# Does Light at Night Affect Weight Gain?

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## Introduction

This paper will provide a statistical analysis on a dataset from a long-range study preformed by researchers at West Virginia University (WVU) on the effects of light at night on the weight gain or loss of mice, with the hope of using this data to extrapolate the affect of the same factor on humans. Through generations of genetic modifications, mammals have evolved to significantly biologically react to light. As artificial forms of light have only existed for a tiny sliver of human and evolutionary history, our mental and physical biology is largely dependent upon natural sunlight and darkness. The circadian rhythm, a neurological mechanism that governs the natural production of melatonin, serotonin, epinephrine, dopamine, and a long list of other crucial neurotransmitters in setting mood, alertness, and wakefulness, is largely set by the activation of light receptors in the retina. The type, duration, and intensity of light intercepted by the eye informs the suprachiasmatic nucleus to emit chemicals at certain times during the day-for instance high levels of yellow light, which is usually produced by the morning sunshine, inform this organ to produce higher levels of serotonin and epinephrine to generate a feeling of wakeful alertness. In areas such as Northern Alaska, where there is a significantly altered night and day cycle, depression skyrockets, and with it, weight gain. Any alteration to this circadian rhythm can severely effect the happiness of organisms, which in turn affects activity and weight gain. As organisms become more sedentary, which often occurs due to depression, they gain weight in an unhealthy way, which can lead to health complications. This study uses mice as a testing group to analyze the effect of light at night, which can interrupt the natural sleep cycle, on weight gain. This data can hopefully be used to advise medical professionals on how to mitigate depression and resultant weight gain. Our belief is that the overall effect of light will have an adverse effect on weight gain, with the goal of this statistical analysis being the rejection of the null hypothesis that true mean weight of the overall population of mice is not effected by light at night (LaN) in favor of the alternative hypothesis that the true mean weight of mice that are exposed to LaN is affected by LaN. We will examine this statistical analysis in four parts: Analysis and Transformation of Data, Development of Data Models and Hypothesis testing, and Analysis of Variation (ANOVA).

# I. Analysis and Transformation of Data

The following data is recorded on a sample population (denoted as *n*) of 27 mice. The mice were apportioned into 3 treatment groups, each with different levels of applied LaN: Dark (no LaN/Control Group) denoted *LD*, Dim (low LaN) denoted *DM*, and Bright (high LaN) denoted *LL*. For each mouse, its classification was recorded along with 8 additional variables: Change in body mass (*BMGain*), Blood corticosterene (a stress neurotransmitter) level (*CorticoSter*), Percentage of calories consumed during the day (*DayPct*), Average daily consumption in grams (*Consumption*), Binary glucose intolerance at the conclusion of the study (*GlucInt*), Glucose levels in the blood 15 minutes after a glucose injection (*GTT15*), Glucose level in the blood 120 minutes after a glucose injection (*GTT120*), and a measure of physical activity level (*Activity*). The raw unprocessed data is presented in the following table:

| Light | BMGain | Corticosterone | DayPct | Consumption | GlucoseInt | GTT15 | GTT120 | Activity |
|---|---|---|---|---|---|---|---|---|
| DM | 10.2 | 128.56 | 40.848 | 3.414 | No | 319.266 | 94.495 | 1409 |
| DM | 7.29 | 124.43 | 47.45 | 3.219 | Yes | 335.772 | 279.675 | 509 |
| DM | 7.57 | 98.517 | 56.429 | 3.613 | Yes | 343.59 | 412.821 | 2003 |
| DM | 3.42 | 208.26 | 55.051 | 3.857 | No | 271.717 | 148.485 | 1084 |
| DM | 5.82 | 80.685 | 48.352 | 3.587 | Yes | 402.941 | 335.294 | 1848 |
| DM | 10.92 | 26.41 | 67.635 | 4.514 | Yes | 380.808 | 274.747 | 1841 |
| DM | 5.21 | 3 | 42.969 | 4.231 | No | 400 | 169.369 | 2716 |
| DM | 13.47 | 3 | 72.864 | 5.324 | Yes | 328.571 | 328.571 | 4622 |
| DM | 8.64 | 49.142 | 66.746 | 4.633 | Yes | 445.833 | 398.958 | 1744 |
| DM | 6.05 | 11.994 | 56.816 | 4.849 | No | 159.048 | 144.762 | 7253 |
| LD | 5.02 | 87.838 | 31.063 | 3.791 | No | 228.448 | 134.483 | 1437 |
| LD | 6.67 | 191.22 | 41.408 | 3.923 | No | 231.183 | 220.43 | 2541 |
| LD | 8.17 | 67.7 | 47.573 | 4.489 | No | 226.563 | 141.406 | 346 |
| LD | 2.79 | 41.017 | 34.947 | 4.161 | No | 323.077 | 199.038 | 5837 |
| LD | 8.13 | 21.817 | 41.94 | 4.416 | No | 500 | 190.361 | 877 |
| LD | 6.34 | 23.403 | 40.5 | 4.89 | No | 280 | 118.333 | 1649 |
| LD | 6.32 | 70.47 | 28.95 | 4.946 | No | 299.174 | 153.719 | 728 |
| LD | 3.97 | 56.718 | 21.846 | 4.004 | No | 461.25 | 230 | 6048 |
| LL | 9.89 | 42.132 | 71.552 | 3.387 | Yes | 378.704 | 328.704 | 5752 |
| LL | 9.58 | 48.238 | 61.453 | 3.451 | No | 379.091 | 227.273 | 1256 |
| LL | 11.2 | 92.191 | 85.978 | 3.501 | Yes | 366.129 | 383.871 | 244 |
| LL | 9.05 | 51.999 | 64.827 | 4.24 | No | 392.373 | 250 | 931 |
| LL | 12.33 | 12.252 | 81.6 | 3.479 | Yes | 466.346 | 470.192 | 3582 |
| LL | 9.39 | 3 | 87.257 | 5.94 | Yes | 259.615 | 413.462 | 2657 |
| LL | 10.88 | 132.4 | 70.441 | 4.586 | No | 348.78 | 126.016 | 153 |
| LL | 9.37 | 8.615 | 84.415 | 4.873 | Yes | 335.652 | 286.957 | 4482 |
| LL | 17.4 | 66.679 | 81.636 | 7.177 | Yes | 435.644 | 405.941 | 6702 |

