Research Proposal

Introduction:

The purpose of this research is to test whether the guinea pig or the hamster is better at learning and retaining information after the given training sessions held.

Hypothesis:

If guinea pigs and hamsters were put into a maze in order to test how they remember, forget, and running the maze perfectly, then the hamster would learn faster, and Guinea Pigs would be harder to teach but would forget harder and remember better as well.

Research questions:

Central research question:

Which animal has better ability to learn/memorize the taught information between the Hamster and the Guinea pig?

There will be 4 research questions:

- How much time do guinea pig and the hamster take to learn the given information? (The time that the guinea pig and hamster would be using to run the maze will be due to the average of other general researches that state the amount of time that both rodents should be given to be able to learn some new informations. The information needed for these animals to learn as "new information" would be the track design and pathways [the maze]). In this experiment, there would be certain time limits that the subjects should not exceed to complete the maze. The time that the subjects spent in the maze would be tracked in a data table and used to represent their respective

abilities to take in and process information.

- How to time the Guinea pig and the Hamster in their forgetting stage? (making sure that there is no biased or unfair timing) (In this stage, the specimens would be left alone with the same amount of time in a daily basis to forget the information that it had been taught. After this process, it would be going through the stage of training once again which would be part of the next process.)

- How much time does the Guinea pig and the Hamster take to recover the information taught? (This is the process where the subjects would be given some amount of time to recover the information that it had forgotten. To make the data accurate, there would be a specific time set up which is expected that both subjects would be able to recover the information that it has forgotten within those amount of time. However, time would be extended if they are not successful with the task taught previously.)

- How much time or how many time does it take for each of the specimens to run the maze without an error? (During this process, there would be a member counting the errors that the subject had done. This data would be graphed and compared to the time that it spent in the learning and recovering stage.)

The procedure would proceed to run in a cycle, this would be more than enough information for the report.

The last question lies on the:

- How accurate can these specimens complete the maze? (The errors collected from each practice in track will be used as one of the data information. This would be the last experiment held and would only have a certain amount of time for the two specimens to run. If there is an error, this error would also be recorded for the final

result.)

What is the topic of study related to that question?

The topic of study related to the previous question is how the animals learn and act. Besides this, it also shows the reward pathway of the animal from using treat to help animal remember the maze. In this case, classical conditioning will be implemented and studied using the subjects provided. Furthermore, treats would be arranged in a way that help lead or guide the subjects out of the maze during the first few times of training and there would be a huge treat at the end of the maze. This is to help the subjects adapt to the maze environment without having potential occurrence of stress, which would also develop thinking patterns that would be rewarded with treats. Since snacks would have an impact on how they behave, the potential for the specimens to finish up the maze as perfectly as they can would be boosted.

Why this is important research, what is its significance, and why should someone reading the proposal care about the outcomes of the proposed study?

This is an important research because it will help other researchers in the future. By doing this, it is possible to observe how the brain of both organisms function and how they react to variables. Besides this, the organisms in the same family will be compared and contrasted in order to find the difference. Aside from this, researchers might be able to compare the data that we have collected to human beings, due to the fact that all of them are mammals. Future researchers are able to use the results the report gained in order to compare it with one's own, conclude or discover something new about other related organisms. If datas that have been collected in both organisms are similar, scientists might be able to conclude that mammals learning development are fairly similar. On the other hand, if datas

turn out to be different, it would lead to a conclusion of which organisms might have different brain functions, therefore indicating that the need for the variety of methods used for different variables would be necessary potential results. The evaluation starting from the normal rodents experiments using those of the information as the guideline to see the important factors that would affect the rodents to learn in their best ability.

