

Effectiveness of Bran Tea Water Extract on Spermatozoa Quality of Mice (*Mus Musculus*)

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ABSTRACT

Bran is a by-product of rice milling that has nutritional content for health due to the presence of bioactive compounds. In addition to Bran, *Piper caninum* is also rich in bioactive compounds. These compounds have antioxidant activity that can counteract exogenous free radicals. If exogenous free radicals enter the body, it will damage the metabolic system including the process of sperm formation. The number of bioactive compounds contained in Bran and forest betel makes this combination suitable as a beverage that can be consumed. This study aims to test the effectiveness of bran tea extract on the quality of spermatozoa mice (*Mus musculus* L.) adults. The experimental design used was a complete randomized design with 4 treatments, namely K (without tea bran extract), P1 (tea bran extract 350 mg/KgBW), P2 (tea bran extract 700 mg/KgBB), and P3 (tea bran extract 1050 mg/KgBW), each treatment consisted of six mice as replicates. The administration of bran tea extract is carried out orally once a day with a volume of 0.5 mL for 35 days. On the 36th day, mice were sacrificed by anesthesia and then performed surgery for spermatozoa collection. Analysis of the quality of spermatozoa is carried out by making a suspension of spermatozoa from the Cauda epididymis. The parameters observed include spermatozoa membrane integrity, motility, viability, morphology and concentration of spermatozoa. The Data obtained were then analyzed statistically using One Way Anova and further tested with DMRT. The results showed that the administration of aqueous extract of bran tea doses of 750 mg/KgBW and 1050 mg/KgBW in mice significantly ($P < 0.05$) improved the quality of spermatozoa.

1. Introduction

Living beings, including humans, have the ability to reproduce. Reproduction is the process by which organisms reproduce themselves which aims to maintain the lineage of their species (Sunaryanto, 2012). The process of reproduction consists of two types, namely asexual and sexual reproduction. Asexual reproduction is reproduction that involves only one individual without the process of exchanging genetic material with other individuals sexually, while sexual reproduction is reproduction that involves combining genetic information from two individuals of different sexes (nature, 2015). Sexual reproduction occurs when a female egg cell meets a male sperm cell, and then decays to later form a zygote (Sinaga et al., 2017).

One of the largest production of food sources in Indonesia, namely rice plants, is a rice-producing plant that is the staple food of the Indonesian people and one source of carbohydrates for most of the world's population (Masdar, 2005). Rice production in Indonesia has increased, in 2020 rice production amounted to 54.65 million tons of dry milled grain (DMG), producing more than 50 thousand tons of DMG or 0.08% compared to 2019 which amounted to 54.60 million tons of GKG (Faizah et al., 2020). Bran is a layer located between the rice grain and the red rice skin (Bran), Bran is a by-product of the rice milling process or creamy grain (Chen et al., 2012).

Bran (rice bran) is good for overcoming diabetes, high cholesterol, high blood pressure, and various other conditions. The content of substances in Bran for health benefits is very diverse such as carbohydrates, protein, fiber (dietary fiber and crude fiber), calories, fat, fatty acids, cellulose, minerals, starch, and vitamins B1, B2, B3, B5, B6 and vitamin E (Muntana and Prasong, 2010). *Piper. caninum* is a creeping plant, with strong and slender stems, leaves are always green, mostly found in Lowlands in primary and mixed forests (Hashim et al., 2019). Research conducted by Suriani (2018) also explained that forest betel nut contains saponin compounds, flavonoids, and polyphenols.

Research on ethanol extraction of 96% brown rice bran by Widarta et al. (2013) showed, that brown rice bran has a fairly good antioxidant activity that reached 62.41 %. Research on antioxidant activity in *Piper caninum* by Nadri et al., (2015) showed positive results in the form of obtaining phenolic compounds and the capture of free radicals. Suyasa et al., (2022) also explained ethanol extract of *P. caninum* at doses of 500mg / KgBW and 1000 mg/KgBW can increase the motility of mice spermatozoa. Until now, studies on the effect of bran tea extract on the quality of spermatozoa have not been conducted. Therefore, it is necessary to conduct research on the effect of bran tea extract on the quality of mice spermatozoa.

2. Methodology

2.1 Time and place of study

This research was carried out from January 2023 to February 2023. Forest betel plant obtained from Tabanan, Bali. Treatment of the experimental mice is carried out at the experimental animal maintenance room of Biology Study Program, Udayana University. Surgery was carried out at the Laboratory of Animal Structure and Development in the Biology Study Program, Udayana University, Bali, Indonesia.