The following are relational tables that define statistical parameters for each variable based upon the data presented above. For each classification group, each variable is processed and Mean ($\mu$), Sample Variance ($S^2$), Standard Deviation ($S$) is computed. Note: the statistical values computed from Glucose Intolerance are based upon a conversion from *(Yes, No)* to *(1, 0)*. The calculated

***Control Group (LD)***

| *Statistic* | *BMGain* | *Corticosterone* | *DayPct* | *Consumption* | *GlucoseInt* | *GTT15* | *GTT120* | *Activity* |
|---|---|---|---|---|---|---|---|---|
| $\mu$ | *5.92625* | *70.022875* | *36.028* | *4.3275* | *0* | *318.712* | *173.471* | *2432.875* |
| $S^2$ | *3.607798214* | *2932.47101* | *70.6041* | *0.188098571* | *0* | *11340.3* | *1759.74* | *5136787* |
| $S$ | *1.899420494* | *54.15229459* | *8.40262* | *0.433703322* | *0* | *106.491* | *41.9493* | *2266.448* |
| $n$ | *8* | | | | | | | |

***Low Exposure (DM)***

| *Statistic* | *BMGain* | *Corticosterone* | *DayPct* | *Consumption* | *GlucoseInt* | *GTT15* | *GTT120* | *Activity* |
|---|---|---|---|---|---|---|---|---|
| $\mu$ | *7.859* | *73.3998* | *55.516* | *4.1241* | *0.6* | *338.75* | *258.72* | *2502.9* |
| $S^2$ | *9.055832222* | *4555.263168* | *118.4* | *0.481350989* | *0.26667* | *6489.9* | *12772* | *3997535* |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| *S* | *3.009290983* | *67.49268974* | *10.88* | *0.69379463* | *0.516397779* | *80.5602* | *113.01* | *1999.384* |
| *n* | *10* | | | | | | | |

***High Exposure (LL)***

| *Statistic* | *BMGain* | *Corticosterone* | *DayPct* | *Consumption* | *GlucoseInt* | *GTT15* | *GTT120* | *Activity* |
|---|---|---|---|---|---|---|---|---|
| $\mu$ | *11.01* | *50.834* | *76.5732* | *4.514888889* | *0.666666667* | *373.593* | *321.380* | *2862.111* |
| $S^2$ | *6.8853* | *1782.175665* | *93.0498* | *1.728958861* | *0.25* | *3495.55* | *11879.6* | *5847891* |
| *S* | *2.623985518* | *42.21582244* | *9.64623* | *1.314898803* | *0.5* | *59.1232* | *108.994* | *2418.241* |
| *n* | *9* | | | | | | | |

# II. Development of Data Models

In this section we will generate the statistical models that will be used to evaluate our hypothesis tests. Our goal in this section is to generate T-Distributions that evaluate the hypothesis that the two population means are equal, as well as generate tables that allow for a more subjective, visual side-by-side comparison of different variables.

## Part A: Developing 2-Sample Distributed Hypothesis Tests

In this section we will utilize a T-distribution to test the hypothesis that 2 means are not significantly different at varying significance levels. In this situation we are testing *"$H_0$: $\mu_{LD,\,i} = \mu_{DM,\,i}$"* and *"$H_0$: $\mu_{LD,\,i} = \mu_{LL,\,i}$"* where $\mu_{LD,\,i}$ is the true mean value of the Control Group on variable $i$, $\mu_{DM,\,i}$ is the true mean value of the Low Exposure Group at variable $i$, and $\mu_{LL,\,i}$ is the true mean value of the High Exposure Group at variable $i$. We will test these hypotheses at arbitrary significance levels $\alpha = 0.2, 0.1, 0.05, 0.02, 0.01, 0.002, 0.001$. Our test statistic for a 2-sample distributed hypothesis test is shown in *figure (a)*. This statistic is used to derive the confidence interval, as shown in *figure (b)*. However, as the population standard deviation ($\sigma$) of the Control Group and the Low Exposure Group are both unknown and not necessarily equal, we must use a more complex formula to derive the number of degrees of freedom, as shown in *figure (c)*.

*Figure (a)*

$$T = \frac{(\bar{X}_{LD} - \bar{X}_{DM\ or\ LL}) - (\mu_{LD} - \mu_{DM\ or\ LL})}{\sqrt{{S_{LD}}^2/n_{LD} + {S_{DM\ or\ LL}}^2/n_{DM\ or\ LL}}}$$

*Figure (b)*

$$(\mu_{LD} - \mu_{DM\ or\ LL}) = (\bar{X}_{LD} - \bar{X}_{DM\ or\ LL}) \pm t_{\alpha/2,\nu}\sqrt{\frac{{S_{LD}}^2}{n_{LD}} + \frac{{S_{DM\ or\ LL}}^2}{n_{DM\ or\ LL}}}$$

