</td></tr></table></td>
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<td>[1] 1 3 9 6 5 7 2 8 4
Levels: 1 2 3 4 5 6 7 8 9</td>
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Levels: 1 2 3 4 5 6 7 8 9</td>
<td>[1] 1 2 8 6 9 7 3 4 5
Levels: 1 2 3 4 5 6 7 8 9</td>
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</table>

4.MODEL AND ASSUMPTIONS

In my experiment my hypothesis is;

Ho: $\tau_1 = \tau_2 = \tau_3$

Ha: at least one of them is different.

I am going to use ANOVA-glm for testing.

I run 3 pilot observations to have information about stDev and max. difference in means. I obtained 12.750, 19.063, 22.024 grams of water. My response values will be always very small and I only run a few pilot observations. Considering this situation and according to these pilot results I decided to take variation between 8 and 24, stdev as 3.14, and max. dif as 16. Based on these assumptions I run the R code for simulation for repeated Latin square design and it gave me the power of 86%.

My significance level is 0.05. I have 27 observations and the power is 86% for this experiment based on the total sample size.

I am comparing the means as a contrast, thus I may see the means of my treatments are different. Such as, my treatments are A, B, C

$\Gamma 1 = 2\mu_A - \mu_B - \mu_C = 0, \qquad 2\mu_A = \mu_B + \mu_C$

$\Gamma 2 = \mu_B - \mu_C = 0, \qquad \mu_B = \mu_C$

I can use Tukey or Fisher test to see if there is a difference between the effect of my blocking factors and treatments. I can use the Main effect plot to see the primary effect on osmotic pressure. The interaction plot can give me an idea about what combinations would give more grams of water.

I am using R programming for cause-and-effect diagrams, randomization layout, and simulation for power calculations. I am also using Minitab for my calculations.

I am not expecting to have missing values. I run all replications, I have the results. If I have outliers, I can try log transformation. If log transformations don't make a difference, I would go on with my original results.

After I collected my data for the first repetition, I checked the descriptive statistics.

Table4.1.Descriptive statistic of 1.repetition

Statistics

Variable	Salt	N	N*	Mean	SE Mean	StDev	Minimum	Q1	Median	Q3	Maximum
Water	Himalayan	3	0	12.848	0.524	0.908	11.827	11.827	13.155	13.563	13.563
	Kosher	3	0	16.074	0.467	0.810	15.172	15.172	16.312	16.738	16.738
	Sea	3	0	16.282	0.688	1.192	14.909	14.909	16.894	17.044	17.044

Depending on the table 4.1 I am expecting to see the osmotic pressure of Himalayan salt is different than other ones. I believe after I complete my experiment, my results are going to give me superior results to now.

5.INFERENCE

I ran the ANOVA-glm test. Based on analysis of variance p-value for the treatment is 0.004 < 0.05 significant.(Table5.1) I have enough evidence to reject the null hypothesis. At least one of them is different. Thus, Salt types have an effect on osmotic pressure.

Onion and lettuce are blocking factors. I do not need to check p-values for blocking factors.

Table5.1.ANOVA-glm

Factor Information

Factor	Type	Levels	Values
Salt	Fixed	3	Himalayan, Kosher, Sea
Onion	Fixed	3	R, W, Y
Lettuce	Fixed	3	Iceberg, Red Leaf, Romaine
Rep	Fixed	3	1, 2, 3

