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Hippocampus Cell Disorders and Neurosensory Tests in Mice (*Mus musculus*) Due to Induction of Excess Sodium Chloride

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Abstract

Salt or sodium chloride (NaCl) is an additive to give food a salty taste. The use of salt in everyday life is difficult to avoid. Salt has both good and bad effects on the body. The electrolyte content of salt can help launch metabolism in the body, whereas if the body contains too much salt it will cause heart attacks and hypertension. The dose of salt consumption that has been set by the government is 5g/day. This study was conducted to examine the effect of consuming excessive sodium chloride (NaCl) on the ability to smell and to what extent it damages cells in the hippocampus of mice (Mus musculus). The method of this study was CRD (completely randomized design) with 6 replications, 1 group control and 3 repetitions induced by sodium chloride (NaCl) is 260mg/gBB (P1), 520mg/gBB (P2), and 780mg/gBB (P3). The parameters of this study are neurosensory coordination in the form of olfactory response of mice (Mus musculus) to ammonia and cell disruption in the hippocampus (DG & CA) which were observed by histological preparations of Hematoxylin eosin (HE) staining. The results of this study indicate the presence of olfactory disorders in mice (Mus musculus) and cell death in the hippocampus also increased due to excessive sodium chloride (NaCl) induction. The more salt is consumed in daily life, it will disrupt the cells in the hippocampus.

Keywords : Sodium Chloride;Hippocampus;Mice (Mus musculus)

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1. Introduction

Salt or Sodium chloride (NaCl) is an additive that is often used to give food a salty taste. Salt has bacteriocid and bacteriostatic properties, so salt can be used as a food preservative. Consumption of salt has a good impact on the body because the electrolyte content plays an important role in metabolic processes and is useful for helping regulate blood pressure, if the dose used does not exceed the predetermined dose [13]. The dose of salt used by the Indonesian Ministry of Health in 2009 is 5g/day or equivalent to 1teaspoon/day [9]. Research by [2] regarding a high-salt diet can damage memory-related synaptic plasticity through increased oxidative stress and suppression of synaptic protein expression. It is known that rats who underwent a high-salt diet for 4 weeks (27.5% NaCl) and 7 weeks (25.3% NaCl) showed impaired Short Term Memory in the task of recognizing objects and could cause serious disturbances in Long Term Memory. Mechanically, a high-salt diet causes a decrease in the amount of synaptic protein levels so that it can inhibit the potentiation of long-term memory, in the hippocampus.

Research by [3], regarding hypertension induced by high salt causes rats to experience cognitive impairment because a high salt diet interferes with spatial memory and affects the decrease in intracellular calcium in the hippocampus. The hippocampus is the repository for short-term and long-term memory, and is responsible for cognitive abilities. Damage to the hippocampus can lead to impaired learning and impaired reception of new information [4].

According to [5], the hippocampus neuronal morphology can change due to several factors, namely stress and chemical content in the hippocampus. Morphological changes in the hippocampus can cause cell death, and disrupt the hippocampal intracellular membrane causing impaired cognitive abilities. Cognitive ability is the process of thinking and acting in making decision, problem solving, and the ability to reduce brain-centeredness [6]. To find out cognitive impairment is to do a smell test. In the olfactory system, odor receptors will be allocated to olfactory sensory neurons located in a small area at the top of the nose, then olfactory receptors are transmitted in the glomerulus, in glomerulus the nerve endings of the receptors stimulate the mitral cells to transmit signals to the brain area. In normal brain conditions, the response is very easy to give when smelling something that smells or interferes with the sense of smell [10]. One of the chemical compounds that have a sharp odor that can test the ability to smell is ammonia. Based on these references, it is suspected that excessive salt consumption can damage brain function, especially in the hippocampus which results in impaired cognitive ability, because it is important to know the developmental disorders of hippocampal cells and cognitive ability disorders of mice (*Mus musculus*) due to excessive salt induction, so this study was carried out.

2. Materials and Methods

This study uses the RAL method. The test animals in this study were male Swiss webster (*Mus musculus*) mice aged 21 days with an average body weight of 25 grams. Animals were acclimatized for 1 week. The test material used in the form of salt/sodium chloride (NaCl) used in this research is salt with the composition: Content of Potassium Iodate (KIO3) min. 30 ppm, and the content of Sodium Chloride (NaCl) is 97.70%.

3. Results and Discussion

3.1 Neurosensory Test (Ammonia Smell) in Mice (*Mus musculus*)

The results of the study on the effect of sodium chloride (NaCl) on the ability to smell ammonia in mice (*Mus musculus*) can be seen in Table 1.

Table 1. Ammonia Smelling Behavior Test in Mice (*Mus musculus*)

Group	Mice (*Mus musculus*) Responses		
	Dodge	Neutral	Approaching
K	100%	0%	0%
P1	67%	33%	0%
P2	0%	73%	27%
P3	0%	60%	40%

From the test results of the ammonia smelling behavior in the control group, it was found that as many as 100% of mice (*Mus musculus*) gave an avoidant response to the smell of ammonia. In the P1 group, it was found that as many as 67% of mice (*Mus musculus*) gave an avoidant response to the smell of ammonia and as many as 33% of mice (*Mus musculus*) gave a neutral response to the smell of ammonia. In the P2 group, it was found that as many as 73% of mice (*Mus musculus*) gave a neutral response to the smell of ammonia and as many as 27% of mice (*Mus musculus*) gave a close response to the smell of ammonia. In the P3 group, it was found that 60% of mice (*Mus musculus*) gave a neutral response to the smell of ammonia and as many as 40% of mice (*Mus musculus*) gave a close response to the smell of ammonia.