*Figure (c)*

$$\nu = \left\lfloor \frac{\left(\frac{{S_{LD}}^2}{n_{LD}} + \frac{{S_{DM\ or\ LL}}^2}{n_{DM\ or\ LL}}\right)^2}{\frac{({S_{LD}}^2/n_{LD})^2}{n_{LD}-1} + \frac{({S_{DM\ or\ LL}}^2/n_{DM\ or\ LL})^2}{n_{DM\ or\ LL}-1}} \right\rfloor$$

This means that for the 8 statistical variables observed by scientists, we will perform 2 Hypothesis Tests with a lot of complex calculations at 7 different significance levels. That's (2 x 7 x 8) = 112 iterations of calculations. This is far too much work to do by hand so I wrote a Java Method that will do all the calculations for us. To begin, I composed a 2-dimensional matrix with attributes as columns and data entities as rows. The proper estimator for the variable $\Delta$, which is a representation of difference between population means ($\mu_1$ - $\mu_2$), is the variable $\Delta_o$, a representation of the difference in sample means ($\bar{X}$ - $\bar{Y}$). I computed these for each variable as displayed above in the table above. In order to use these formulas on each piece of data, I also needed to find the variance, which I computed for each variable, shown in the table above. With these two values, I finally had enough data to generate a reliable confidence interval for the true mean difference between any two data. Using the java method I constructed, which requires only the two data sets, a significance level, and the variable name, I was able to generate the following data. For each set of confidence intervals, I will discuss the meaning in context.

Control Group to Low Light Group

1. Body Mass Gain

```
We can say with 80.0% Confidence that the true mean difference of BMGain is between
0.3708689305795416 and 3.4946310694204556.
We can say with 90.0% Confidence that the true mean difference of BMGain is between
-0.10899311312010651 and 3.974493113120104.
We can say with 95.0% Confidence that the true mean difference of BMGain is between
-0.5492548910775508 and 4.414754891077548.
We can say with 98.0% Confidence that the true mean difference of BMGain is between
-1.097835042976906 and 4.963335042976903.
We can say with 99.0% Confidence that the true mean difference of BMGain is between
-1.499661268890447 and 5.365161268890445.
We can say with 99.8% Confidence that the true mean difference of BMGain is between
-2.4151262357543386 and 6.280626235754336.
We can say with 99.9% Confidence that the true mean difference of BMGain is between
-2.811128893176379 and 6.676628893176376.
```

First, lets examine our sample means and make a projection our sample mean for the control group is 5.92625, and for the low exposure group is 7.859. Body mass gain is a measure of weight that is contingent upon a mouses volume. The hypothesis here is that the affect of light at night is an increase in weight as a result of depression or lack of energy. Mice that have low energy or low dopamine may be prone to eat more, which increases body mass. Based on the sample means, this is likely to be true. However, as shown on our confidence intervals, this cannot be proven true definitively above a 80% confidence level. This means that there is a 20% chance that the true mean difference in the two populations is actually less in the control group than in the low exposure group.

2. Corticosterone

```
We can say with 80.0% Confidence that the true mean difference of Corticosterone is
between -35.07226462785809 and 41.82611462785809.
We can say with 90.0% Confidence that the true mean difference of Corticosterone is
between -46.88514018839316 and 53.63899018839316.
We can say with 95.0% Confidence that the true mean difference of Corticosterone is
between -57.72316679490349 and 64.47701679490349.
We can say with 98.0% Confidence that the true mean difference of Corticosterone is
between -71.22769201095208 and 77.98154201095208.
```

```
We can say with 99.0% Confidence that the true mean difference of Corticosterone is
between -81.11954169149723 and 87.87339169149723.
We can say with 99.8% Confidence that the true mean difference of Corticosterone is
between -103.65575574630444 and 110.40960574630444.
We can say with 99.9% Confidence that the true mean difference of Corticosterone is
between -113.40424528655184 and 120.15809528655184.
```

For corticosterone, the sample mean was 70.022875 for the control group and 73.3998 for the low exposure group. This is a relatively small difference in means, as it is less than 5% of the low exposure sample mean. Corticosterone is a neurochemical found in birds, amphibians, and mammals. A slight correlation has been proven to exist between corticosterone, glucose levels, and stress levels, however LaN does not seem to affect it, as none of the tests at any significance level showed any large change in corticosterone, at least at low exposure to light.

3. Calorie Consumption

```
We can say with 80.0% Confidence that the true mean difference of CalConsump is
between 0.37573962677180384 and 1.1747805547281958.
We can say with 90.0% Confidence that the true mean difference of CalConsump is
between 0.2521704721138637 and 1.298349709386136.
We can say with 95.0% Confidence that the true mean difference of CalConsump is
between 0.13810663704499582 and 1.4124135444550037.
We can say with 98.0% Confidence that the true mean difference of CalConsump is
between -0.004176115553930493 and 1.5546962970539302.
We can say with 99.0% Confidence that the true mean difference of CalConsump is
between -0.1090316722708844 and 1.659551853770884.
We can say with 99.8% Confidence that the true mean difference of CalConsump is
between -0.34963507436927754 and 1.9001552558692771.
We can say with 99.9% Confidence that the true mean difference of CalConsump is
between -0.45449063108623156 and 2.005010812586231.
```

As the DayPct variable is a proportion of the total consumption for the day, I surmised that a far more statistically significant variable for consideration in terms of its effects on increased body mass would be the percent of calories consumed multiplied by the daily consumption. The confidence intervals for this new variable, CalConsump, are shown above. The data produced here shows that we can say with 95% confidence that the true mean difference in populations lies above 0.

