Name: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Date: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Period: \_\_\_\_\_\_

**Cell Signaling Statistics Assignment**

AP Biology, U5P2



Tetrodotoxin (commonly known as TTX) is a neurotoxin found in several species including pufferfish, rough-skinned newts, ocean sunfish, blue-ringed octopus, etc. Tetrodotoxin prevents action potentials from traveling down nerve cells by binding to voltage-gated sodium channels on the nerve cell membrane. This prevents the sodium channels from opening, so Na+ will not be able to enter the nerve cell through the channels to carry out the depolarization phase of the action potential. Nerve cells cannot signal to muscle cells to contract, and this could result in death if muscles like the diaphragm are paralyzed and unable to initiate breathing.

Let’s say scientists wanted to design an experiment to see if applying TTX to nerve cells would affect the amount of Na+ ions entering the nerve cell. They compared the TTX nerve cells to normal nerve cells that were not exposed to TTX.

What is the null hypothesis for their experiment?

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

What is the alternate hypothesis for their experiment? You should express this in two ways (see below).

A) Write the alternate hypothesis as the exact opposite statement of the null hypothesis given above.

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

B) Write the alternate hypothesis in “If, then” format using the following stem… “If nerve cells are exposed to TTX, then…”

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

Let’s say the scientists collected the following data… (Note: The scientists submerged the TTX nerve cells and normal nerve cells in the same solution containing Na+ ions.)

|  |  |  |
| --- | --- | --- |
| **Trial** | **# of Na+ ions entering the TTX nerve cells** | **# of Na+ ions entering the normal nerve cells** |
| 1 | 10 | 62 |
| 2 | 3 | 74 |
| 3 | 7 | 65 |
| 4 | 4 | 69 |
| 5 | 11 | 61 |
| Sum for each column | 35 | 331 |
| Total Na+ ions entering cells | 366 |

1. Use the data in the chart above to calculate the **mean** for each set of data.

***Why use this formula?***

Use the mean formula when you want to calculate the average of a set of values (data points). When you calculate the mean for two sets of values, this allows you to easily compare the sets of values

***Formula***



*Note: I agree with you… this formula makes NO SENSE! The easiest way to calculate mean is simply by adding the data points together and dividing by the number of data points (n).*

***Additional Information from the Formula Sheet***



Record your calculated means in the table given below.

|  |  |  |
| --- | --- | --- |
|  | **# of Na+ ions entering the TTX nerve cells** | **# of Na+ ions entering the normal nerve cells** |
|  (Mean) |  |  |

2. Use the data in the chart on the previous page to calculate the **standard deviation** for each set of data.

***Why use this formula?***

Use standard deviation formula to determine the amount by which your values (data points) typically differ from the mean value. In other words, the standard deviation determines the amount of variation in your data.

***Formula***



***Additional Information from the Formula Sheet***



Record your work in row 1 of the table below, and record your calculated standard deviations in row 2 of the table given below.

|  |  |  |
| --- | --- | --- |
|  | **# of Na+ ions entering the TTX nerve cells** | **# of Na+ ions entering the normal nerve cells** |
| Work |  |  |
| s (standard deviation) |  |  |

3. Use the standard deviations you calculated to find the **standard error of the mean (SEM)** for each set of data.

***Why use this formula?***

Use the standard error formula to determine the precision of the mean value. In other words, we are determining how confident we are in our mean value by considering both the standard deviation (s) and the number of data points (n). Typically, when we have more data points, we can be more confident in our data (i.e. a lower standard error).

***Formula***



***Additional Information from the Formula Sheet***



Record your calculated SEM values in the table given below.

|  |  |  |
| --- | --- | --- |
|  | **# of Na+ ions entering the TTX nerve cells** | **# of Na+ ions entering the normal nerve cells** |
|  (SEM)  |  |  |

4. Next you will calculate the **95% confidence limit.**

***Formula***

There is no formula given on the test, but if you are asked to use the 95% confidence limit to draw error bars on a graph, they will tell you that the 95% confidence limit includes all values within the following range…

95% Cl = Mean ± 2SEM

***Why use this formula?***

The (mean + 2SEM) gives you the **error bar upper limit** (the top of the error bar). The (mean – 2SEM) gives you the **error bar lower limit** (the bottom of the error bar). The error bar provides a range of values around the mean for this sample of data. If we sampled a much larger number of data points (in this case, more trials of TTX and normal nerve cells), we would be 95% confident that the mean of this larger sample was within the error bar range.