Background information:

People, as well as animals, have *selective attention,* this means that they choose which things are important and which things they can forget or pay the least attention to. However, before remembering even happens, the animal must notice a stimulus first. They then "encode" the information from their sensory memory (what you see, touch, taste, hear, smell) into their short term memory and then decides whether or not it is something that is important enough to store in the long term memory which could be anything that is necessary or vital information, such as where to find food, shelter, clothing, etc. Anything that is stored in the long term memory must be pulled out of the memory banks by what scientists call "Retrieval". But since the memory isn't a straight line where one leads to two and so on, and are more like an interconnected webs of associations, "Retrieval Cues" are needed to help remember. They're memories that are associated to the targeted memory and assists the animal in remembering the memory. Priming is what scientists call this non-conscious activation of associations between memories. In addition, the emotion and state of the organism will affect the type of associations they're priming. For example, if a hamster was trying to remember a maze, and a mark was made on a section of a maze before the hamster has to turn right, then the next time when the hamster sees the mark it will have to remember it has to turn right and if they were having a good time from the food, the next time they run a

maze with a positive mood, they're more likely to recall that running the maze successfully gives them food, a positive association.

The next part about the learning process is how memory loss happens. Which can be broken down into three ways; storage decay, memory interference (retrieval failure), and storage failure. Storage decay is when a memory had been left unused for a sufficient amount of time that it disappears. When an outside stimulus triggers an alteration of the memory stored, that is called memory interference. Storage failure defines when the memory wasn't encoded or stored. Hamsters and guinea pigs in this experiment would test these concepts. Storage decay, or forgetting due to length of time, and whether or not they experience storage failure, would be tested. If anything, the hamsters not storing anything in time is evidence that supports the fact that they can't remember in time. Then the "Priming" would also be tested on how the hamsters and guinea pigs remember from retrieval cues. (*Remembering and Forgetting- Crash Course)*

The research here is trying to gather evidence to tell which animal between a hamster and a guinea pig can learn, remember, and relearn (the learning process) more efficiently by making them run a maze. Understanding the learning process first requires understanding on how the specimen learns a piece of information(in this case how to run a maze), how long they're able to retain that information, and how do they relearn something that they have learned before but have forgotten. The questions here are, how do this animal learn? How does it forget? How does it remember again? The experiment would record the teaching time, the relearning time, and the forgetting time of both species and compare them with one another. That all begs the question, isn't this what all the scientists have been doing all these years? Studying the ways hamsters and guinea pigs run a maze? Now, the answer to both is

yes, but no one has ever done *both* of the animals simultaneously before. In this experiment, the two animals' learning processes will be tested by running a maze.

Literature Review

In order to evaluate which types of cages would suit the hamster most, researches have to be conducted. According to *A Renewed Look at Laboratory Rodent Housing and Management* (Gonder & Laber, 2007), there are various factors that would affect the physical and mental health of rodents. Factors such as; cage space had surprisingly affect the mortality rate of rodents. Many conducted researches had proven that rodents living in small spaces tend to survive more, increase their weights, and turns out to be less aggressive. Furthermore, the environmental enrichment also had an impact towards rodents. Rodents that were placed in cages with rigid shelter had demonstrated an increase in aggression and a decrease in weight, which were factors that determine stress. The journal includes various of results from different experiments. Some of which tested the rodents' behavior, brain development, stress levels, and others.

These experiments presented shows the exact same result that small cages are better for rodents, although various researches had proven that the belief of rodents inhabiting in small cages performed an outstanding result, was in fact inaccurate. In other research experiments, it had been proven that rodents performed better in large cages since they have more space to run around and relax. This results them to relieve against stress and gain happiness. On the contrary, this journal states facts that are fairly much in contrast with others, which is very doubtful to believe. Since the information given in the article could be biased or shows one sided success, the experiment which proves the doubtful result should have further experiments to find better and even more accurate results. From the information

stated above, it is critical to find cages that suit the subjects which would be used as an experimental subject. This is due to the fact that it might have an impact on its learning behavior which includes forgetting and recovering stage. If animals are not comfortable in the environment that they are placed in, it would be hard for them to concentrate on the tasks that they are given.