4. Daily Consumption

```
We can say with 80.0% Confidence that the true mean difference of Consumption is
between -0.5623459494908702 and 0.15554594949087153.
We can say with 90.0% Confidence that the true mean difference of Consumption is
between -0.6726261368064845 and 0.2658261368064858.
We can say with 95.0% Confidence that the true mean difference of Consumption is
between -0.773805531964985 and 0.3670055319649863.
We can say with 98.0% Confidence that the true mean difference of Consumption is
between -0.8998782703767674 and 0.49307827037676866.
We can say with 99.0% Confidence that the true mean difference of Consumption is
between -0.9922245437357163 and 0.5854245437357176.
We can say with 99.8% Confidence that the true mean difference of Consumption is
between -1.2026134447795824 and 0.7958134447795838.
```

```
We can say with 99.9% Confidence that the true mean difference of Consumption is
between -1.2936213663507206 and 0.8868213663507218.
```

The sample mean between the low exposure group and the control group is -0.2065, which is a pretty small proportion of the mean values. In fact the sample mean of the low exposure group in this case is less than that of the control group, which means that the mice that had less exposure to light actually ate more. Our goal by recording this is to distill our data and ensure that the weight gain observed in BMGain is in fact due to LaN and not increased eating habits. If these two were more correlated, we might say that increased LaN generally points to increased volume of consumption.

5.Glucose Intake

This variable is a proportion, which means that a regular t-test will not function properly. As this is only one variable and I'm sufficiently sick of programming, I will do this one by hand. For this problem we will use a score confidence interval, shown below

$$\textit{Score CI} \qquad (p_1 - p_2) = (\hat{p}_1 - \hat{p}_2) \pm Z_{\alpha/2}\sqrt{\frac{\hat{p}_1(1-\hat{p}_1)}{n_1} + \frac{\hat{p}_2(1-\hat{p}_2)}{n_2}}$$

To find our precise Z-score, as I don't desire to compute 7 different significance level confidence intervals by hand, we will use the test statistic

$$Z_o = \frac{(\hat{p}_1 - \hat{p}_2)}{\sqrt{\hat{p}(1-\hat{p})(1/n_1 + 1/n_2)}} \qquad \text{Where} \qquad \hat{p} = \frac{n_1\hat{p}_1 + n_2\hat{p}_2}{n_1 + n_2}$$

The sample proportion ($\hat{p}$) for the sample groups are

$\hat{p}_{LD} = 0$
$\hat{p}_{DM} = 0.6$
$\hat{p}_{LL} = 0.6667$

Meaning the pooled proportions for both difference tests are

$$\text{DM-LD} = \frac{8(0)+10(0.6)}{18} = 0.333$$

$$\text{LL-LD} = \frac{8(0)+9(0.6666667)}{17} = 0.352958$$

And our test statistics, $Z_{DM\text{-}LD}$ and $Z_{LL\text{-}LD}$, are

$$Z_{DM-LD} = \frac{(0.6-0)}{\sqrt{0.333(1-0.333)(1/8+1/10)}} = 2.684$$

$$Z_{LL-LD} = \frac{(0.6667-0)}{\sqrt{0.352958(1-0.352958)(1/9+1/8)}} = 2.871$$

And the confidence intervals for these scores are

$$(p_{DM} - p_{LD}) = (0.6 - 0) \pm 2.684\sqrt{\frac{0.6(0.4)}{10} + \frac{0}{8}}$$

$$(p_{LL} - p_{LD}) = (0.6667 - 0) \pm 2.871\sqrt{\frac{0.6667(0.3333)}{9} + \frac{0}{8}}$$

Which corresponds to the p-values $0.003 > \alpha > 0.002$ for the first test and $0.0047 > \alpha > 0.0046$ for the second. In context this means that the difference in population proportions in each respective test is above 0 for the inverse of the probability range above or higher. So, test 1 proves the alternate hypothesis correct for significance levels greater than 0.003, and test 2 for values greater than 0.0047.

6. Blood Glucose Levels After 15 Minutes

```
We can say with 80.0% Confidence that the true mean difference of GTT15 is between
-41.599860817593246 and 81.6853108175934.
We can say with 90.0% Confidence that the true mean difference of GTT15 is between
-60.96545193727964 and 101.05090193727979.
We can say with 95.0% Confidence that the true mean difference of GTT15 is between
-79.01272816853667 and 119.09817816853682.
We can say with 98.0% Confidence that the true mean difference of GTT15 is between
-101.83321347858967 and 141.91866347858982.
We can say with 99.0% Confidence that the true mean difference of GTT15 is between
-118.83492962591998 and 158.92037962592013.
We can say with 99.8% Confidence that the true mean difference of GTT15 is between
-158.6116719492195 and 198.69712194921965.
We can say with 99.9% Confidence that the true mean difference of GTT15 is between
-176.24981597372258 and 216.33526597372273.
```

This is a measurement of how the mice's bodies processed glucose within 15 minutes. It is slightly less reliable than the measurement at 2 hours, but it still provides useable data. Higher glucose levels usually indicate diabetes. At least for this group, the data did not significantly affect the mean.

7. Blood Glucose Levels After 2 Hours

```
We can say with 80.0% Confidence that the true mean difference of GTT120 is between
32.508300106790195 and 137.9845998932099.
We can say with 90.0% Confidence that the true mean difference of GTT120 is between
15.754361072483618 and 154.73853892751646.
We can say with 95.0% Confidence that the true mean difference of GTT120 is between
0.08381763392897312 and 170.40908236607112.
We can say with 98.0% Confidence that the true mean difference of GTT120 is between
-19.92030818763338 and 190.41320818763347.
We can say with 99.0% Confidence that the true mean difference of GTT120 is between
-34.93307572876722 and 205.4259757287673.
We can say with 99.8% Confidence that the true mean difference of GTT120 is between
-70.4916668893394 and 240.98456688933948.
We can say with 99.9% Confidence that the true mean difference of GTT120 is between
-86.43305922683203 and 256.9259592268321.
```

Here, we can say with at least a 95% confidence that the true mean difference of the population is above 0. At 80%, we can conclude that that mean is at least ~32.508 units higher. This is concrete evidence that there is a very strong likelihood of LaN affecting glucose levels.