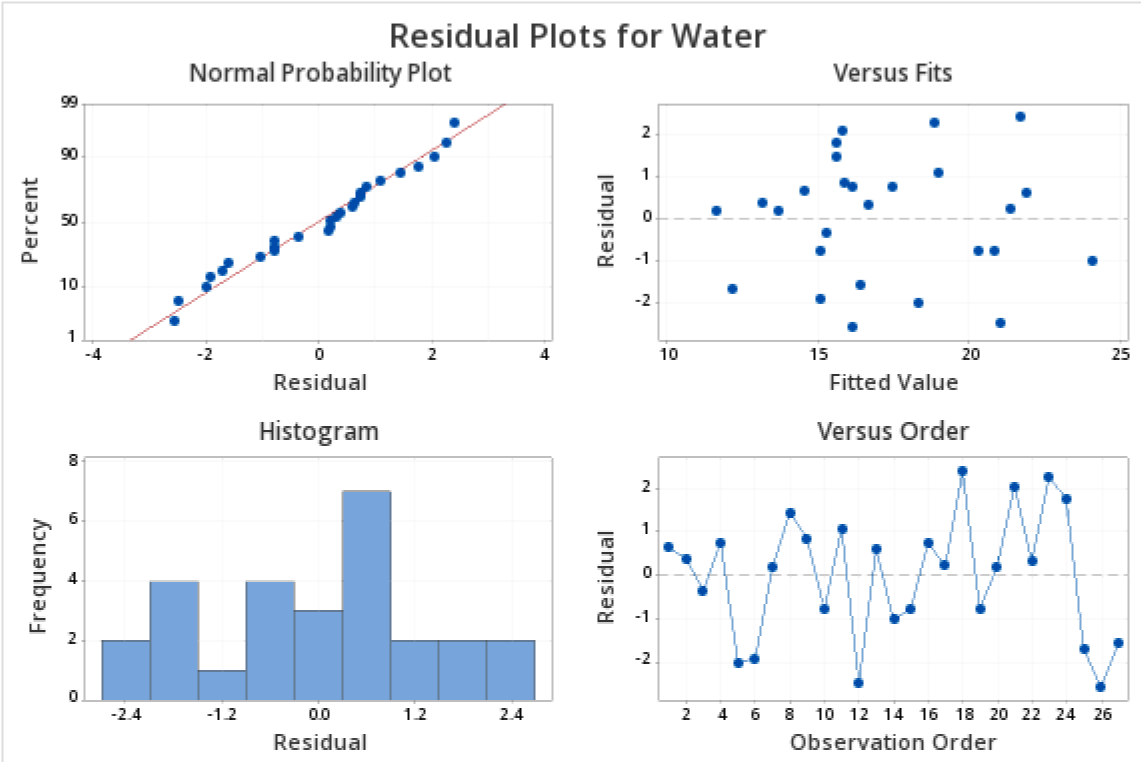
Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Salt	2	43.92	21.959	7.43	0.004
Onion	2	12.97	6.483	2.19	0.140
Lettuce	2	27.85	13.926	4.71	0.023
Rep	2	185.02	92.508	31.30	0.000
Error	18	53.21	2.956		
Total	26	322.96			

I am going to check residuals to see if the model is good to use. (Table 5.2)

As looking at the residuals there is no big concern. Distribution looks normal.Histogram doesn’t look exactly symmetric, but there is no violation for normality. There is no pattern on Residuals vs fitted value plot, that is good. As looking at observation order I can say observations are independent. Thus, I can go with the model that I obtained.

Table5.2.Residuals



I ran the Tukey test to see which treatment caused the max difference. I can also use the Fisher lsd test .

Depending on the Table5.3 I can say that Himalayan salt has the least osmotic pressure. The difference between Sea salt and Kosher salt is very close to 0 and they are in the same group. I can say that Himalayan salt causes the max difference in means.