Two different studies discuss and analyze the behaviors and memory of mices and rats gaining results that vary. A journal named *Using the Morris Water Maze to Assess Spatial Learning and Memory in Weanling Mice* (Barnhart, Yang, & Lein, 2015) indicates that weaning mice develops spatial learning and memory (encoding, storage, and recovering of a memory), sex differences does not affect how they perform, and training enhances transcription of genes particularly in the synaptic plasticity (the capability of synapses to become stronger or weaker over time, responding to increases or decreases in their activity). The evidence given is that training results in a decrease in escape latency (the time it takes to find the platform) and spend more time on finding the path to target the quadrant. On another journal named *Assessing Spatial Learning and Memory in Rodents* (Vorhees & Williams, n.d.), they compare morris water maze (MWM) with radial-arm maze (RAM). There are two types of radial arm maze, one tests on trial-dependent memory while the other tests the combination of both working and spatial memory. It concludes that by using RAM, rats tend to gain both spatial and associative aspects which benefits them more than MWM. On the contrary, RAM takes more time in their process of learning compared to MWM.

To compare, both of these experiments tested the memory of subjects whether which method will be most effective for them. By this, their results show that the subjects are able to remember the information that is given to them. Data had been collected and these subjects

are observed various of time before scientists would be analyzing the results.

On the other hand, the result gain from both experiments are different. This might be due to one experiment using mices while the other uses rats. The development of how the subjects learn and reacted from different processes varies. The main focus on the first journal involves what types of learning do mices gain from MWM and the factors that is affected by it. However, the second journal focuses on which type of mazes benefits the rats more and how is it different. Furthermore, the first research consider sex as a factor to whether their learning process of males and females would be different while the other does not. Despite this fact, the first one tend to emphasizes on how the mices learn and develop its times and distance while the other concentrate on how much information is memorized by the rats.

To criticize, the first experiment is more accurate because it focuses on a particular experiment. It is able to analyze more information while the other one is only able to analyze the different learning stages. Although, the other experiment is also useful due to the fact that it shows the advantages and disadvantages of learning processes and future researchers are able to use this information and adapt it to their maze that they would be making.

Connecting this trials to future implications, if mices or guinea pigs were to be experimented, mazes would be a better option for them since there is only a small amount of time to conduct the experiment. Radial arm maze would take too long to be implemented on the subjects and would be hard to test the spatial and associative aspects on them. Furthermore, since researches have stated that training is the most essential part of a successful result, the main point that should be focused on future mazes would be this particular part. Moreover, if their activity affects their memory on the maze both species should be controlled in order to make the data as precise as it could become.

List of Materials:

1. Feature board

2. Velcro tape

3. Ruler

4. Scissor

5. Rubber

6. Cutter

7. Paper

8. Camera

9. Pencil

10. Markers

11. Guinea pig (male)

12. Hamster (male)

13. Computer

14. Phone (for timing)

15. Snack (for training)

16. Toys

17. Cage

Timeline

Date	Tasks
10th May 2017	-Prepare the materials -Start building the maze
11th - 14th May 2017	-Get the rodents and materials needed -Finish the maze -Create the data table
15th May 2017	-Bring the rodents to school - Start the experiment (day 1) -Collect the data needed
16th May 2017	-Experiment (day 2) -Collect the data needed
17th May 2017	-Experiment (day 3) -Collect the data needed
18th May 2017	-Experiment (day 4) -Collect the data needed
19th May 2017	-Experiment (day 5) -Collect the data needed -Collect all videos need to be edited -Bring the rodents back home
20th - 23th May 2017	-Analyze the data -Finish the papers
24th May 2017	-Finish the video
25th May 2017	-Turn in the project

Methodology:

The given specimens would be having a hard time upon learning about the maze, due to the fact that specimens would respond with becoming scared or becoming uneasy upon the given obstacle course that they will be placed in for the experiment. Which this is a natural response of the specimens due to their natural instinct of encountering unusual environments, people, pressure or task that they've never been experienced in, but before putting the

specimens through training, we must first allow them to get used to the trainers then proceeding towards recognizing that the maze is not a threat for them. As for these two conditions "Counterconditioning", as well as "Habituating" are needed to ease the specimens to adapt towards possible obstacles therefore, inhibit the ability for them to be challenged upon the maze and tricks that will be taught in the future.