2.2 Tools and materials

The materials that have been applied to this research were bran tea extract, mice (*M. musculus*), 0.9% NaCl, hypoosmotic solution, aquadest, 1% and 0.5% eosin, tissue, filter paper and husks. The tools used were microscopes, optilab, surgical instruments (scissors, knives, tweezers, needles), glass objects, glass slides, trays, gastric tubes, a rectangular cage (plastic tub), and drinking bottles for mice.

2.3 Methods

Research design

Complete randomized design (CRD) with four treatments and each treatment has six repetitions. The treatment included K (without water extract of bran tea), P1 (water extract of bran tea 350 mg/KgBW), P2 (water extract of bran tea 700 mg/KgBW), and P3 (water extract of bran tea 1050 mg/KgBW).

Measurement of bran tea water extract dosage

In this research, the dosage of the extract was made refers to research by Olsen et al., (2008):

Average body weight of mice = 30g = 0,030kg

- Dose of 350 mg/kgBW

350mg X 0,030 kg X 6 mice

= 63mg

63mg will be dissolved with 3mL of distilled water

- Dose of 700 mg/kgBW

700mg X 0,030 kg X 6 mice

= 126mg

126mg will be dissolved with 3mL of distilled water

- Dose of 1050 mg/kgBW

1050mg X 0,030 kg X 6 mice

= 189mg

189mg will be dissolved with 3mL of distilled water

Bran tea water extract application on mice

Bran tea water extract that has been dissolved with water is given orally with a volume of 0.5 mL (the volume does not exceed the intragastric volume) once a day for 35 days using a gastric tube. The dose of the extract given was 300 mg/KgBW for P1 treatment, 750 mg/kgBW for P2 treatment and 1050 mg/KgBW for P3 treatment. On day 36, the test animals were sacrificed under anesthesia and then surgery was carried out to collect spermatozoa.

Integrity of the cell membrane of spermatozoa

A suspension of spermatozoa is introduced into 1 mL of a hypoosmotic solution (0.31 g of sodium citrate and 0.565 g of fructose are dissolved in 50 mL of aquades), then incubated at a temperature of 37°C for 30 minutes. Then dripped on the glass object and then made observations using a light microscope with a magnification of 400 times.

Motility of spermatozoa

A suspension of spermatozoa in a 0.9% NaCl solution is pipetted and placed on a glass object, then observed under a light microscope with a magnification of 100 times against 100 spermatozoa. Calculations were carried out between motile and non-motile spermatozoa from 100 spermatozoa and the results were converted into percent form.

Viability of spermatozoa

Observation of spermatozoa viability was conducted by spermatozoa smear preparation method using 0.5% eosin staining. One drop of spermatozoa suspension is dripped on a glass object, then one drop of eosin is added and a smear is made for drying. Observation of the smear preparation is carried out under a light microscope with a magnification of 400x against 100 spermatozoa. Each test animal is counted three times, the result is then averaged from these three calculations. Each observation is expressed as a percentage.

Morfology of spermatozoa.

Observation of spermatozoa morphology was carried out by the method of spermatozoa smear/smear preparation using 1% eosin staining. One drop of spermatozoa suspension is dripped on a glass object, then one drop of eosin is added And then a smear is made and dried. Morphological observation is emphasized on the deformity or abnormality of spermatozoa.

Concentration of spermatozoa.

The concentration of spermatozoa is calculated using a Hemacytometer. First, the testes are placed in a Petri dish containing 0.9% NaCl in the amount of 2 mL, then the Cauda part of the epididymis is taken and cut into pieces so that the spermatozoa can exit and do not coagulate. The solution is then taken a little and dropped on a Hemacytometer and then placed under a microscope. Observations were made as many as five fields of view as a repetition.

2.4 Data analysis

The data were analyzed statistically with normality test using Kolmogorof - Smirnof test, then the data were analyzed with One-Way Anova and DMRT further test. Data analysis using SPSS program.

3. Results and Discussion

3.1 Result

Integrity of the cell membrane of spermatozoa

The mean integrity of spermatozoa obtained in the control group (K) with the treatment group P1, P2 and P3 (dose 300 mg/KgBB, 750 mg/KgBW and 1050 mg/KgBW) with mean values of 42.67%; 68.33%; 80.50% and 86.67% respectively. Further test results showed that there was a significant difference between treatment K with treatment P1, P2 and P3. There are also significant differences between the treatment of P1 with P2, P1 with P3 and between P2 with P3. The results of the data obtained, P3 treatment with a dose of 1050 mg/kg of bran tea water was able to increase the integrity of the spermatozoa membrane by 86.67% (Table 1).