8. Activity

```
We can say with 80.0% Confidence that the true mean difference of Activity is between
-1302.8318670168253 and 1442.8818670168255.
We can say with 90.0% Confidence that the true mean difference of Activity is between
-1727.4478199380144 and 1867.4978199380146.
We can say with 95.0% Confidence that the true mean difference of Activity is between
-2119.4010072498813 and 2259.4510072498815.
We can say with 98.0% Confidence that the true mean difference of Activity is between
-2608.321779964424 and 2748.371779964424.
We can say with 99.0% Confidence that the true mean difference of Activity is between
-2968.6329130922595 and 3108.6829130922597.
We can say with 99.8% Confidence that the true mean difference of Activity is between
-3795.4091675782292 and 3935.4591675782294.
We can say with 99.9% Confidence that the true mean difference of Activity is between
-4155.720300706065 and 4295.7703007060645.
```

The physical activity of mice can decrease their weight overall. However, the variance for this variable was incredibly high, and as a result of that it yielded low statistical significance. To say with any surety what the true mean difference between these two populations would be, we would need a much higher sample size.

<u>Control Group to High Exposure Group</u>

1. Body Mass Gain

```
We can say with 80.0% Confidence that the true mean difference of BMGain is between
1.42289630936165 and 4.879103690638352.
We can say with 90.0% Confidence that the true mean difference of BMGain is between
0.894254267872431 and 5.40774573212757.
We can say with 95.0% Confidence that the true mean difference of BMGain is between
0.41085054289206946 and 5.891149457107932.
We can say with 98.0% Confidence that the true mean difference of BMGain is between
-0.18758775835367203 and 6.489587758353673.
We can say with 99.0% Confidence that the true mean difference of BMGain is between
-0.6244606434963509 and 6.926460643496352.
We can say with 99.8% Confidence that the true mean difference of BMGain is between
-1.6132409900470908 and 7.915240990047092.
We can say with 99.9% Confidence that the true mean difference of BMGain is between
-2.0384811652303494 and 8.34048116523035.
```

Here, there is at least a 95% confidence that the true mean difference in population means is above 0, which is a significantly higher confidence level than the BMGain in the previous test.

2. Corticosterone

```
We can say with 80.0% Confidence that the true mean difference of Corticosterone is
between -56.84785154131635 and 11.716251541316346.
We can say with 90.0% Confidence that the true mean difference of Corticosterone is
between -67.38044306631437 and 22.248843066314357.
We can say with 95.0% Confidence that the true mean difference of Corticosterone is
between -77.04384014507468 and 31.91224014507467.
We can say with 98.0% Confidence that the true mean difference of Corticosterone is
between -89.08473967972046 and 43.95313967972046.
We can say with 99.0% Confidence that the true mean difference of Corticosterone is
between -97.9045068547795 and 52.772906854779485.
We can say with 99.8% Confidence that the true mean difference of Corticosterone is
between -117.9982372883922 and 72.86663728839221.
We can say with 99.9% Confidence that the true mean difference of Corticosterone is
between -126.69018175076923 and 81.5585817507692.
```

In this iteration of comparison tests, as well as the last one, corticosterone seems to have little effect, even though its sample mean is significantly lower than the sample mean of the control group, by about 20 units. This, in conjunction with the confidence intervals shown above, proves that even at an 80% significance level, there is not significant proof that shows that there is a decrease in corticosterone when LaN is high.

3. Daily Percent Calories

```
We can say with 80.0% Confidence that the true mean difference of CalConsump is
between 0.48822412848790475 and 1.8347424730676518.
We can say with 90.0% Confidence that the true mean difference of CalConsump is
between 0.27826700883306277 and 2.044699592722494.
We can say with 95.0% Confidence that the true mean difference of CalConsump is
between 0.08426862511398059 and 2.2386979764415758.
We can say with 98.0% Confidence that the true mean difference of CalConsump is
between -0.16009951890234375 and 2.4830661204579005.
We can say with 99.0% Confidence that the true mean difference of CalConsump is
between -0.34063271917562843 and 2.663599320731185.
We can say with 99.8% Confidence that the true mean difference of CalConsump is
between -0.7595495374893273 and 3.082516139044884.
We can say with 99.9% Confidence that the true mean difference of CalConsump is
between -0.9435737112485594 and 3.266540312804116.
```

Here, as before we can say with 95% confidence that the true mean difference in the derived data value CalConsump is above 0.

4. Daily Consumption

```
We can say with 80.0% Confidence that the true mean difference of Consumption is
between -0.27727807591802844 and 1.0588558536958053.
We can say with 90.0% Confidence that the true mean difference of Consumption is
between -0.48951064801002775 and 1.2710884257878046.
We can say with 95.0% Confidence that the true mean difference of Consumption is
between -0.6880191738697501 and 1.469596951647527.
We can say with 98.0% Confidence that the true mean difference of Consumption is
between -0.9414238846585805 and 1.7230016624363573.
We can say with 99.0% Confidence that the true mean difference of Consumption is
between -1.1315999538772772 and 1.913177731655054.
```

```
We can say with 99.8% Confidence that the true mean difference of Consumption is
between -1.5820427570009434 and 2.36362053477872.
We can say with 99.9% Confidence that the true mean difference of Consumption is
between -1.7839822944187347 and 2.5655600721965115.
```

Here, we can see that here, just as in consumption for the first comparison tests, we cannot determine with statistical surety that the true mean difference in consumption is above 0, and consequently we cannot say that it increases. This will be valuable information when deciding which variables to include in our ANOVA test.