There will be 3 learning evaluation for the rodents, where each of the rodents would be given with the same starting time, the same starting point which would develop their progresses and the speed of each rodents' intelligence:

Evaluation 1 (Learning)

- This part of the evaluation stage is where the specimens would be taught with new informations. At first, food would be provided along the correct path of the maze in order for them to learn and remember the path. Then, before they would start running, a certain time would be set as a standard in order to determine their ability to learn. After that, subjects would be running in the maze and both species would be timed and number of errors would be counted (when they run to a wrong path). These data would be collected daily during the experiment.

Evaluation 2 (Forgetting)

- In this stage of the experiment, the specimens will be placed in their own housing with an exact schedule to prevent the confusion for the training sessions, each of the specimen would be trained with 2 specific people, to prevent time loss when one of the given trainers are absent.

Evaluation 3 (Recovering)

- This part of the evaluation stage is where the specimens would be given a certain amount of time to recover their learnt information. They will be placed at the same position and the same part of the maze, however, food won't be provided to guide them anymore except the food at the finish line. Therefore, the rodents will have to recover the path by themselves.The rodents will be timed from the start to the end while, at the same time, the error will also be counted. After this stage has been done, they will proceed back to Evaluation 1.

Maze Design and Justification:

The maze was designed to facilitate as many variations of mazes as possible with the least materials needed, which means the maze is capable of redesigning the pathway without the need to rebuild the whole maze again. Since the time is fixed, the variable maze would allow us to retest the rodents' learning abilities by having the rodents run a different maze designs, therefore the data collected would be valid, although issues that we might face with this factor is that the time given for this experiment might run short, which we may not be able to try out multiple maze trainings. The walls would be made differently for 2 specimens, the guinea pigs wall would be about 10-12 cm, whilst the hamster would be about 7-8 cm due to the fact that hamsters will have trouble sniffing out for the treat if the guinea pig becomes larger, therefore enabling the ability for the hamsters to have the same ratio of wall height would be the fairest option.
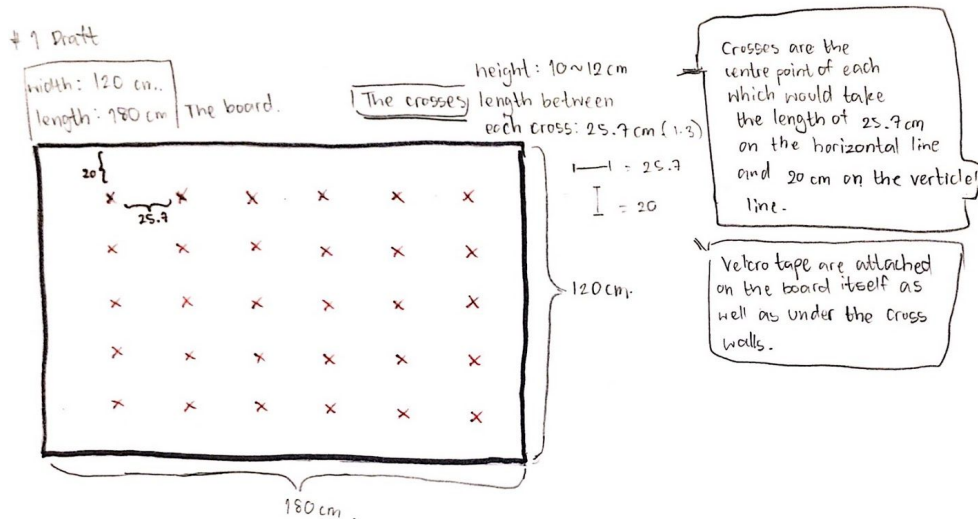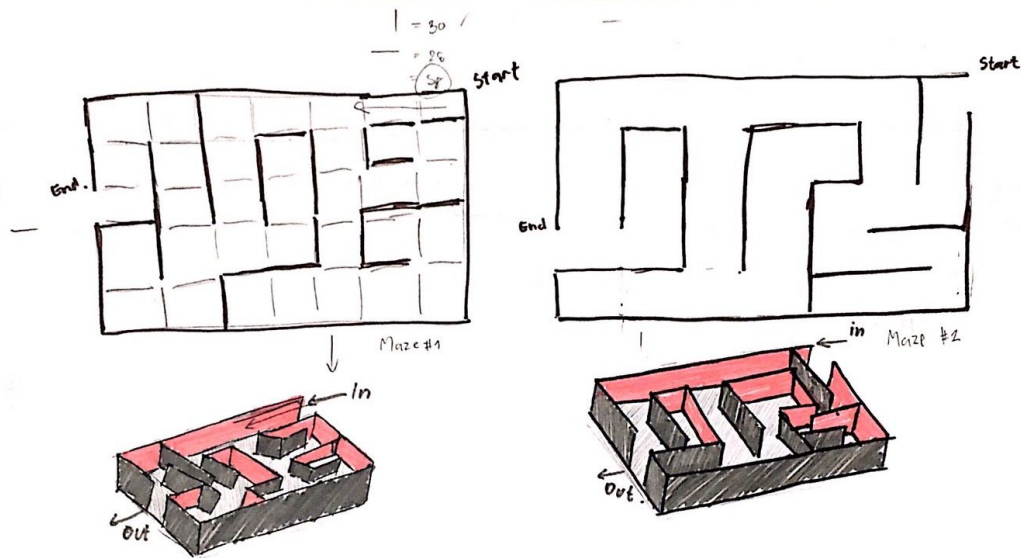
Methodology Literature Review