Table 1. The results of the average statistical analysis of the integrity of the membranes of spermatozoa

Treatment	Integrity of the membranes of spermatozoa (%)	
	Intact	Broken
	Mean $\pm$ Standard Deviation	Mean $\pm$ Standard Deviation
K	42,67 $\pm$ 3,33 ^a	57,33 $\pm$ 3,33 ^a
P1	68,33 $\pm$ 2,42 ^b	31,67 $\pm$ 2,42 ^b
P2	80,50 $\pm$ 1,05 ^c	19,50 $\pm$ 1,05 ^c
P3	86,67 $\pm$ 1,21 ^d	13,33 $\pm$ 1,21 ^d

Note: The values in the table above show the average of 24 mice and 4 groups, the numbers accompanied by a different letter notations in the same column show significantly different average values based on the DMRT (Duncan Multiple Range Test) at the level of 5% after statistical test using ANOVA; P0= control treatment (without application of extract); P1= treatment of extract dose 350mg/KgBW; P2= treatment of extract dose 700mg/KgBW; P3= treatment of extract dose 1050mg/KgBW

Motility of spermatozoa

The average number of spermatozoa motility obtained in Group K (untreated group) is 52.96%, then group P1 is the group that was given water extract of bran tea with a dose of 300 mg/KgBW has an average number of spermatozoa motility of 58.17%. Treatment of P2 given water extract of tea Bran 750 mg/KgBW and P3 given water extract of tea Bran 1050 mg/KgBW get the average number of sperm motility respectively 66.34% and 75.02%. The results of statistical analysis showed that Group K with P1 showed no significant difference. These results indicate that the water extract of bran tea on the P1 treatment (dose 300 mg/KgBW) did not exert a significant effect when compared with the P2 and P3 groups (Table 2).

Table 2. The results of the average statistical analysis of the motility of spermatozoa

Treatment	Motility of Spermatozoa (%)	
	Motile	Non Motile
	Mean $\pm$ Standard Deviation	Mean $\pm$ Standard Deviation
K	52,96 $\pm$ 6,56 ^a	47,04 $\pm$ 6,56 ^a
P1	58,17 $\pm$ 4,85 ^a	41,83 $\pm$ 4,85 ^a
P2	66,34 $\pm$ 5,87 ^b	33,66 $\pm$ 5,87 ^b
P3	75,02 $\pm$ 3,18 ^c	24,98 $\pm$ 3,18 ^c

Note: The values in the table above show the average of 24 mice and 4 groups, the numbers accompanied by a different letter notations in the same column show significantly different average values based on the DMRT (Duncan Multiple Range Test) at the level of 5% after statistical test using ANOVA; P0= control treatment (without application of extract); P1= treatment of extract dose 350mg/KgBW; P2= treatment of extract dose 700mg/KgBW; P3= treatment of extract dose 1050mg/KgBW

Viability of spermatozoa

The average number of viable spermatozoa in the control group K was 53.77%. Then in the treatment group P1 (dose 300 mg/kg) of 58.16%; P2 (dose 750 mg/kg) of 66.72% and P3 (dose 1050 mg/kg) with an average value of 75.30%. The results of statistical analysis showed that there is a significant difference between Group K with group P1, P2 and P3. Significant differences were also found between groups P1 with P2 and P3 and groups P2 with P3. The best treatment group that can increase the viability of viable spermatozoa is P3 with an average of 75.30% (Table 3).

Table 3. The results of the average statistical analysis of the viability of spermatozoa

Treatment	Viability of Spermatozoa (%)	
	Viabel	Non Viabel
	Mean $\pm$ Standard Deviation	Mean $\pm$ Standard Deviation

K	53,77 ± 8,06 ^a	46,23 ± 8,06 ^a
P1	58,16 ± 5,54 ^b	41,84 ± 5,54 ^b
P2	66,72 ± 6,66 ^c	33,28 ± 6,66 ^c
P3	75,30 ± 3,67 ^d	24,70 ± 3,67 ^d