Record your calculated error bar upper limits in row 1 of the table below, and record your calculated error bar upper limits in row 2 of the table below.

|  |  |  |
| --- | --- | --- |
|  | **# of Na+ ions entering the TTX nerve cells** | **# of Na+ ions entering the normal nerve cells** |
| Error Bar Upper Limit  |  |  |
| Error Bar Lower Limit  |  |  |

5. In the grid given on the next page, create a bar graph showing the mean # of Na+ ions entering TTX vs. normal nerve cells. (You should be graphing only two bars.) For each set of data, please graph an error bar using the error bar upper limit and error bar lower limit. Make sure to include the following elements of a proper scientific graph.

-A title that includes information about the x and y axis

-X and Y axis labels (with units where applicable)

-An appropriate scale on your X and Y axes



Is there overlap in the error bars for the two data sets? What does this mean?

5. Next, we will use a Chi square test to determine if we should reject or FAIL TO REJECT our null hypothesis. Note: In the past, we have used the phrase “accept the null hypothesis.” Statistically, this is incorrect, as we will never have enough data to fully prove / confirm our null hypothesis. Therefore, we will use the phrase “fail to reject the null hypothesis” in place of “accept the null hypothesis.”

Use the steps given below to complete your Chi square test…

***How do I perform a Chi square test?***

1. ***State the null hypothesis***
* This is a negative statement, basically saying that there is no statistically significant difference between the observed and the expected results or between two sets of data.

Please rewrite your null hypothesis from page 1 in the space below.

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

1. ***Determine your expected (and observed) values***
* If the null hypothesis is true, and the TTX and normal nerve cells are submitted in the same Na+ solution, how many Na+ ions total would you expect to enter the TTX vs. normal nerve cells? (Note: According to the data table on the first page, there are 366 Na+ ions available to enter the nerve cells.)

|  |  |
| --- | --- |
| **EXPECTED # of Na+ ions entering TTX Nerve Cells** | **EXPECTED # of Na+ ions entering normal nerve cells** |
|  |  |

* Note, your observed values represent the data actually collected during the experiment. Over the five trials conducted, how many Na+ ions total entered the TTX nerve cells vs. normal nerve cells?

|  |  |
| --- | --- |
| **OBSERVED # of Na+ ions entering TTX Nerve Cells** | **OBSERVED # of Na+ ions entering normal nerve cells** |
|  |  |

***C. Calculate 2***

* The formula is: $X^{2}=\frac{(o-e)^{2}}{e}$
* Where o = observed value, e = expected value, and ∑ = the sum of
* So you would need to calculate $\frac{(o-e)^{2}}{e}$ separately for each data set (ex: TTX nerve cells vs. normal nerve cells) and then add the results together

|  |  |  |  |
| --- | --- | --- | --- |
| Data Set | o | e | $$\frac{(o-e)^{2}}{e}$$ |
| TTX Nerve Cells |  |  |  |
| Normal Nerve Cells |  |  |  |
| $X^{2}$ (sum of all values from the last column together) |  |

***D. You will also need to know the degrees of freedom.***

* This is calculated using the formula (n-1), where n = the number of data sets.

How many degrees of freedom do you have for this experiment? \_\_\_\_\_\_\_\_\_\_\_\_\_



***E. Compare the X2 value against a table of critical numbers.***

* On the table to the right, refer to the row that corresponds to the correct number of degrees of freedom for your data set
* Look up the critical number at the p = 0.05 level. “p” stands for probability

***F. Make a conclusion***

* If the X2 value that you calculated in Step 3 is higher than the critical number at the p = 0.05 level then you can reject the null hypothesis. In other words, there is a statistically significant difference between the observed and expected results. (i.e. the observed results do not match the expected results)

*Note: A high X2 value corresponds with a low p value (below 0.05)*

* If the X2 value is less than the critical number then you fail to reject the null hypothesis. In other words, there MAY NOT be a statistically significant difference between the observed and expected results (i.e., any differences between the observed and expected results are probably due to chance alone.)

*Note: A low X2 value corresponds with a high p value (above 0.05)*

 What is your conclusion for this Chi square test? (Do you reject or fail to reject the null hypothesis?)

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

What does that mean in the context of this experiment? (Make sure to mention the alternate hypothesis in this section).

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