According to the research of *Spatial Memory Performances of Aged Rat in the Water Maze* (Drapeau et al., 2003), the study that predicts levels of hippocampal neurogenesis. The experiment was performed on twenty-three months old rats. These subjects were tested in a morris water maze, with a diameter of 180 centimeters and 60 centimeters high made opaque by adding milk powder. Before experiments were conducted, they were trained daily to find platforms which will lead them out. During their actual tests, they are tracked based on the

time and the distance that they traveled to find the platform using a computerized tracking system. To collect data, BrdUrd (Sigma) was injected to rats which show that learning increases the survival rate of newly born cells during their training. However, the entire procedure of the water maze has no impact on the subjects.

In another research called *Learning and memory impairments in a neuroendocrine mice model of anxiety/ depression* (Darcet et al., 2014), mices were placed in a morris water maze and a barnes maze. Similarly to the previous experiment, mice were to find the platforms. However, the timing scale is different. If rats are not able to find the platform within 60 seconds, it would be guided to the platform and the time recorded would be 60 seconds. On the barnes maze, the mices are placed in a clear gray circular platform with 20 equally spaced holes and one target box would be placed underneath a particular hole. It would not see any holes due to the fact that it would be dark and the mice would have to use spatial extra cues. They would have 180 seconds and if they are not able to escape, they would be guided and the time recorded would still be 180 seconds. These trails were recorded by camera and analyzed by ANY-maze software. Primary latency, latency to escape, primary errors, and total errors were used to score these mices. This experiment proves that mice placed on an anxiety/depressed situation is caused by the excess of glucocorticoids.

According to a research mentioned previously in the literature review, the Radial arm maze is like a wheel which has arms coming off from it. The rats would be placed and the center and foods would be placed on the arms. By this, animals would have to be starved before they would be place into the maze. Since there are two types, each of them have different procedures. In the first type, the food would be placed in all arms and rats would be able to choose which arm they would be going to. From this, if the rats were entering the

same one that they had entered before that would count as an error. However, the problems to this way are that rat are able to go in a particular way or pattern from going only on left or right side. As a result, this methodology would not help test the memory of rats, due to the fact that there are other efficient ways to do so. Therefore, for the second type number of arms would include food and some would not include food. The rats would be placed in this maze several time and tested if they are able to remember which ones include food and which one doesn't. This helps prove whether they have spatial memory.