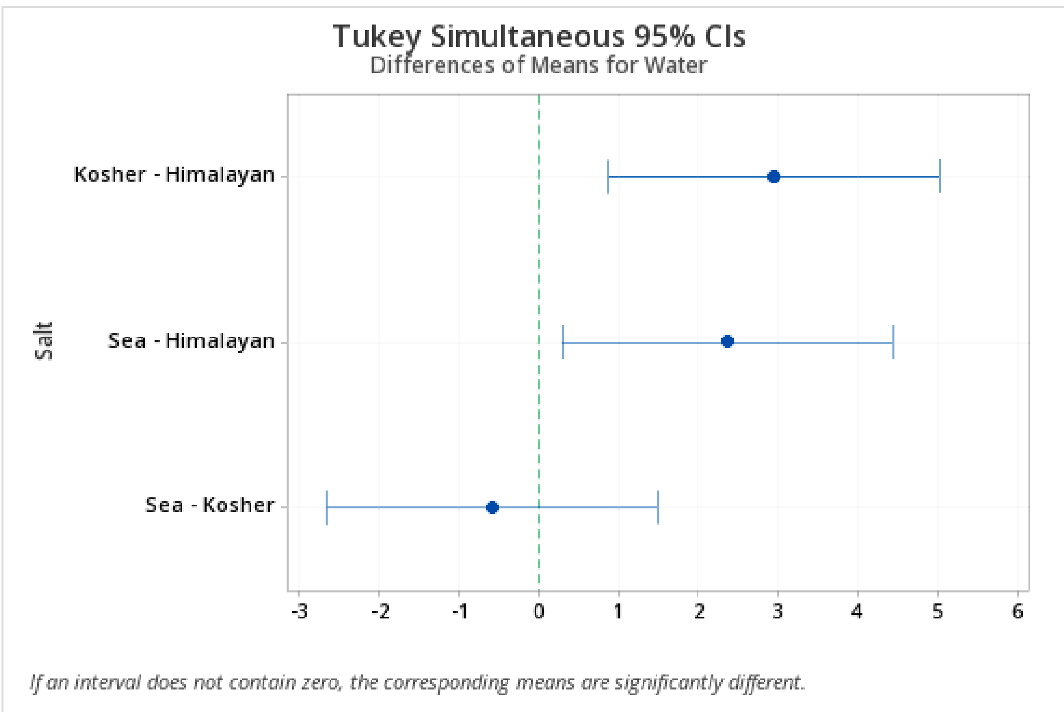
Table5.3.Tukey PAirwise Comparisons

Tukey Pairwise Comparisons: Salt

Grouping Information Using the Tukey Method and 95% Confidence

Salt	N	Mean	Grouping	
Kosher	9	18.3556	A	
Sea	9	17.7766	A	
Himalayan	9	15.4074		B

Means that do not share a letter are significantly different.



6.CONCLUSIONS

I have enough evidence to say that at least one of the treatment effect is different. Thus, Salt types have an effect on osmotic pressure. I can say that Himalayan salt's osmotic pressure is different from the other two salts. I chose the salt types from all kinds of salts randomly. So, I can say that at least one of the salt types has different osmotic pressure.

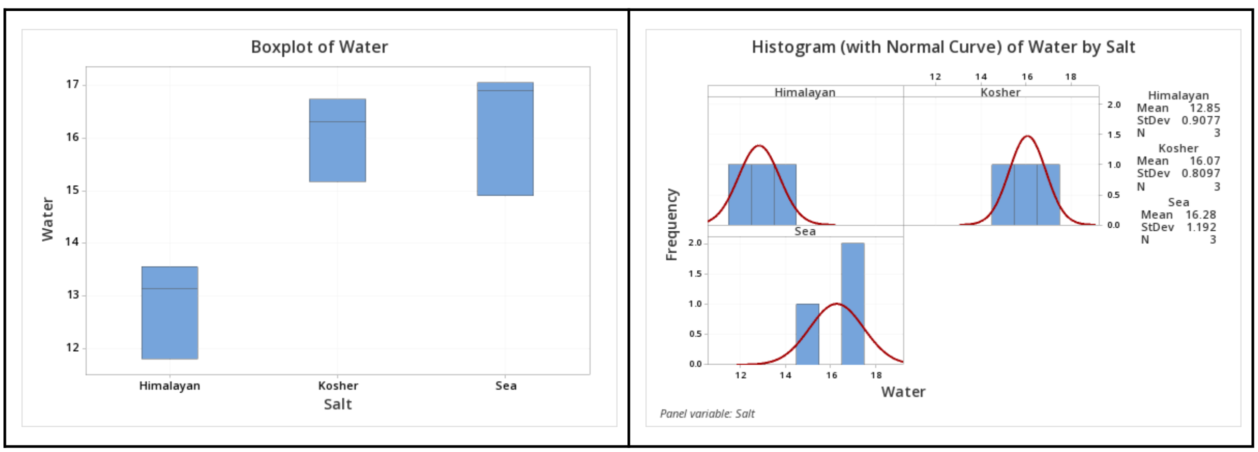
For my further study I can research which salt type could be the best preserver for food conservation. I may have another experiment with more treatment levels to see what kinds of salt has less osmotic pressure.