Note: The values in the table above show the average of 24 mice and 4 groups, the numbers accompanied by a different letter notations in the same column show significantly different average values based on the DMRT (Duncan Multiple Range Test) at the level of 5% after statistical test using ANOVA; P0= control treatment (without application of extract); P1= treatment of extract dose 350mg/KgBW; P2= treatment of extract dose 700mg/KgBW; P3= treatment of extract dose 1050mg/KgBW

Morphology of spermatozoa

The average results of normal sperm morphology obtained in the control group (K) with the treatment group P1 (dose 300 mg/KgBB), P2 (dose 750 mg/KgBB) and P3 (dose 1050 mg/KgBB) with mean values of 33.50%, 54.33%, 74.83%, and 84.33% respectively. The results of statistical analysis showed that there is a significant difference between Group K with group P1, P2 and P3. Significant differences were also found between groups P1 with P2 and P3 and groups P2 with P3. The highest morphological mean of normal spermatozoa was obtained in P3 treatment with a value of 84.33% (Table 4).

Table 4. The results of the average statistical analysis of the morphology of spermatozoa

Treatment	Morphology of spermatozoa (%)	
	Normal	Abnormal
	Mean ± Standard Deviation	Mean ± Standard Deviation
K	33,50 ± 5,86 ^a	66,50 ± 5,86 ^a
P1	54,33 ± 6,12 ^b	45,67 ± 6,12 ^b
P2	74,83 ± 4,36 ^c	25,17 ± 4,36 ^c
P3	84,33 ± 5,01 ^d	15,67 ± 5,01 ^d

Note: The values in the table above show the average of 24 mice and 4 groups, the numbers accompanied by a different letter notations in the same column show significantly different average values based on the DMRT (Duncan Multiple Range Test) at the level of 5% after statistical test using ANOVA; P0= control treatment (without application of extract); P1= treatment of extract dose 350mg/KgBW; P2= treatment of extract dose 700mg/KgBW; P3= treatment of extract dose 1050mg/KgBW

Concentration of spermatozoa

The average concentration of spermatozoa obtained in the control group (without treatment) was 10.23x106/mL, then in the treatment groups P1 (dose 300 mg/KgBW), P2 (dose 750 mg/KgBW) and P3 (dose 1050 mg/KgBW) with an average value of 25.80x106/mL, 40.80x106/mL and 52.97x106/mL. Further test results showed that there was a significant difference between treatment K with treatment P1, P2 and P3. There are also significant differences between the treatment of P1 with P2, P1 with P3 and between P2 with P3. As a result of the data obtained, P3 treatment with a dose of 1050 mg/kg of bran tea water was able to increase the highest concentration of spermatozoa by 52.97% (Table 5). An increase in the concentration of spermatozoa in each treatment group showed that the aqueous extract of bran tea significantly increased the concentration of spermatozoa.

Table 5. The results of the average statistical analysis of the concentration of spermatozoa

Treatment	Concentration of Spermatozoa (Million/mL)
	Mean ± Standard Deviation
	Mean ± Standard Deviation
K	10,23 ± 0,37 ^a
P1	25,80 ± 0,68 ^b
P2	40,80 ± 0,38 ^c
P3	52,97 ± 0,60 ^d

Note: The values in the table above show the average of 24 mice and 4 groups, the numbers accompanied by a different letter notations in the same column show significantly different average values based on the DMRT (Duncan Multiple Range Test) at the level of 5% after statistical test using ANOVA; P0= control treatment (without application of extract); P1= treatment of extract dose 350mg/KgBW; P2= treatment of extract dose 700mg/KgBW; P3= treatment of extract dose 1050mg/KgBW

3.2 Discussion

Spermatozoa quality

Treatment of bran tea water extract to mice had a significant effect on increasing the quality number of spermatozoa.

Free radicals are molecules that have a group of atoms with unpaired electrons in their outer orbits, radical forms and are very labile and reactive and can damage all types of cellular macromolecules including proteins, carbohydrates, nucleic acids and lipids, and oxidize nucleic acids, proteins, fats. These compounds are formed in the body triggered by various internal and external factors. This unpaired electron tends to look for its partner by attracting or attacking electrons from other compounds and this leads to the formation of new radical compounds. This reaction will continue and will stop if the reactivity is dampened by compounds that are antioxidants.