This experiment here shows the same result as other researches where learning has proven to increase new cells that would grow in organisms. On the other hand, running an experiment that shows a null result is not very efficient. Before conducting any experiments on animals, researchers should collect data from previous labs. By this, they are wasting their time and harming organisms. As a result, time and distance would be collected to run the maze. Learning stages would be highly emphasized with the recovering stage being the main part that would be use to track the mices.

As known in the researches above, there were 3 given methods used for the same research purpose, which is to find whether there were any intellectual development in specified time with the given complexity of the distance towards the given goal. With 2 of the experiments, the factor that that excites the rodents upon all the experiments were "the way out" which acts as a trigger that makes the response of how active, how much concentration they put on to achieve the rewards. *Learning and memory impairments in a neuroendocrine mice model of anxiety/ depression* as for both Morris' and Barnes' methods were used. With Morris' and Barnes' method, they were both given the puzzle to 'find their way out' rather than just achieving the prize.

To each of the experiments held, there were several differences that were distinct; age, species, methods, technology used for the experiments. Starting from the age mentioned on "*Spatial Memory Performances of Aged Rat in the Water Maze*" experiment, they specified their age limit for the specimens, which in "*Learning and memory impairments in a neuroendocrine mice model of anxiety/ depression*" and in *Radial arms* experiment none were mentioned. With the species difference, "*Spatial Memory Performances of Aged Rat in the Water Maze*" with, they were both focusing on the rat's intellectual development whilst "*Learning and memory impairments in a neuroendocrine mice model of anxiety/ depression*" focuses on the mice's anxiety/depression which would affect the intellectual development as well as the performance of the mice. Methods that were used comprehend the performance of the rodents there were 3 distinct experiments taken place: *Morris' water maze*, *Barnes' maze* and *Radial arms*. Which *Morris' water maze* clearly states that the importance of water is needed to make it work with the tracking computerized system, as for *Barnes' maze* the main focus lies on a plate like form that the mice must find holes underneath them, which this is tracked by the ANY-maze software programme. Lastly the Radial arm method, focuses only on how desperate they will be to get the 'reward' which the concept of the method is to starve the creature instead of giving time for the creature to figure it out.

Both experiments did well in the fact that they didn't conclude anything that the scientific community didn't prove already through legitimately proven research. The methodology used is designed with an eye towards minimizing errors caused by humans such as computerizing measurements, this should result in accurate data. However, experiment #2 did say that when the rodents don't finish in time, they record it as 60, which would promptly throw the average off. Another one of experiment #2's mistakes is that they still went on with the experiment knowing fully that running mazes such as these makes the mice feel stressed.

That is just plain unethical. Nobody should have the right to do that to any creature or anyone, it's just not right to stress an organism "for science", no cause would justify putting animals to such stress. Experiment #1 didn't really do any better, they did what was already well known, so it didn't actually become anything new. Since they're using live animals, they should consider that doing an experiment that doesn't prove anything harms animals for no purpose at all, and so they should find background information instead of experimenting the next time. Overall, these two experiments had the same undoing: they unnecessarily inflict animals with uncomfort be it physical or mental, and it was a thing that could have been avoided.

Specifically related methods that were used and found as a similarity towards the methods used for this experiment, which the ideal is to use 'rewards' as similar as the Radial arms, but allowing the rodents to 'find their own way' to get around the maze, although the technological adaptation were not possible for this experiment, therefore the need to substitute the ANY-maze software and computer tracking system with records based on writing manually, which is needed to evaluate upon detailed information.

