

A Comparison Study of the Effects of Music, Cannabis and Alcohol on Endorphin Levels of Individuals Aged between 21 and 25

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I. Abstract:

Endorphin is a chemical secreted by the central nervous system to relieve stress or pain. Often, when they are naturally released, Endorphins serve as painkillers and happiness boosters. Therefore, we dug into what substances may potentially be able to assist the Central Nervous System to produce this chemical. Our aim was to determine whether Cannabis Tea, Alcohol, or Music were effective stimulants for the levels of Endorphin. Therefore, we created a two factorial randomized block design to help us create an analysis of the effects of the aforementioned substances on the island population. As seen below, our outcomes drawn from the ANOVA tables and Interaction plots can help us determine whether the substances are proven as valid stimuli. Prior research conducted by *Impulse: The Premier Journal for Undergraduate Publications in the Neurosciences* has suggested that opioidergic activity plays a role in addictions by mediating the development of reinforcing qualities of certain activities and substances (Leuenberger, A., 2016).

II. Introduction:

People have long been obsessed with the feeling of happiness what factors could cause it. *According to a study by Harvard*, three out of four students have reported having experienced at least one stressful life event in the last year. More than 20 percent of students reported experiencing six or more stressful life events in the last year. Stress exposure was strongly associated with mental health diagnoses, self-harm and suicidality (Younghans, J., 2018). As a way to relieve stress, many college students have enjoyed the occasional recreational drugs and alcohol with good music. Roughly 80 percent of college students – four out of every five – consume alcohol to some degree (Galbicsek, C., 2019) . This study aims to determine what can make people the happiest— when they are listening to music or under the influence of drugs and alcohol and whether there are interactions between these three factors that make music more enjoyable when they consumed recreational drugs and alcohol. To measure the level of happiness, we determine to measure subjects' blood endorphin level.

Studies have shown that endorphins interact with opiate receptors in the brain to reduce our perception of pain and act similarly to drugs such as morphine and codeine (Alikhani, A. H. et al., 2015). Endorphin is a hormone releasing peptide that is released by the central nervous system and the pituitary gland. A

primary function of endorphins is to inhibit communication of pain signals, which in turn, allows an individual to attain a feeling of euphoria. This is similar to an effect produced by numerous other opioids. Endorphins are primarily known to be naturally released in response to injury or pain. The release first begins in the Peripheral Nervous System, in which a special type of endorphine (β -endorphin), is released by the pituitary gland and is eventually binded with μ -receptors. This binding, in turn, causes the pain signal of the peripheral nerves to be to be inhibited. On the other hand, the Central Nervous System also acts similar to the Peripheral Nervous System, but only differentiate in neurotransmitter type. Nevertheless, when endorphin binds to the μ -receptor, a neurotransmitter named Gamma-aminobutyric acid(GABA) is inhibited. Without GABA, dopamine, a chemical released by neurons is finally released. Similarly, dopamine is also known for being a neurotransmitter that is associated with pleasure.

Studies have shown that music releases endorphins in blood and could change people's mood (Rokade, P. B., 2011). There are not many studies done on the direct relationship between cannabis and endorphin levels, but increased level of dopamine, which is a similar hormone responsible for the feeling of happiness, in animals after being treated with cannabis has been observed (Oleson, E. B., & Cheer, J. F., 2012). Scientists have also shown that alcohol consumption could induce the release of endorphins (Mitchell, J. M. et al., 2012). Therefore, previous literature has established the basis for the relationship between music, cannabis and alcohol, the three factors that we are interested in, with the release of hormones that could create feelings of pleasure. Our study wants to further compare the endorphin level released under each factor and determine if there are any interaction effects between these factors. The targeted subjects of this study are college students at legal age, so we are only going to collect from individuals aged 21 to 25. In addition, studies have also shown that laughing exercises, meditation, music, exercise, workouts all these exercises of keeping the mind calm, feeling confident, happy are related with endorphin release (Rokade, P. B., 2011). We decided to delve deeper into the connection between endorphin release and listening to music.

III. Methods

2.1 Participants:

Our participants will be individuals from the open-ended virtual environment named *Island*. To take into account randomization, we will select 108 islanders between the ages of 18 and 25 and use the sample

function in R to randomly select them and then randomly assign treatment to the selected islanders who consented to be in the study. Our seed in this experiment will be set for 2 and 3. Our goal is to reach a sample that will represent the entire population.