5. Glucose Intake

See part 5 of test 1.

6. Blood Glucose Levels after 15 minutes

```
We can say with 80.0% Confidence that the true mean difference of GTT15 is between
-8.224712477158384 and 77.90084581049166.
We can say with 90.0% Confidence that the true mean difference of GTT15 is between
-21.3979934568326 and 91.07412679016588.
We can say with 95.0% Confidence that the true mean difference of GTT15 is between
-33.443976553160624 and 103.12010988649389.
We can say with 98.0% Confidence that the true mean difference of GTT15 is between
-48.35651712428327 and 118.03265045761654.
We can say with 99.0% Confidence that the true mean difference of GTT15 is between
-59.24299382631233 and 128.9191271596456.
We can say with 99.8% Confidence that the true mean difference of GTT15 is between
-83.88250470516509 and 153.55863803849837.
We can say with 99.9% Confidence that the true mean difference of GTT15 is between
-94.47910480861941 and 164.15523814195268.
```

In truth, it is unlikely that this variable will have much statistical significance, as glucose levels measured at 15 minutes rarely indicate whether or not an organism has developed diabetes. According to multiple academic sources, the litmus test for diabetic mice is a blood glucose level of 200 mg/dL. This is clearly higher in the high exposure group than it is in the control group, however, even at significance level 0.2 (80), there is not enough evidence to say that the difference in population means is certainly above 0.

7. Blood Glucose Levels After 2 Hours

```
We can say with 80.0% Confidence that the true mean difference of GTT120 is between
-5.474156506624894 and 130.79786761773582.
We can say with 90.0% Confidence that the true mean difference of GTT120 is between
-26.317558850253732 and 151.64126996136466.
We can say with 95.0% Confidence that the true mean difference of GTT120 is between
-45.37729595665273 and 170.70100706776367.
We can say with 98.0% Confidence that the true mean difference of GTT120 is between
-68.97263895200763 and 194.29635006311855.
We can say with 99.0% Confidence that the true mean difference of GTT120 is between
-86.1977489572559 and 211.52146006836682.
We can say with 99.8% Confidence that the true mean difference of GTT120 is between
-125.18357485670839 and 250.5072859678193.
```

```
We can say with 99.9% Confidence that the true mean difference of GTT120 is between
-141.950028086669 and 267.27373919777995.
```

Here, we may have slightly more statistically significant data, as the data is approaching a definite increase in the population mean. This is not necessarily completely insignificant.

8. Activity

```
We can say with 80.0% Confidence that the true mean difference of Activity is between
-1014.5902962405335 and 1733.012518462756.
We can say with 90.0% Confidence that the true mean difference of Activity is between
-1436.6679844052446 and 2155.090206627467.
We can say with 95.0% Confidence that the true mean difference of Activity is between
-1823.9140186922855 and 2542.336240914508.
We can say with 98.0% Confidence that the true mean difference of Activity is between
-2306.4348709388364 and 3024.857093161059.
We can say with 99.0% Confidence that the true mean difference of Activity is between
-2659.8737117563737 and 3378.295933978596.
We can say with 99.8% Confidence that the true mean difference of Activity is between
-3465.0995925754587 and 4183.521814797681.
We can say with 99.9% Confidence that the true mean difference of Activity is between
-3813.416131352162 and 4531.838353574385.
```

Just as in the previous tests, the variance was high enough that we cannot say with any degree of statistical surety that activity increases or decreases when high levels of LaN are applied, as the lower bound of the confidence interval is not even close to surpassing 0.

# III. Analysis of Variation

For each of the tests performed in section 2, some variables were more effective than others in showing a difference in the true mean value of the difference of the population. The following table provides a side-by-side comparison of what variables allowed us to say with statistical significance that the true mean difference in population was above 0, and at the corresponding significance levels.

| | Test 1 (DM-LD) | Test 2 (LL-LD) |
|---|---|---|
| BMGain | 80% | 95% |
| Corticosterone | - | - |
| Calorie Intake | 95% | 95% |
| Consumption | - | - |
| Glucose Intake | 97% | 95.3% |
| GTT15 | - | - |
| GTT120 | 95% | - |
| Activity | - | - |

These values are helpful in determining the statistical significance of LaN, however, they lack the ability to measure interactive affects between variables in DM and LL. I could do an entire set of these calculations, but instead, a more comprehensive approach to multivariable analysis is with an ANOVA table. These calculate a useable F ratio to test the null hypothesis that all population means in a dataset are equal (sum = 0). For example, and to begin with, the following is the completed ANOVA table for the null hypothesis that all three population means of BMGain are the same:

| Source of Var | DF | SS | MS | F |
|---|---|---|---|---|
| Variable | 2 | 113.0829 | 56.54146 | 8.384821 |
| Error | 24 | 161.8395 | 6.743312 | |
| Total | 26 | 274.9224 | | |

This table has an f ratio with numerator degrees of freedom = 2 and denominator degrees of freedom = 24. The result of our calculations for this table is F(2, 24) = 8.384821. Luckily, in our case for this series of experience, our degrees of freedom do not change, so using F tables, the following f ratios correspond with their given significance level at F(2, 24).

| *Significance Level* | *0.2* | *0.1* | *0.5* | *0.02* | *0.01* | *0.002* | *0.001* |
|---|---|---|---|---|---|---|---|
| *F(2, 24)* | *1.72* | *2.54* | *3.40* | *4.63* | *5.61* | *8.15* | *9.33* |

So, if our F values fall above a given significance level, then we can say that the there is at least 1 instance of a population mean not being equal to another population mean. For example, the F value calculated in the table above for BMGain indicated that the null hypothesis is rejected at any significance level higher than 0.001. This performs significantly better than our T-tests in part II.