Ethical/ Humane Considerations:

The materials that will be used to make the maze are composed of feature board and the velcro tape. These materials were chosen because they are not toxic towards the rodents, which they would likely eat and bite everything, as well as the fact that feature board is easy to clean. After extraneous research, it is determined that these materials would be safe if the animals were to ingest them. On the other hand, the rodents will be very stressful when they're moved into a new environment or change their foods. Based on the research done, the

rodents will adjust to their new "home" after a few days alone, although the toys would provide additional help in accomplishing this. Smell and voice would help the rodent familiarize itself with humans who are going to be experimenting on them.

Data Collections and Limitations:

As they're 3 different phases of the experiment, there are different data that needs to be collected for each phase.

Phase 1: Forgetting stage

After the subjects are trained and are well prepared, it will be left alone for some period of time so that it would forget the information that it was taught and trained. There is only 1 data needed in this stage which is the forgetting time of both rodents which will be controlled every day of the experiment.

Phase 2: Recovery stage

Continuing from the previous stage,when the forgetting time is over, the subjects would now be placed into the same mazes and made to run it, this is in order to observe how much information have been loss with time. However, there is a certain time that it is expected to complete the maze.

Phase 3: "Perfect running" stage

Since both subjects are from different species, the rates at which they run would differ. if that is factored in, their must be something else other than time used to calculate the results. Therefore, the errors made by the subjects would be another factor that would help consider. However, time could be calculated in order to determine its intelligence. Firstly, the subjects would be running in a straight line that contains the same length and would be timed.

Secondly, a new unit would be created in order to compare the time that both species run. In order to calculate this, the time that it takes to complete this run would be divided by the length of the path. From this, the rate that each species run would be calculated. As a result, this unit could be used in comparing the time that each species run during the mazes.

Error tracking stage

Using these 3 stages of evaluation as well as error, the species' intelligence would be determined in a way that does not harm them. For instance, the member counting the errors made by the hamsters or guinea pig miscounted.

Conclusion:

There are 3 stages of our experiment which are learning, forgetting, and recovering. By collecting the data of each guinea pig and hamster from each stages and compare them at the end, its intelligence could be determined. The factors that would be used to determine their intelligence are time, errors, and recovery. These factors will be graphed respectively to see which subject has made more learning progress, standard deviation and trend lines would be calculated from the given graph. Moreover, these three factors (the initial time, recovering time, and errors) might be combined and averaged out in order to make the data more accurate and more reliable.

References

C. (2014, May 12). Retrieved May 08, 2017, from https://www.youtube.com/watch?v=HVWbrNls-Kw

Barnhart, C. D., Yang, D., & Lein, P. J. (n.d.). *Using the Morris Water Maze to Assess Spatial Learning and Memory in Weanling Mice.* Retrieved May 08, 2017, from http://journals.plos.org/plosone/article?id=10.1371%2Fjournal.pone.0124521

Darcet, F., Mendez-David, I., Tritschler, L., Gardier, A. M., Guilloux, J., & David, D. J. (2014, April 03). *Learning and memory impairments in a neuroendocrine mouse model of anxiety/depression.* Retrieved May 08, 2017, from http://journal.frontiersin.org/article/10.3389/fnbeh.2014.00136/full

Drapeau, E., Mayo, W., Aurousseau, C., Moal, M. L., Piazza, P., & Abrous, D. N. (2003). *Spatial memory performances of aged rats in the water maze predict levels of hippocampal neurogenesis.* Proceedings of the National Academy of Sciences, 100(24), 14385-14390. doi:10.1073/pnas.2334169100

Gonder, J. C., & Laber, K. (2007, January 01). *A Renewed Look at Laboratory Rodent Housing and Management.* Retrieved May 08, 2017, from https://academic.oup.com/ilarjournal/article/48/1/29/690040/A-Renewed-Look-at-Laboratory-Rodent-Housing-and?searchresult=1

Vorhees, C. V., & Williams, M. T. (2014). *Assessing Spatial Learning and Memory in Rodents.* Retrieved May 08, 2017, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4240437/