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Motility and Viability of Spermatozoa Treated with Sambiloto Leave Extract

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INTRODUCTION

Sambiloto (*Andrographis paniculata*) is one of the herbs that can be found easily in Indonesia as it has been widely used as a traditional medicine.

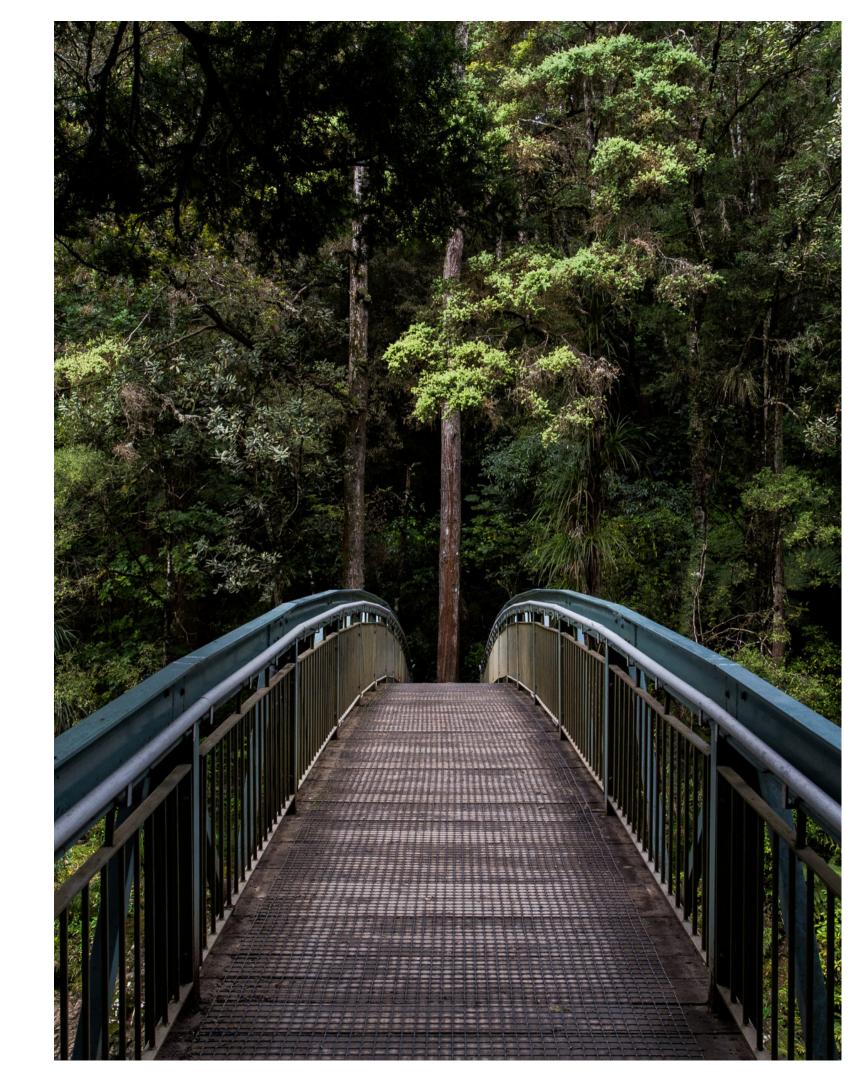
Many researchers in the world are beginning to highlight the used of herbs as alternatives to overcome reproductive problems, as the herbs is considered as natural and not harmful.

Andrographis paniculata is an example of herbs with a pervasive use in Indonesian society. Previous research has suggested that the the introduction of Andrographis paniculate extract on male mice may cause the disruption of seminiferous epithelium.

The potential benefits of **A. paniculata** to be used as antierlitiy for male has put an urge to conduct further research on the effect of A. paniculate leaf extract on the quality of spermatozoa, particularly on mice.

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RESEARCH METHOD

TIME AND PLACE

The research was conducted from May to June 2013 at the Biology Microtechnics Laboratory, UNM

TYPE OF RESEARCH

The Research is an experimental research, consists of 2 stages:
(1) Preparation stage
(2) The implementation Stage

RESEARCH DESIGN

COMPLETELY RANDOMIZED
DESIGN (CRD), consisting of 4
treatment groups with 5
replications to obtain
20 combinations.

RESEARCH DESIGN

The research design used was a **Completely Randomized Design (CRD)** consisting of **four treatment groups** with five replications to obtain twenty combinations.