2.2 Design:

We will be using a two by three factorial randomized block design, in which the parameters are assigned below:

	No Music	Music
Control	No Music No Substance	Music No Substance
Cannabis Tea	No Music Cannabis Tea	Music Cannabis Tea
Alcohol	No Music Alcohol	Music Alcohol

2.3 Instruments:

Endorphin levels will be measured from the islanders through the procedure of a blood test. Alcohol and cannabis tea will both be administered through an oral liquid. In deciding between cannabis tea and a reefer, cannabis tea seemed the better choice so we can compare liquids on both ends instead of any inhalation techniques. Additionally, because alcohol takes approximately thirty minutes to metabolize in our system, we will measure the endorphin levels of the islander a half an hour after the administration.

For cannabis tea, we had to wait for two hours to take the second reading after its administration. Due to Endorphin being in the form of a hormone, the primary method of release is through the bloodstream.

2.4 Procedures:

Step 1: Subject Selection

Since it would be difficult to randomly select subjects from the whole islands, we would try to randomly select subjects that fit the age between 21 to 25 from different villages. We aim to select 108 subjects total, 54 males and 54 females.

Step 2: Random Assignment

We will divide the subjects into two blocks of 54 based on the gender. For each block, we will use R to randomly select sample sizes of 9 without replacement for each of the 6 treatment groups.

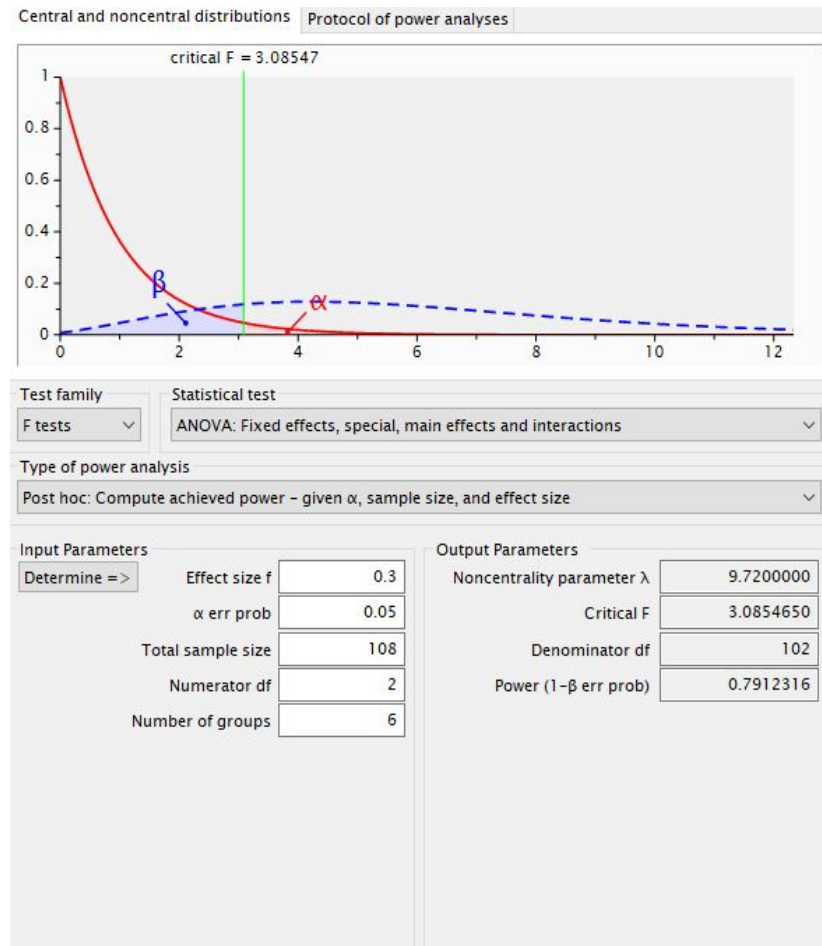
Step 3: Measurement

We would conduct a blood test for all the subjects and measure their endorphin level. Every subject would then receive the treatment of their assigned group. Blood test endorphin level would be measured again to calculate the difference in endorphin level before and after the treatment. For music treatment group, we conducted a survey to determine their favorite music genre.