To avoid further unneeded clutter on this paper, I will simply make a table of the results of all F tests with corresponding rejection boundaries based on the significance levels above. If the F value is less than the 0.2 significance level, it is safe to assume that the true means are approximately equal.

| *Variable* | *F value* | *Rejection Boundary* |
|---|---|---|
| *BMGain* | *8.38* | *$\alpha > 0.002$* |
| *Corticosterone* | *0.43* | - |
| *CalConsumpt* | *9.61* | *$\alpha > 0.001$* |
| *Consumption* | *0.45* | - |
| *GlucoseInt* | *6.18* | *$\alpha > 0.01$* |
| *GTT15* | *0.96* | - |
| *GTT120* | *5.01* | *$\alpha > 0.02$* |
| *Activity* | *0.09* | - |

Clearly, there are some noticeable differences here between ANOVA results and the T Distribution results. This could be a result of interplay between the sample means of the 2 non-control groups, but we won't know until we take a Studentized Range Test of all the variables but Corticosterone, Consumption, GTT15, and Activity, as they each had a low enough F value to say that all three population means are equal.

Tukey's Studentized Range Test

This test utilizes the following confidence interval to apply to a set of means to determine which means are significantly different. Here is a table like above that shows Q distribution values at given significance levels that we will use to evaluate the range test:

| *Significance Level* | *0.2* | *0.1* | *0.5* | *0.025* | *0.01* | *0.005* | *0.001* | *0.0005* |
|---|---|---|---|---|---|---|---|---|
| *Q(2, 24)* | | *2.42* | *2.92* | *3.38* | *3.96* | *4.37* | *5.30* | *6.41* |

Unfortunately, I could not find a value of 0.2 online, however I did not end up needing it. These values will be plugged into the following Measure of Error (MoE) formula to create a width to compare to the sample means to determine which population means are significantly different. I determined the Q value by using the first one in the above table to exist in the rejection region found in the ANOVA test.

$$MoE = Q_{\alpha,k,\nu}\sqrt{\frac{Mean\ Squared\ Error}{N}}$$

| *Variable* | *Q value* | *MoE* | *Significantly Different Means* |
|---|---|---|---|
| *BMGain* | *5.30* | *2.6487* | *{LD,LL}, {DM,LL}* |
| *CalConsumpt* | *6.41* | *113.5580* | *{LD,LL}, {DM,LL}* |
| *GlucoseInt* | *4.37* | *0.3601* | *{LD,DM}, {LD,LL}* |
| *GTT120* | *3.38* | *52.3133* | *{DM,LD}, {DM, LL}, {LD,LL}* |

Result

So, as a result of our ANOVA calculations, we can say that there is significant evidence to claim that the population means of Body Mass Gain, Glucose Intake, Calorie Intake, and Diabetes Positivity (GTT120) are significantly higher for mice who are exposed to high levels of LaN, and the population means of Glucose Intake and Diabetes Positivity are also significantly higher for mice who are exposed to at least dim LaN. In addition to that we can say that the population means of Body Mass Gain, Calorie Intake, and Diabetes Positivity rates are statistically significantly higher in high LaN exposure mice than low exposure mice, however this is not a solution of "Does LaN Affect Weight Gain", though the data is interesting.

# Conclusion

In this paper, I modeled and analyzed upwards of 100 T distributions and surmised as a result of these tests that generally speaking there is significant data to conclude that Body Mass, Calorie Intake, Glucose Intake, and Diabetes Rates are significantly higher for mice when LaN is applied. However, after we concluded our ANOVA calculations, there was enough evidence to say that the interplay between the low exposure group and the high exposure group went unmeasured. We then were able to pin down exactly which groups have significant differences and which do not. But what does all of this mean in context?

Looking only at the difference in means for the Control Group and the high LaN exposure group, as those show us the most concrete change in population means, we can say that following variables increased: Body Mass Gain, Calorie Consumption, Glucose Intake, and

Diabetes Rate. However, the crucial question that we have to ask is whether or not the gain in body mass is a result of LaN. In the experiment, we can conclude that the population mean of body mass is very likely higher in mice exposed to high light at night, but we have to consider the other variables that changed with this mean. When an organism takes in more calories and does not expend them, those unused calories are stored as fat, which causes weight gain. In our analysis, we found that for high LaN groups compared to the control group, the weight and the calorie intake increased significantly, but not the activity, meaning these unused calories could be what is causing the increase in weight. Additionally, the same can be said for glucose intake, which if too high causes diabetes, and consequently diabetes rate and their effects on weight gain. Though Body Mass Gain was higher for the LaN group, so was the Diabetes rate.

Conversely, when we consider which effects did not end up being statistically relevant, it gives us more information as to why Body Mass Gain was so much higher. Corticosterone, the neurochemical that been shown to affect and be affected by both your adrenal system and your thyroid system, did not increase with statistical significance. Our hypothesis that LaN affected weight gain was based upon this interplay of weight gain and stress due to this chemical and as it did not increase, it likely means that our hypothesis was incorrect. So**:** does LaN affect weight gain? Yes. Does it do so as a result of neurochemical imbalances created by sleep quality deprivation? Not necessarily.

But what does all this say about human populations? While corticosterone does have similar effects in humans as it does in mice, that does not necessarily mean that human populations that experience LaN will not have a statistically significant difference in corticosterone for a similar test. The brains of mice are complicated, but the average mouse brain is a handful of interplaying variables that form a relatively simple feature space when compared to the almost metaphysically incomprehensible complexity of the human central nervous system. But now we can definitively say that mice that have an exposure to excess light at night may need to consider intermittent fasting or keto.