7.APPENDIX

R-code (Fishbone diagram)

```
cause.and.effect(cause=list(
        Measurements=c('in milliliters','in grams'),
            Salt=c('Sea Salt', 'Kosher Salt', 'Himalayan Salt'),
        Temperature=c('69-66F', '72-69F','75-72F'),
        Ingredients=c('Cucumber', 'Tomatoes'),
        Time=c('30min','1hour','1.5hour'),
        Block=c('Lettuce', 'Onion')),
        effect='Reflectance', title = "Salt effect on Osmotic Pressure Cause-and-Effect diagram",
        cex = c(0.8, 0.8, 0.8))
```

Box-plot and histograms after 1.repetition



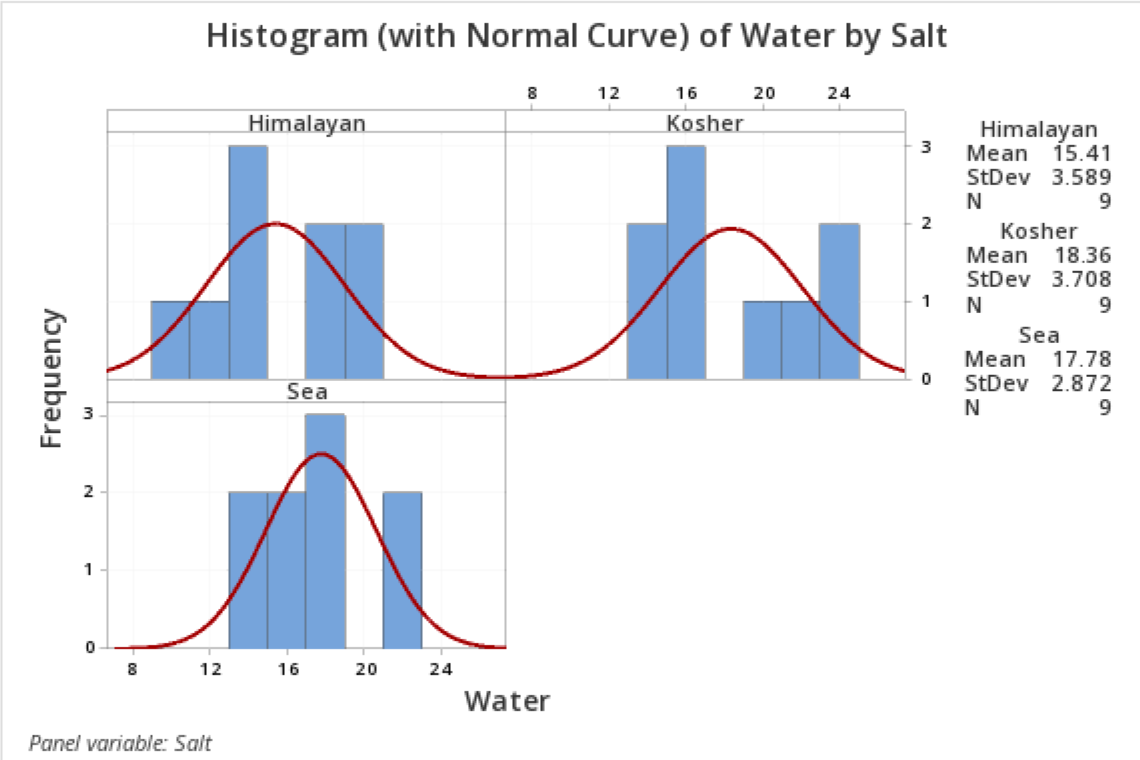
R-code (simulation for replicated Latin square design-power calculation)

