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THE EFFECT OF BEET (BETA VULGARIS L.) TO SPERMATOZOA MOTILITY OF MOSQUITO COIL-EXPOSED WISTAR RATS

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ABSTRACT

Background : Mosquito coils contain substances such as allethrin, transflurin, pralethrin, cypenothrin and esbiothrin. Alletrin inhalation will pass into the bloodstream. In the liver, the detoxification occurs and produces metabolites that play a role as free radicals. Beets contain betanin, one of the betalalain groups with functions to binding free radicals in the body system. **Objective :** To investigate the effect beets on spermatozoa motility of wistar rats exposed by mosquito coils. **Methods :** In this study used post test only control group design. The number of samples was 20 male wistar rats divided into 4 groups. The Control Group (K-) is a group was given standard feed. The Control Group (K+) is only given exposed to mosquito coils. The P1 group was exposed to to mosquito coils and given 8 ml of beet juice. Group P2 was exposed to to mosquito coils and given 16 ml beet juice. The treatment were done for 56 days. **Result :** The mean value of motility of spermatozoa are group K (-) = 50; group K (+) = 10; group P1 = 29; group P2 = 46. Oneway Anova test showed significant differences in sperm motility in all groups. Post Hoc test showed significant difference between group K (+) with K (-) (p = <0,001), group K (+) with P1 (p = 0.005), group K (+) with P2 (p = <0,001). **Conclusion :** Beet juice can affect the spermatozoa motility of wistar rats exposed by mosquito coils.
Keyword : mosquito coils; beet juice; sperm motility

INTRODUCTION

Infertility defined as a failure to achieve pregnancy, after 12 months or more of regular unprotected sexual intercourse.[1] The average prevalence of infertility in developed countries are 3.5-16.7% and in developing countries are at 6.9- 9.3%. Infertility in men amounts to 20-40% while in women amounts to 30-55%. Combined, those factors are 35% and unknown causes are 5-15%.[2]

Mosquitoes are insects in which can cause various diseases in which can lead to death. Mosquito are carries (vector) of disease such as malaria, filariasis, Dengue Hemorrhagic Fever (DHF) and other disease. Based on the 2013 RISKESDAS, in amount 48% households in Indonesia choose mosquito coils as an effort to prevent mosquito bites in the house, followed by the use of mosquito nets (25.9%), repellent (16.9%), mosquito spray (12.2%), and ventilation closure by gauze (8%).[3]

Most mosquito coils in Indonesia contain active ingredients such as allethrin, transflurin, pralethrin, cypenothrin or esbiothrin, which are derived from pyrethroid.[4] Inhaled allethrin will enter the bloodstream, go to the liver, detoxify, and produce metabolites that act as free radicals. Then, those free radicals will re-enter the blood circulation and go to the entire body, including the testes.[5]

Beet (Beta vulgaris) is known to be rich of useful ingredients that are beneficial to the body. They include vitamins A, B1, B2, B3, B5, B6, Folate, Vitamin C, Calcium, Iron, and Magnesium. It also contains betanin betaxanthin, betasianin, and flavonoids which are families of betalain. The betanin is an antioxidant that acts as the

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most effective inhibitor to reactions that can cause damage to DNA.[6,7]

METHOD

This research implemented an experimental research design with a post test only control group design. The samples were male wistar rats (Rattus novergicus). The sample size was based on the provision of the World Health Organization (WHO) with a minimum number of samples amounting to 5 experimental animals per group.

This study used four groups of rats; one group which was given food and drink for 56 days that was not given treatment (K (-)), one group that was exposed to mosquito coils for 8 hours per day for 56 days (K (+)), one group that was given exposure to mosquito coils for 8 hours per day for 56 days and was given beet juice at a dose of 8 mg/day (P1), one group that was given exposure to mosquito coils for 8 hours per day for 56 days and was given beet juice with a dose of 16 mg/day (P2). The assessment was done by dissection where 2 cm of cauda epididymis was taken, placed on a petri dish, given a physiological NaCl solution, then performed a massage using a spatula on 2 cm cauda epididymis to get the sperm cells needed, and then stirred to be homogeneous to facilitate examination. Sperm cells that had come out of the epididymis were taken using a pipette and then placed in an obect glass. The cells were then observed under a microscope at 100x magnification using three fields of view. The notes taken were the percentage of spermatozoa that move straight (progressive), move in place (non-progressive), and do not move (immotile), then they were record on the observation sheet.

The data obtained from those four groups were processed using a computer program. A normality test was done using the Shapiro-Wilks test because the number of samples was <50. The data distribution was normal so that the parametric test was carried out with the One Way Anova test and continued with Post-Hoc Least Significant Differences (LSD). The Differences were considered significant if the p value <0.05.