IV Data Analysis

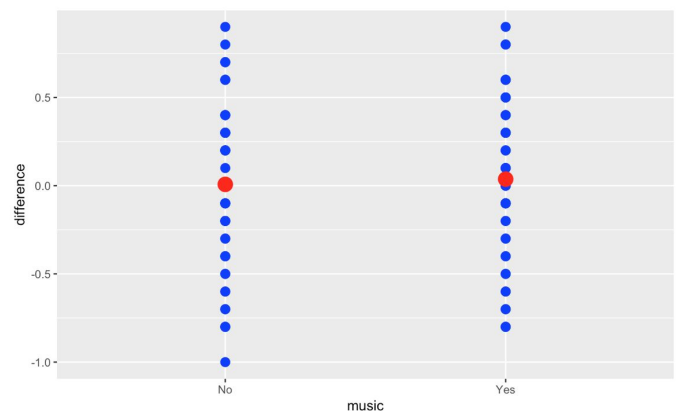
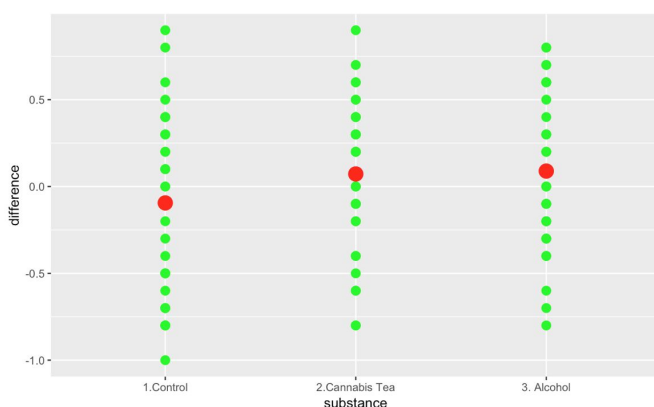
- Using R, we will conduct an ANOVA analysis on the 2 x 3 factorial experiment design. The ANOVA will conduct a F-test within treatments and blocks to determine if there is a significant difference between endorphin levels of our groups and a potential interaction between the two treatment groups. To set a benchmark statistic, we will add a control measure in both of the treatment groups. This will help us understand the effects our treatments better. Furthermore, we are placing a block on the gender of the subjects to rule out any nuisance caused by that factor. We will use the results from the ANOVA analysis to determine whether the block is significant.

- **Sample Size Determination:** To determine the sample size, we decided to use a power of 0.8 and a slightly conservative effect size of 0.3. Using G-Power, we received a sample size of 111. We decided to reduce the sample size to 108 in order to distribute the subjects evenly across the 6 groups. That yielded a power of 0.79132, which is very close to our desired power of 0.8, as seen below.