In the test of intact or not the sperm membrane (integrity) in a hypoosmotic solution, spermatozoa with good membranes (intact) are shown with tails that look circular or bent, but in spermatozoa whose membranes are damaged (broken) are marked with straight tails (Maulidyah, 2015). Treatment groups P1, P2, and P3 were compared with the control group (K) has a significant difference, it shows that all treatment groups have antioxidant activity that is able to fight free radicals so as to increase the integrity of the membrane of spermatozoa mice. Nurhadijah et al., (2018) argue that, antioxidant compounds such as flavonoids and steroids have the ability to maintain sperm cell stability and scavenger against ROS compounds. The effects of these two antioxidants can enhance the regeneration process by reducing free radicals, accelerating the repair mechanism of damaged cell membranes and supplying competitive substrates to unsaturated lipids into cell membranes as well as steroids have the ability to inhibit lipid peroxidation. Prolonged lipid peroxidation can produce MDA (malondialdehyde), MDA is a dialdehyde compound which is the end product of lipid peroxidation, and the appearance of MDA as a sign if there is damage to cell membranes due to oxidation processes. The occurrence of lipid peroxidation causes oxidative stress, one of the factors that causes many cases of infertility in men. Oxidative stress arises due to an increase in ROS, causing DNA damage and apoptosis in sperm, the content of unsaturated fatty acids in large quantities contained in the sperm cell membrane makes ROS compounds can easily enter the sperm plasma membrane because the electrons contained in fat are attracted directly by ROS, the loss of electrons in the sperm cell membrane fat makes damage to the cell membrane and adversely affects the quality of spermatozoa (Suarni et al., 2016). Flavonoids as antioxidants work by giving contained electrons to free radical compounds so that free radicals such as ROS can be neutralized and damage can be prevented and the chain reaction can be interrupted. The break in the follow-up reaction of free radicals that damage sperm cell membranes due to antioxidants that donate their hydrogen ions, makes the sperm cell membrane remain intact and can improve sperm quality (Tethool and Purwaningsih, 2019).

Spermatozoa motility is the ability (power) of sperm to move. Sperm movement is very important because without good movement, sperm are unable to fertilize an egg. The greater the percentage of nonmotiles, the greater the chance of infertility, which is why normal sperm motility is necessary to reach the egg for fertilization to occur. The results of statistical analysis showed that there was a significant difference between the control group (K) with P2 and P3 treatment groups. It was shown that the treatment groups P2 (dose 750 mg/KgBB) and P3 (1050 mg / KgBB) were able to increase the motility of spermatozoa of mice. Bran tea contains flavonoids that are antioxidants and serve as an antidote to free radicals. Free radicals that interfere with the process of spermatogenesis have the effect to reduce the frequency of movement of spermatozoa because it can cause mitochondrial ATP production in sperm to decrease. The source of energy in the sperm to carry out its movement is supplied in the form of ATP by mitochondria located on the neck of the sperm. Damage that occurs in mitochondrial cell membranes can affect the supply of ATP to decrease and result in disruption of spermatozoa metabolism that making spermatozoa motility to decrease (Permatasari et al., 2020). If antioxidants react with free radicals then these radicals will not damage other cells that are nearby. This makes the seminiferous tubules, mitochondrial cell membranes of spermatozoa, and spermatogenic cells free of free radicals that cause ATP production in spermatozoa mitochondria for spermatozoa movement to normalize and spermatozoa motility to increase (Nurliani et al., 2005). antioxidants contained in Forest betel can protect spermatozoa from cell damage caused by oxidation and can improve sperm quality and increase reproductive efficiency so as to increase spermatozoa motility (Suyasa et al., 2022).

In this study, bran tea with doses of 300 mg/KgBW, 750 mg/KgBW and 1050 mg / kgbb can increase the number of viable spermatozoa. This means that bran tea is able to increase the viability of spermatozoa. Bran tea contains flavonoids as antioxidants. Antioxidants found in bran tea water extract can maintain the viability of spermatozoa by supporting the resistance of membrane integrity to oxidative stress. The plasma membrane in spermatozoa composed of PUFA (Polyunsaturated fatty acids) is very sensitive to oxidative stress. The accumulation of antioxidants in the plasma membrane keeps free radicals and antioxidants in balance. Therefore, the integrity of the spermatozoa membrane can be maintained so that the viability of spermatozoa remains viable (Widotama and Gupta, 2008). Research conducted by Suarni et al., (2021) States, that the substitution of commercial feed with moringa leaf feed can increase the viability of male rabbit spermatozoa because the antioxidant compounds contained in moringa leaves can inhibit the negative effects of free radicals.