Group I (A0) is the control group that was not given the n-hexane extract of *A. paniculata*



Group II (A1) is the experimental group that were given the n-hexane extract of *A. paniculata*, **125 mg/Kg bw** for 18 consecutive days

Group III (A2) is the experimental group that were given the n-hexane extract of *A. paniculata,* **250 mg/Kg bw** for 18 consecutive days

Group IV (A3) is the experimental group that were given the n-hexane extract of *A. paniculata*, **500 mg/Kg bw** for 18 consecutive days



Body Weight

Table 1. The average body weight (g) on various treatment groups with n-hexane extract of *A. paniculata* for 18 days

NO	Dosage (mg/bw)	Average ± SD	LSD α 0,05
1	0	24,546 ± 0,3242c	
2	125	22,622 ± 1,2726b	1 527
3	250	21,083 ± 1,1403b	1,527
4	500	22,483 ± 0,9538a	

Note: the numbers followed by the same letter are not statistically different on α 0,05 of LSD test.

The decreasing of mice body weight along with the shrinkage of its testis may occur due to the presence of androgenic and cytotoxic compounds in A. paniculata extract. The compounds is **β-sitosterol** which belong to the group of phytosterol and cytotoxic saponins.

Testis Weight

Table 2. The average weight of testis (g) on various treatment groups of n-hexane extract of *A. paniculata* for 18 days

NO	Dosage (mg/bw)	Average ± SD	LSD α 0,05
1	0	0,609 ± 0,075b	
2	125	0,567 ± 0,009ab	0.100
3	250	0,515 ± 0,396a	0,108
4	500	0,500 ± 0,062a	_

Note: the numbers followed by the same letter are not statistically different on α 0,05 of LSD test.

A decreased in testicular size occurs due to cytotoxic effects contained in n-hexane extract of *A. paniculata* which can cause the death of spermatogenic cells in seminiferus tubule, thus causing a decrease in testicular weight.

Spermatozoa Count

Table 3. The average count of spermatozoa (10⁵ million/ml) on various treatment groups of n-hexane extract of *A. paniculata* for 18 days

NO	Dosage (mg/bw)	Average ± SD	LSD α 0,05
1	0	59,863 ± 2,9744d	
2	125	41,162 ± 1,6996c	0.254
3	250	26,275 ± 2,8712b	9,354
4	500	20,790 ± 3,1819a	

Note: the numbers followed by the same letter are not statistically diferent on $\alpha\,0,\!05$ of LSD test.

The decrease in counts, motility, and viability of sperm is caused by the presence of saponin compounds, β -sitosterol, which can cause the increase of testosterone levels along with the inhibition of energy formation in the body of tested animals.

Spermatozoa Count

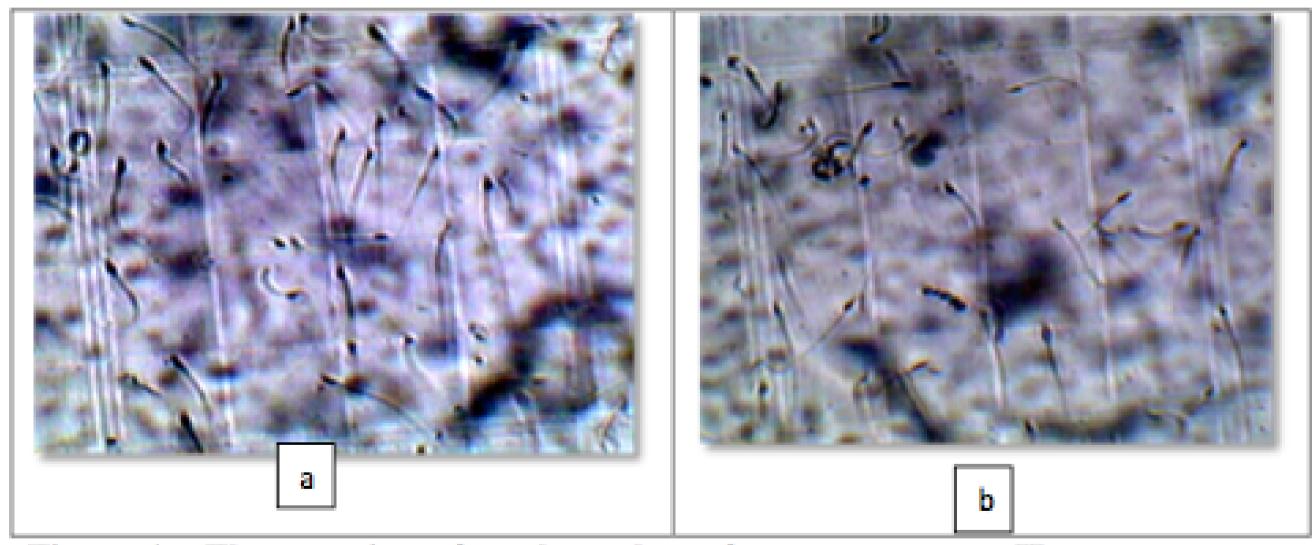


Figure 1. The counting of total number of spermatozoa on Haemocytometer with the magnification of 400 x: a. control group, b. Experimental group