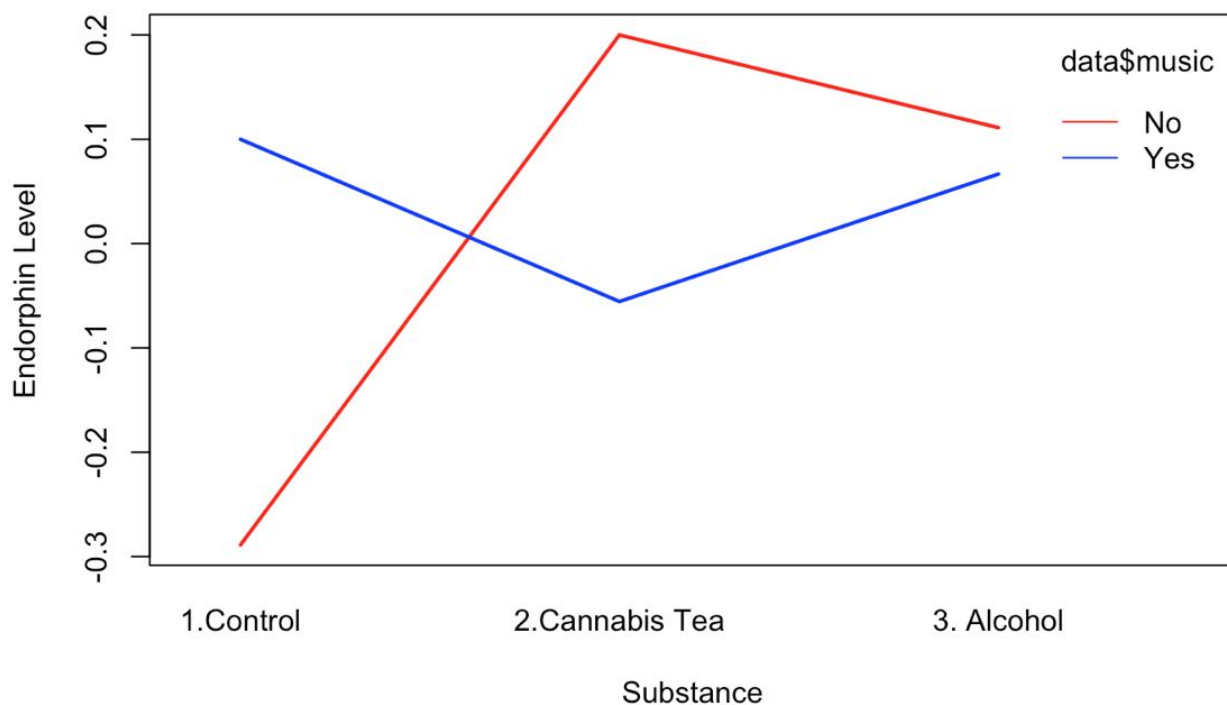
V. Results

5.1. Differences in Means



We first checked the differences in means. The variation between groups did not seem to be significant and there were huge variation within groups. It seemed that the two factors: substance and music were not significant in causing subject's Endorphin level to increase.

5.2 Interaction Plot



From the above interaction plot, we suspect that there is a significant interaction between our 2 factors, that is, music and substance. The Endorphin Levels on the y-scale seemed to be small because we only measured the differences as a response variable and all the differences were ranged between -0.3 and 0.2 pg/ml. Due to the differences in slope, we suspect that in the presence of cannabis tea and alcohol, subjects received no music had higher Endorphin Level in blood than subjects who received music treatments. This seemed to be contradicting to what we initially hypothesized, but further conclusions should be confirmed by the ANOVA table.

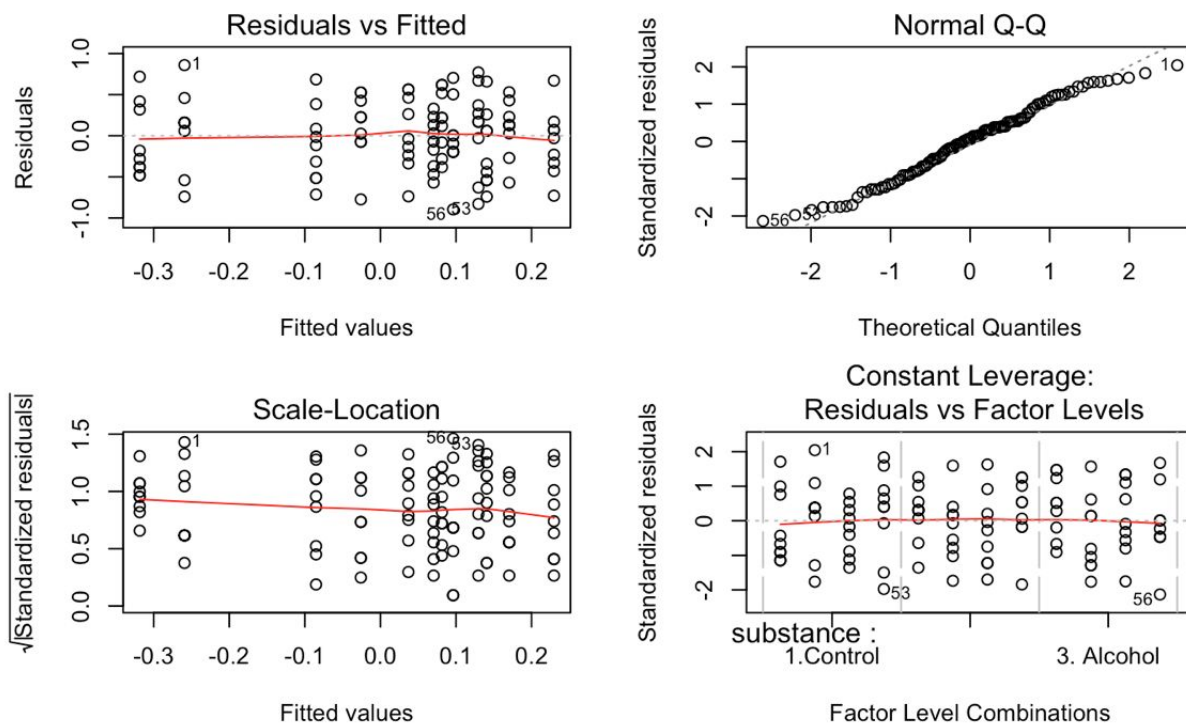
5.3 Anova Analysis

	Df	Sum Sq	Mean Sq	F-value	p-value
Substance	2	0.7400	0.37000	1.9601	0.14616
Music	1	0.0237	0.02370	0.1256	0.72381
Gender	1	0.0948	0.09481	0.5023	0.48013
Substance * Music	2	1.9430	0.97148	5.1465	0.00744
Residuals	101	19.0652	0.18876		