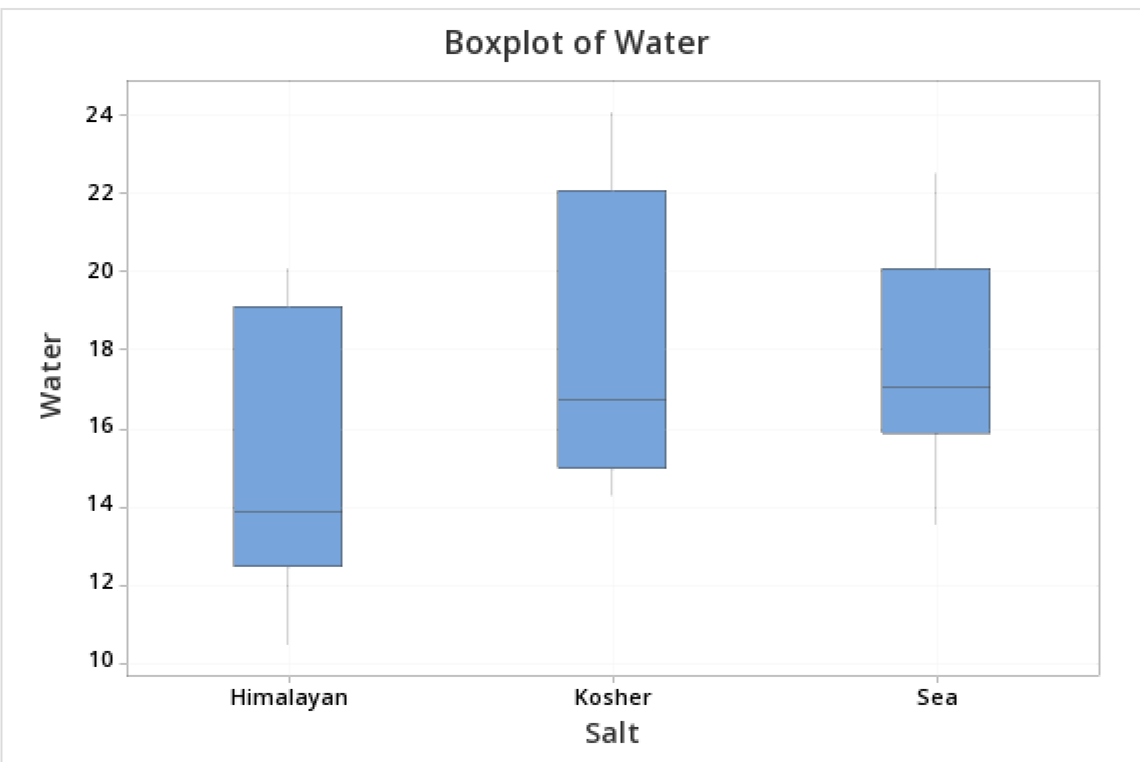
```
mu <- c(8,19,24)
n.sim  <- 1000
sigma2 <-3.14^2
rep=3
g    <- length(mu)
r=g
```

```
c=g
install.packages('agricolae')
library(agricolae)
trt<-LETTERS[1:g]
outdesign <-design.lsd(trt,serie=2,seed=23)
outdesign<-data.frame(outdesign$book)
for (i in 2:rep){
    v <-design.lsd(trt,serie=2,seed=23*i)
    outdesign<-rbind(outdesign, v$book)
}
lsd=outdesign
lsd$trtn=as.numeric(lsd$trt)
lsd$rep=rep(1:rep,each=r*c)
lsd
results <- numeric(n.sim) ## vector to store results in
aux=0
for(i in 1:n.sim){
  for(i in 1:g^2){
  lsd$y[i] <- rnorm(1, mean = mu[lsd$trtn[i]], sd = sqrt(sigma2))
  }
  data <- data.frame(lsd)
  fit  <- aov(y ~ trt+row+col+rep,data = data)
  aux1 = ifelse(summary(fit)[[1]][1, "Pr(>F)"] < 0.05,1,0)
  aux=aux+aux1
}
power=aux/n.sim
power
```

My Data (Number of data/Lettuce type/Onion type/Salt type/Water in grams/ Randomized order in the day/ Replication number)