RESULT

This research was conducted at Semarang State University (UNNES) for 56 days. The following results were obtained:

Table 1. Descriptive Analysis and Standard Deviation of Spermatozoa Motility in each group

Groups	N	*Progresif*/SD	*Non-Progresif*/SD	*Imotil*/SD
K (-)	5	50,00/11,06	32,00/3,80	18,00/7,67
K (+)	5	10,00/11,06	24,67/6,91	65,33/12,83
P1	5	29,33/7,23	42,00/3,80	28,67/5,58
P2	5	46,67/7,82	30,00/8,50	23,33/7,45

Based on table 1 above, it can be seen that the average percentage of spermatozoa moving straight shows the highest is from K (-) group (50.00) and the lowest is K (+) group (10.00). For the percentage of spermatozoa that move in place, the table shows that the highest is P1 group (42.00) and the lowest is K (+) group (24.67). As for the percentage of immobile spermatozoa, it is shown that the K (+) group has the highest average (65.33) and the K group (-) has the lowest percentage of immunity (18.00).

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Table 2. The Normality Test Result of Spermatozoa Motility

Kelompok	N	p*		
		Progresif	***Non-Progresif***	***Imotil***
K (-)	5	0,394	0,815	0,685
K (+)	5	0,394	0,753	0,083
P1	5	0,747	0,815	0,314
P2	5	0,103	0,537	1,000

Then, homogeneity test was done using the Levene test to find out whether the groups had the same variants or not.

Table 3. The Homogeneity Test Result of Spermatozoa Motility

			p*
Uji Varian	*Progresif*	*Non-Progresif*	*Imotil*
Levene	0,730	0,503	0,589

Based on the homogeneity test above, it is shown that p value> 0.05, which means that all groups have the same variant. Therefore, all groups are considered homogeneous.

Table 4. The One Way Anova Test Result of Spermatozoa Motility

	p*		
Kelompok	*Progresif*	*Non-Progresif*	*Imotil*
K (-) K (+) P1 P2	0,001	0,003	0,001

The One Way Anova test result is p <0.05, which means that there are significant differences between those five groups. Then, the Post Hoc test was carried out to find out significant difference between each group.

Table 5. The Post Hoc LSD Test Result of Spermatozoa Motility

Progressive Category

	p*		
	P1	**P2**	**K (-)**
K (+)	0,005*	<0,001*	<0,001*
P1		0,010*	0,003*
P2			0,585

The result of the progressive Post Hoc table above shows that there are significant differences in K (+) with P1 (p = 0.005), K (+) with P2 (p = <0.001), K (-) with K (+) (p = < 0.001), P1 with P2 (p = 0.010) and groups P1 and K (-) (p = 0.003). While in the other groups, there is no significant differences.

Non-Progressive Category

	p*		
	P1	**P2**	**K(-)**
K (+)	<0,001*	0,186	0,075
P1		0,007*	0,020*
P2			0,611

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The non-progressive Post Hoc table above shows that there are significant differences in K (+) with P1 (p = <0.001), P1 with P2 (p = 0.007) and P1 with K (-) (p = 0.020). While in the other groups, there is no significant difference.

Immotile Category

	p*		
	P1	**P2**	**K (-)**
K (+)	<0,001*	<0,001*	<0,001*
P1		0,352	0,073
P2			0,353

The immotile Post Hoc table above shows that there are significant differences in K (+) with P1 (p = <0.001), K (+) with P2 (p = <0.001), and K (+) and K (-) (p = <0.001), While in the other groups, there is no significant differences.

DISCUSSION

The result of Post-Hoc LSD test shows that there is a significant difference in the percentage of spermatozoa motility in the K (-) group compared to the K (+) group (p = <0.001). These results support previous research conducted by Faisal Yusuf et al (2018) which states that allethrin contained in mosquito coils can reduce motility, number, viability and morphological abnormalities of spermatozoa.[8]

The spermatozoa motility of mosquito coil-exposed male wistar rats that were given beet treatment has higher percentage compared to those with no treatment. This can be seen from the average treatment group which was exposed to mosquito coils and given beets i.e. P1 and P2 had an increase compared to K (+).

In the assessment of immotile spermatozoa motility, it is also found that there are significant differences between the groups given exposure to mosquito coils and given different doses of beets. Specifically, it is found that a dose of 8 ml gave a difference that showed the protective effect of beets with their anti-oxidant content. There are several research that support this research, one of which is a research conducted by Clifford et al (2015) about the benefits of beet fruit supplementation for health that shows one of the beet supplementation is beneficial as an anti-oxidant.[6]

An existing limitation in this research is that the spermatozoa motility of wistar rats cannot be examined before the treatment, so it cannot be compared with motility after treatment is conducted.

CONCLUSION AND SUGGESTION

Conclusion

Beet juice can affect the spermatozoa motility of wistar rats exposed by mosquito coils..

Suggestion

Further research is needed to investigate the effects of beet juice intake towards the spermatozoa motility of wistar rats with various types of mosquito repellent.

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