Lipid peroxidation is the process most responsible for the destruction of spermatozoa membranes. Lipid peroxidation can be prevented by flavonoid antioxidants and works very effectively in suppressing the number of free radicals. Nurliani et al., (2005) explained, If the structure of the spermatozoa membrane is damaged (bad integrity), the viability of sperm will also decrease, therefore the sperm cell membrane has an important role in maintaining the structure and activity of sperm.

The morphology of spermatozoa is the overall shape of sperm, both size, structure, and appearance. Normal sperm morphology consists of one (1) oval-shaped head and an elongated tail with a size of 9-10 times the length of the sperm head (Nugraheni et al., 2005). Increasing the number of spermatozoa whose morphology is abnormal can decrease male fertility. Abnormalities in sperm morphology can occur due to the presence of ROS in spermatogenesis. ROS compounds can interfere with the normal process of apoptosis and DNA damage, rising levels of ROS compounds, and lack of antioxidants resulting in cellular DNA damage and morphological abnormalities (head, neck, and tail) (Suarni et al., 2021). Free radicals inhaled into the body can cause abnormalities in spermatozoa and disrupt the process of spermatogenesis, resulting in damage to the membranes of Sertoli cells and Leydig cells. Damage to both membranes will affect the production of hormones, testosterone, LH (lutein hormone), and FSH (follicle stimulating hormone). The decline of these three hormones results in disruption of the feedback mechanism between the testes and hypothalamus-pituitary which can cause disruption of the process of spermatogenesis and result in the formation of abnormal spermatozoa (Ningrum et al., 2018). The percentage of normal morphology that increased in groups P1, P2 and P3 occurred due to the antioxidant activity of bran tea water extract that can balance the levels of free radicals found in the body of mice. In the opinion of Hardiningtyas et al., (2014) the steroid functions as a scavenger of hydrogen peroxide, causing hydrogen peroxide to not react further into hydroxyl radicals and decreasing free radical production. Flavonoids in bran tea water extract act as antioxidants because they can suppress the number of free radicals such as peroxyl, peroxynitrite, peroxyl radicals, superoxide, hydrogen peroxide and hydroxyl. Flavonoids can also inhibit the process of lipid peroxidation at the initiation stage so that free radicals are not able to develop into new free radicals (Dewanto et al., 2017).

The concentration of spermatozoa is the number of spermatozoa per unit in a unit volume or per one milliliter of semen. Generally, the normal concentration amount of sperm is 10-200x10⁶ cells/mL (Safwan et al., 2016). The decreased concentration of spermatozoa is caused by the presence of toxic compounds in the form of free radicals that can affect hormones so the spermatogenesis process is disrupted (Susilo and Akbar, 2016). Free radicals that inhibit the process of spermatogenesis can attack estrogen receptors, causing inhibition of LH and FSH production and spermatozoa production. The FSH hormone has a direct influence on Sertoli cells in the testes, namely by producing Androgen Binding Protein (ABP) to bind to proteins and stimulate protein synthesis (Muchtaromah, 2009). Free radicals can also induce apoptosis and necrosis in spermatogenic cells causing a decrease in the number of such spermatogenic cells. When the number of spermatogenic cells decreases, causing disruption of spermatozoa formation and maturation thereby decreasing the number of spermatozoa (Ayustina et al., 2022). When the process of spermatogenesis is disrupted, the development of spermatogonium cells can affect the number of spermatozoa to be formed. Therefore, the number of spermatozoa produced is highly dependent on the process of spermatogenesis that occurs in the seminiferous tubules. In this study, the concentration of spermatozoa of the treatment groups P1, P2, and P3 increased markedly compared to the control. This is because bran tea extract contains flavonoid compounds as antioxidants that can ward off free radicals so as to improve the process of spermatogenesis. Nugraheni et al., (2005) argued, flavonoids also function as protective cells that produce the hormone testosterone. If the production of the hormone testosterone is not disturbed by the presence of free radicals, the process of spermatogenesis will run normally. The normal process of spermatogenesis that occurs in the seminiferous tubules makes the production of spermatozoa is not disturbed because the antioxidants contained in the bran tea extract are able to ward off free radicals so that the number of free radicals decreases along with the number of doses of bran tea water extract given.

4. Conclusion

Giving bran tea water extract can improve the quality (membrane integrity, motility, viability, morphology and concentration) of Mouse spermatozoa (*Mus musculus* L.). Then, the treatment group on the provision of water extract of bran tea is the most optimal among the groups tested to improve the quality of spermatozoa mice P3 treatment group (dose 1050 mg / KgBW).

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