---

Java Method:

```
public static void twoSampleDistHypothesisTest(double [] dataSet1, double dataSet2[], double sl, String varName)
	{
		int n1 = dataSet1.length;
		int n2 = dataSet2.length;

		double sum1 = 0;
		double sum2 = 0;

		for (int i = 0; i < n1; i++)
		{
			sum1 += dataSet1[i];
		}

		double avg1 = sum1/n1;

		for (int i = 0; i < n2; i++)
		{
			sum2 += dataSet2[i];
		}
```

```java
		double avg2 = sum2/n2;

		sum1 = 0;
		sum2 = 0;

		for(int i = 0; i < n1; i++)
		{
			sum1 += Math.pow((dataSet1[i] - avg1), 2);
		}

		double var1 = sum1/(n1 - 1);

		for(int i = 0; i < n2; i++)
		{
			sum2 += Math.pow((dataSet2[i] - avg2), 2);
		}

		double var2 = sum2/(n2 - 1);

		double DF = Math.floor((Math.pow((var1/n1) + (var2/n2), 2))/
									((Math.pow(var1/n1, 2)/(n1-1)) +
(Math.pow(var2/n2, 2)/(n2-1))));

		double [] sigLevels = {	0.2,	0.1,	0.05,	0.02,	0.01,	0.002,	0.001};
		double [][]tTable ={ {	3.078,	6.314,	12.706,	31.821,	63.656,	318.289,	636.578},
				{	1.886,	2.920,	4.303,	6.965,	9.925,	22.328,	31.600},
				{	1.638,	2.353,	3.182,	4.541,	5.841,	10.214,	12.924},
				{	1.533,	2.132,	2.776,	3.747,	4.604,	7.173,	8.610},
				{	1.476,	2.015,	2.571,	3.365,	4.032,	5.894,	6.869},
				{	1.440,	1.943,	2.447,	3.143,	3.707,	5.208,	5.959},
				{	1.415,	1.895,	2.365,	2.998,	3.499,	4.785,	5.408},
				{	1.397,	1.860,	2.306,	2.896,	3.355,	4.501,	5.041},
				{	1.383,	1.833,	2.262,	2.821,	3.250,	4.297,	4.781},
				{	1.372,	1.812,	2.228,	2.764,	3.169,	4.144,	4.587},
				{	1.363,	1.796,	2.201,	2.718,	3.106,	4.025,	4.437},
				{	1.356,	1.782,	2.179,	2.681,	3.055,	3.930,	4.318},
				{	1.350,	1.771,	2.160,	2.650,	3.012,	3.852,	4.221},
				{	1.345,	1.761,	2.145,	2.624,	2.977,	3.787,	4.140},
				{	1.341,	1.753,	2.131,	2.602,	2.947,	3.733,	4.073},
				{	1.337,	1.746,	2.120,	2.583,	2.921,	3.686,	4.015},
				{	1.333,	1.740,	2.110,	2.567,	2.898,	3.646,	3.965},
				{	1.330,	1.734,	2.101,	2.552,	2.878,	3.610,	3.922},
				{	1.328,	1.729,	2.093,	2.539,	2.861,	3.579,	3.883},
				{	1.325,	1.725,	2.086,	2.528,	2.845,	3.552,	3.850},
				{	1.323,	1.721,	2.080,	2.518,	2.831,	3.527,	3.819},
				{	1.321,	1.717,	2.074,	2.508,	2.819,	3.505,	3.792},
				{	1.319,	1.714,	2.069,	2.500,	2.807,	3.485,	3.768},
				{	1.318,	1.711,	2.064,	2.492,	2.797,	3.467,	3.745},
				{	1.316,	1.708,	2.060,	2.485,	2.787,	3.450,	3.725},
				{	1.315,	1.706,	2.056,	2.479,	2.779,	3.435,	3.707},
				{	1.314,	1.703,	2.052,	2.473,	2.771,	3.421,	3.689},
				{	1.313,	1.701,	2.048,	2.467,	2.763,	3.408,	3.674},
				{	1.311,	1.699,	2.045,	2.462,	2.756,	3.396,	3.660},
				{	1.310,	1.697,	2.042,	2.457,	2.750,	3.385,	3.646}};

		double tVal = 0;

		for(int i = 0; i < 7; i++)
		{
			if (sl == sigLevels[i])
			{
				tVal = tTable[(int) DF - 1][i];
			}
		}

		double CILowerBound = (avg1 - avg2) - ((tVal) * Math.pow((var1/n1) + (var2/n2), 0.5));

		double CIUpperBound = (avg1 - avg2) + ((tVal) * Math.pow((var1/n1) + (var2/n2), 0.5));

		System.out.println("We can say with " + ((1 - sl) * 100) + "% Confidence that the true mean difference
of " + varName + " is between " + CILowerBound + " and " + CIUpperBound + ".");
	}
```

# Works Cited

Huberman, Andrew. “Master Your Sleep and Be More Alert When Awake.” Huberman Lab, Stanford University, 11 Jan. 2021, hubermanlab.libsyn.com/master-your-sleep-be-more alert-when-awake-episode-2.

Devore, Jay L. Probability and Statistics for Engineering and the Sciences. Nelson, 2018.

Soper, Daniel. “Calculator: Critical F-Value.” Free Critical F-Value Calculator - Free Statistics Calculators, www.danielsoper.com/statcalc/calculator.aspx?id=4.

Fandino, Wilson, et al. “Wilson Fandino.” Real Statistics Using Excel, www.real statistics.com/statistics-tables/studentized-range-q-table/.