Ammonia has a strong pungent odor that causes mice (*Mus musculus*) to become dizzy and stay away from it. In mice *(Mus musculus*) that give a neutral and close response to the smell of ammonia, it is possible that there has been a disturbance in the nerves due to the administration of excess sodium chloride (NaCl) so that it cannot interpret the smell well.

Behavior is a functional indicator of the sensory and motor integrative processes of the peripheral nervous system. Behavioral disorder is a symptom that can be seen, due to a disturbance in the central nervous system which functions as a center for processing behavior, memory and learning processes [1]. Consumption of excess sodium chloride (NaCl) can damage the central nervous system and pose a permanent risk and interfere with intelligence [3].

Olfactory disorders in mice (*Mus musculus*) are triggered by decreased cortical function and decreased concentrations of acetylcholine, serotonin, norepinephrine in the striatum, serotonin in the cortex, norepinephrine and dopamine in the base of the hippocampus, and norepinephrine in the ventral hippocampus. This decrease in concentration causes a decrease in the ability of the nervous system to digest and

transmit impulses between neurons [7]. Decreased concentration of neurotransmitters in the brain can cause behavioral deviations, while learning and remembering behavior and responding are important because they have adaptive value for an organism [11].

3.2 Observation of Mice (*Mus musculus*) Hippocampal Cells

The results of the study on the effect of sodium chloride on the hippocampal cell disorders of mice (*Mus musculus*) can be seen in Figure 1. Different levels of cell death occurred in the control group (K), Treatment 1 (P1) 260mg/gBB, Treatment 2 (P2) 520mg/ gBB, and Treatment 3 (P3) 780mg/gBB to the hippocampus in the *Dentate Gyrus* (DG) and in the *Cornu Ammonis* (CA).

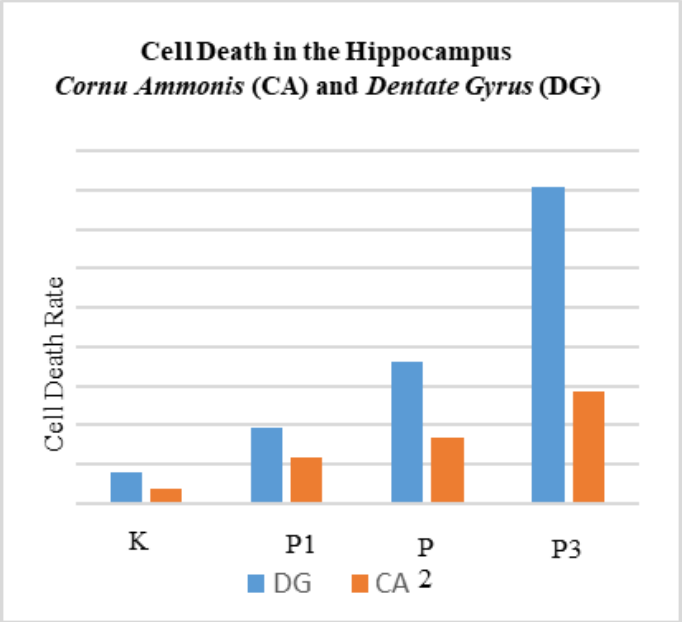

Figure 1. Total Cell Death in *Cornu Ammonis* (CA) and *Dentate Gyrus* (DG).

The cell death rate of each treatment group had a significant difference. From each treatment group which was divided into several groups, namely the control group (K), P1, P2 and P3, it can be seen that the control group was the group that experienced the lowest cell death and P3 was the group that experienced the highest cell death rate, both in the *Dendate Gyrus* (DG) and *Cornu Ammonis* (CA). In the death of nerve cells in the hippocampus (CA&DG), the administration of excess sodium chloride (NaCl) triggers hypertension and disrupts the sodium balance in the body causing accumulation in the arteries so that blood cannot flow normally.

The occurrence of blockage of blood vessels causes a lack of oxygen to the body's organs, including the brain. This can disrupt the nervous system and muscles due to loss of body fluid balance and cause crenation in brain cells, especially the hippocampus. Cells that undergo crenation experience shrinkage due to lack of cells which lose function and experience damage and even death [8].

Cell damage or disruption to cells occurs in three stages, first the clumping of chromatin or pinoxys occurs, after pinoxis occurs the chromatin in the cell nucleus will dissolve and this stage is called karyolysis, then the process of splitting the cell nucleus is called karyorexia [12]. Cell damage that cannot be repaired will cause cell death, cell death is divided into two, namely apoptosis and necrosis. Apoptosis is a cell death event that can be said as cell death that has been programmed by the body because these cells will die by themselves and be replaced by new cells. In the body there are many different cells with different lifespans, some from a few days to 80 years or more. Necrosis is an event of cell death caused by the presence of toxic substances [1].

4. Conclusion

Based on the results of the study, it can be seen that excessive salt consumption can cause neurosensory disturbances and increase cell death in the hippocampus.

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