#	Lettuce	Onion	Salt	Water	Randomized order	Replication
1	Romaine	Y	Kosher	15.172	1	1
2	Romaine	W	Himalayan	13.563	9	1
3	Romaine	R	Sea	14.909	6	1
4	Iceberg	Y	Sea	16.894	7	1
5	Iceberg	W	Kosher	16.312	5	1
6	Iceberg	R	Himalayan	13.155	8	1
7	Red Leaf	Y	Himalayan	11.827	2	1
8	Red Leaf	W	Sea	17.044	4	1
9	Red Leaf	R	Kosher	16.738	3	1
10	Romaine	Y	Kosher	19.551	4	2
11	Romaine	W	Himalayan	20.064	1	2
12	Romaine	R	Sea	18.58	7	2
13	Iceberg	Y	Sea	22.531	6	2
14	Iceberg	W	Kosher	23.082	8	2
15	Iceberg	R	Himalayan	20.101	2	2
16	Red Leaf	Y	Himalayan	18.183	5	2
17	Red Leaf	W	Sea	21.629	9	2
18	Red Leaf	R	Kosher	24.1	3	2
19	Romaine	Y	Kosher	14.295	1	3
20	Romaine	W	Himalayan	13.905	8	3
21	Romaine	R	Sea	17.848	6	3
22	Iceberg	Y	Sea	16.986	2	3
23	Iceberg	W	Kosher	21.101	9	3
24	Iceberg	R	Himalayan	17.38	3	3
25	Red Leaf	Y	Himalayan	10.489	7	3
26	Red Leaf	W	Sea	13.568	4	3
27	Red Leaf	R	Kosher	14.849	5	3


Histogram (with Normal Curve) of Water by Salt
Himalayan
Kosher
Sea
Frequency
Water
Himalayan
Mean 15.41
StDev 3.589
N 9
Kosher
Mean 18.36
StDev 3.708
N 9
Sea
Mean 17.78
StDev 2.872
N 9
Panel variable: Salt


Boxplot of Water
Water
Himalayan
Kosher
Sea
Salt

Fisher Pairwise Comparisons: Salt

Grouping Information Using Fisher LSD Method and 95% Confidence

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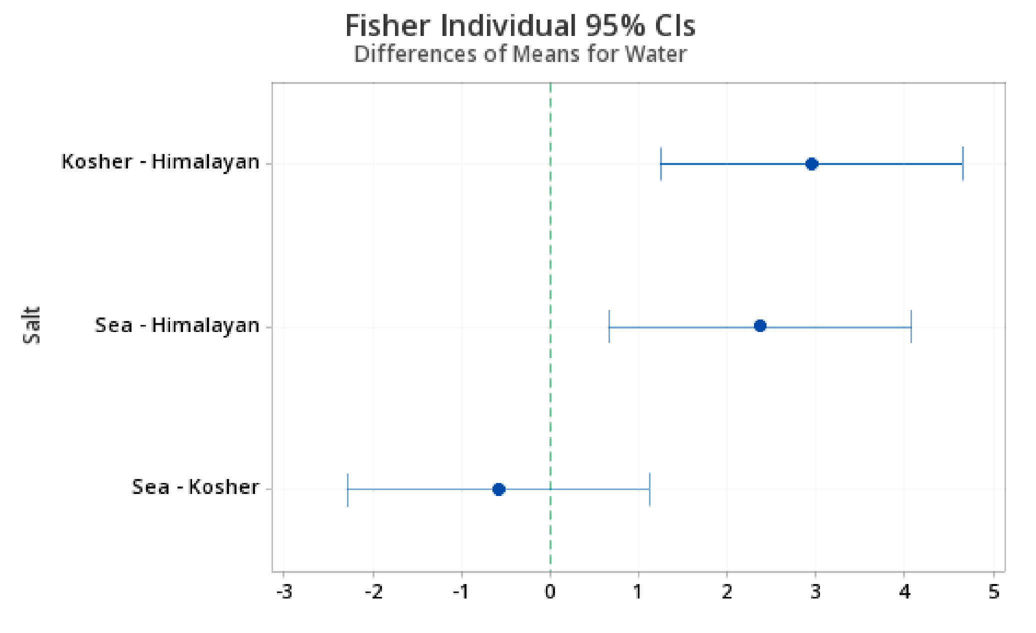

If an interval does not contain zero, the corresponding means are significantly different.

8. REFERENCES

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