

RESEARCH ARTICLE

The Effect of Mesenchymal Stem Cell Wharton Jelly on Alzheimer's Rat with Y-Maze Test Method

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Abstract

Objective: This study aims to examine the y-maze test in alzheimer's rat after addition mesenchymal stem cell wharton jelly. **Methods:** This study used the y-maze test method to test the cognitive test of rat with the Y arm model. The treatment group consisted of a control, $AlCl_3$, MSC-WJ and a combination of $AlCl_3$ and MSC-WJ. The results obtained were analyzed in the form of a percentage. **Results:** The results of this study showed that the y-maze test showed that MSC-WJ had a significant effect compared to the $AlCl_3$ control group in the alzheimer's rat, namely 51.33% and 12.31%. **Conclusion:** In this study it was concluded that that MSC-WJ has a significant effect on alzheimer's rat with y maze test.

Keywords: $AlCl_3$; Alzheimer's; Mesenchymal Stem Cell Wharton Jelly

INTRODUCTION

The y-maze test was used to test the behavior of rat in exploring new environments. The mouse will usually choose to investigate the maze's new arm rather than returning to the previously visited arm. According to Zhou *et al.*, (2015), MSC-WJ transplantation can improve the recovery of learning and memory functions.¹ According to Ru and Liu (2018), learning and memory processes are very important for an organism's ability to adapt to environmental changes.²

In humans, learning and memory are usually expressed through spoken or written language, whereas animal cognitive functions can only be expressed through behavior. The y maze test is a useful tool for detecting the learning and memory abilities of animals because of its simple structure and easy operation.² As science develops, the y-maze test is an accurate test for cognitive testing in animal models. Therefore, this study aims to examine the y-maze test in alzheimer's rat after giving mesenchymal stem cell wharton jelly.

METHODE

Types of research

This research is an experimental research. The experimental animals used were 24 *Rattus novergicus* aged 2 months with body weights ranging from 200-300 grams.

Thawing Mesenchymal Stem Cells Wharton Jelly (MSC-WJ)

A complete medium consisting of alpha Modified Eagle Medium (α -MEM) high glucose, penicillin / streptomycin and Fetal Bovine Serum (FBS) was prepared and put into a 15 ml falcon tube with a ratio of 1:9. Then, cells from the liquid tank were taken and put into a water bath for 10 seconds.

Next, the cells in the cryotube were transferred to a 15 ml falcon tube which already contained the complete medium. Centrifugation was carried out at a speed of 2000 rpm for 10 minutes at room temperature. After centrifugation is complete, the supernatant is removed and the remaining pellets are added with 4 ml of complete medium (depending on the size of the flask to be used). Then, homogenized and transferred into a 25 cm² flask and checked under an inverted microscope. Cells were incubated in a CO₂ incubator at 37 °C and 5% air pressure for 3 days.

Subculture of MSC-WJ

Subculture can be done in the following ways. Briefly, discard the culture media and rinse the cell layers with PBS. Then the PBS was removed and a 0.25% trypsin EDTA solution was added to a 1 ml flask. Incubate for 10-15 minutes in a CO₂ incubator. Next, the cells were observed under an inverted microscope until the cell layers detached from the base of the flask. Cells that are difficult to release can be incubated again in a CO₂ incubator at 37 °C for 5 minutes. The next step is to add 4 ml of complete medium and aspirate cells with a transfer pipette and carry out centrifugation. Substitution of the medium was carried out 3- 4 days until the cells were ready for use in this study, namely passage 3.

Injection of MSC-WJ

All animal groups were anesthetized using ketamine (50 mg / kg) and xylazin (15 mg / kg) intraperitoneally.³ Rat were injected with 1 x 10⁶ cell / mouse stem cells in 300 ul of complete medium⁴ by i.p.⁵. The control group was injected with the same volume of PBS for each mouse. Then the rat brain tissue was harvested one month

after the injection of stem cells and put into a film bottle that had been prepared.

Y-Maze Test

The y-maze measures the spatial workings and memory of the experimental animals by harnessing the rat natural exploration instincts. The y-maze consists of three arms of equal length and interconnected to form an angle of 120 °C. This test was used to measure working memory in rat by counting the amount of movement of the rat when the animal visited all three arms without going to the same arm twice in succession. After a 10-15 minutes break, the y-maze is cleaned. If the memory of the rat was still normal, the rat had to remember the first two arms and be more curious about the third. This method can be used to compare the exploratory behavior of the control with experimental animals to determine the impaired working memory. Motor activity will be recorded as the number of arms visited during the duration of the test.⁶

RESULT AND DISCUSSION

The results showed that the y-maze test showed that MSC-WJ had a significant effect compared to the AlCl₃ control group in the Alzheimer's mouse model. In the AlCl₃ group, the percentage was 12.31% while the combination was 51.33%.

Table 1. Y-Maze Test

Group	Mean (%)
Control	77.14
AlCl ₃	12.31
MSC-WJ	67.97
AlCl ₃ +MSC-WJ	51.33

The low percentage in the AlCl₃ group illustrates a decrease in memory in the rats brain, while the percentage of the

combination of AlCl₃ and MSC illustrates the improvements made by stem cells to disturbed neurons in the rat brain.

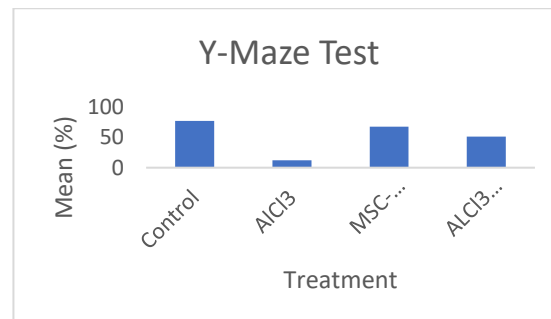


Figure 1. Percentage of Y-Maze Test on Alzheimer's Disease

According to Tong, Fong and Huang (2015), the cognitive decline that underlies Alzheimer's disease is the result of the loss of neuronal processes due to various factors. With the advent of stem cell technology and the ability to convert these cells into various types of central nervous system neurons and glial cells, some success in stem cell therapy has been reported in animal models of alzheimer's disease.⁷

The percentage of the y-maze test can be seen in Figure 1 above. MSC was found to increase autophagy and exert a neuroprotective effect by modulating Aβ clearance in an alzheimer's disease rat model. Shin, Park and Kim (2014), showed that the damaged alzheimer's disease brain could potentially be repaired using MSCs through the modulation of the autophagy pathway.⁸

According to Yao *et al.*, (2020), states that in the brains of alzheimer's disease patients and animal models, excessive activation of microglia and astrocytes can cause an inflammatory response and lead to nerve damage.⁹ In this study, the y-maze test described the cognitive ability of the rat to recognize what they had passed

during the test period. In normal mouse conditions, mice will recognize something they have passed because mice have strong memories.

In the research of Wahl *et al.*, (2017), the y-maze test reflects several aspects of memory including episodic memory, recognition memory, semantic memory, spatial memory and emotional memory. The types of grade memory that deteriorate during alzheimer's disease are episodic and semantic memory. This memory loss often coincides with lapses in spatial learning and navigation and memory.¹⁰Spatial learning is a process that involves many cognitive and perceptual processes and refers to the attainment and maintenance of a specific trajectory from

one place (or object) to another and is learned over time. Therefore, the assessment of spatial memory is very important in rodent assessment.

CONCLUSION

In this study it was concluded that that MSC-WJ has a significant effect on alzheimer's rat with y maze test.

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