The Null hypothesis for an F test states that all slopes must be equal to 0 while the alternative hypothesis states that at least one slope is not equal to zero. If we observe the ANOVA table above, we can see that the factor ‘substance’ has an F-statistic of 1.9601. This is quite a small F-statistic. This essentially tells us that the difference in means between the 3 groups (alcohol, cannabis and control) is only 1.9601 times higher than the difference within each group. The probability of observing an F-statistic as extreme or more extreme than 1.9601 is 0.14616. Maintaining a significance level alpha of 5%, we fail to reject the null hypothesis as we do not have enough evidence to conclude that at least one of the slopes is not equal to 0. After performing the same analysis on music and gender, we find that the p-value for these variables (one factor and one block) is much higher than 0.05 and therefore we can conclude that using music (binary variable) as a factor was not a statistically significant predictor of endorphin levels and further conclude that gender was not a significant blocking factor. For a blocking factor to be significant, we would require the variation between blocks to be much greater than the variation within blocks. However, from the above anova table and the graphical representations, we can see that the mean difference in endorphin levels for males is quite similar to the mean difference in endorphin levels for females. Therefore, gender is certainly not a significant predictor or possible nuisance factor when considering the effects of music and substances on the amount of endorphins released. The only statistically significant

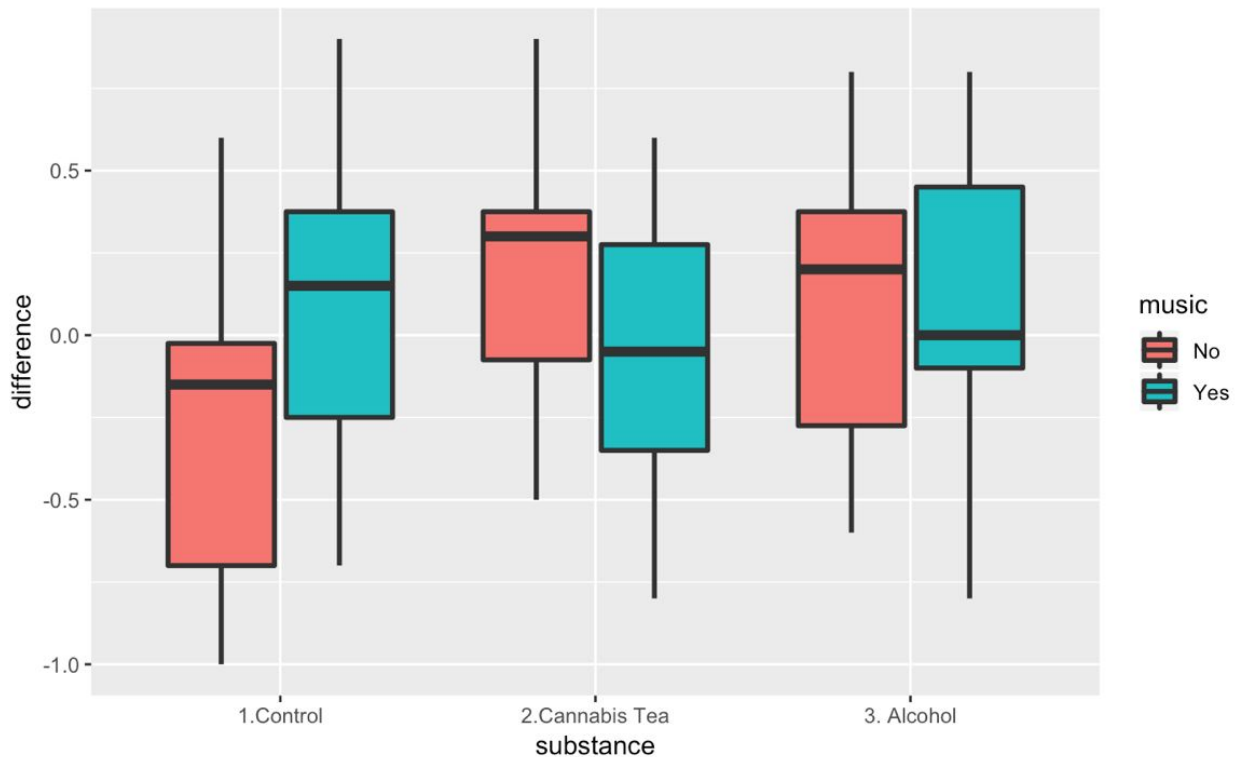
factor in the ANOVA table is the interaction between substance and music. This fact can also be seen if we observe the interaction plot displayed below.

5.3 Model Validity



When creating a linear model, it is essential to make sure that the model is valid. This is done by making sure certain assumptions are satisfied such as the constant variance assumption and the assumption that our errors come from a normal distribution. In order to check whether our linear model was valid, we first created a 'Residuals vs Fitted' plot (displayed above). If we observe the plot carefully, we can see that the residuals (actual values - predicted values) are evenly distributed both above and below the $y=0$ line. There is no trend in the residuals and this fact can be confirmed by observing the red line running through the center of the plot. Since the red line is absolutely straight without any ups and downs, we can safely conclude that the constant variance assumption is indeed satisfied. Next, we observe the QQ plot also displayed above. Almost all the of the points lie on the line. There is a slight right and left tail, indicating that there are indeed a few errors in our model that do not come from a normal distribution. However, the QQ plot does look good as it tells us that most of our errors are normally distributed. To conclude, our model is certainly valid and as a result, we were able to proceed with our analysis.

5.5 Boxplot



After observing the above boxplot, we can see that the difference in medians for each substance is NOT greater than the variation within each substance. We can also see that there is not that much of a difference in the median of each substance with and without exposure to music. This supports our ANOVA table analysis that music and substance are indeed NOT statistically significant due to a small F-statistic which implies that the difference in means between groups is relatively small compared to the variation within each group.

VI. Discussion

After our thorough analysis, we achieved our objective of determining whether the three factors of Cannabis Tea, Alcohol, or Music were effective factors in influencing the levels of Endorphin. Within this analysis, we discovered whether the data rejected or accepted or initial hypotheses, which were:

- 1) Music, cannabis and alcohol will increase our subjects' endorphin levels after being treated.
- 2) The interaction between music and these two substances will increase the endorphin levels even more.

We achieved a power of approximately .8 by sampling 108 islanders, 54 of which were female and 54 of which were male. Once the two factorial block design was conducted, we interpreted a few results key results from the plot function in R, ANOVA table, and interaction plots. Importantly, we noted only one significant factor from the ANOVA analysis, that was the interaction between substance and music.

Therefore, if we considered both the influence of substance and music simultaneously, we see a statistically significant difference in endorphin levels. However, in the scenario of just listening to music or just the use of a substance, there would not be a statistically significant difference in endorphin levels.

Furthermore, offering further evidence to the conclusion, our interaction plot shows a significant interaction between the two factors of music and substance. The information it offers is critical to our analysis because it is telling us that music and substance are dependent on one another. In other words, they are not independent of each other. Additionally, we concluded that the islanders who were exposed to music in the control environment experienced a much higher difference in endorphin levels compared to the islanders who were not exposed to music. On the other hand, our boxplot shows us that the difference in medians for the substances were not greater than the variation within each substance. This directly substantiates the data from the ANOVA analysis, in which music and substance alone were not statistically significant factors due to have a diminutive F-statistic.

Our data analysis was optimal in our scenario because we successfully chose a large enough sample size along with a high power, and conducted numerous tests that provided us great information to justify our results. However, there are always pitfalls and methods that can be adjusted in order to better perform a controlled experiment. One of the most prominent improvements that could have been made in our experiment was timing. Alcohol and Cannabis Tea has different effect times on every human being, and the islanders as well. Even though we began to measure the Cannabis Tea two hours after ingested to provide the most concrete data, this was simply an average amount of time presumed for Cannabis Tea to influence neural functions. However, this span of time might only apply to a limited number of islanders. Similar to Cannabis Tea, this is the same case with Alcohol because it also has different effect timings for the buzz to initiate. In the best case scenario, if we had actual data from the creators of the Island that

explained the effect and time intervals of the various substances, this would be able to contribute greatly to our final conclusion.

VII. References

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