Spermatozoa Motility

Table 4. The average motility of spermatozoa (%) on various treatment groups of n-hexane extract of *A. paniculata* for 18 days

NO	Dosage (mg/bw)	Average ± SD	LSD α 0,05
1	0	34,662 ± 3,4119d	
2	125	24,490 ± 1,0464c	2 20
3	250	16,832 ± 0,3350b	2,29
4	500	12,997 ± 0,2735a	

Note: the numbers followed by the same letter are not statistically different on α 0,05 of LSD test.

Spermatozoa Viability

Table 5. The average viability of spermatozoa (%) on various treatment groups of n-hexane extract of *A. paniculata* for 18 days

NO	Dosage (mg/bw)	Average ± SD	LSD α 0,05
1	0	18,330 ± 1,1885a	
2	125	36.162 ± 3,5559b	4 40 4
3	250	49,495 ± 3,4179c	4,484
4	500	46,283 ± 1,1940c	

Note: the numbers followed by the same letter are not statistically different on α 0,05 of LSD test.

Spermatozoa

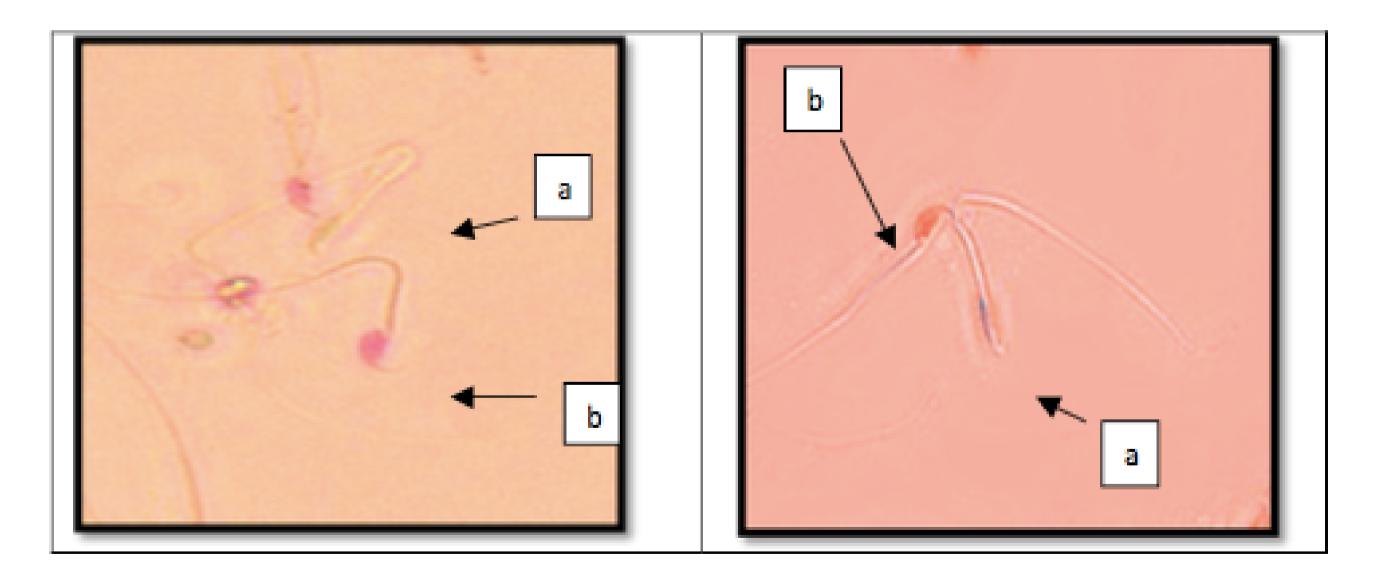


Figure 2. Observation of spermatozoa viability, a. alive spermatozoa = do not absorb dye, b. dead spermatozoa = absorb dye.

Abnormal Spermatozoa

Table 6. The average number of abnormal spermatozoa (%) on various treatment groups of n-hexane extract of *A. paniculata* for 18 days

NO	Dosage (mg/bw)	Average ± SD	LSD α 0,05
1	0	18,330 ± 1,1885a	4 404
2	125	36.162 ± 3,5559b	
3	250	49,495 ± 3,4179c	4,484
4	500	46,283 ± 1,1940c	

Note: the numbers followed by the same letter are not statistically different on α 0,05 of LSD test.



CONCLUSION

Administration of n-hexane extract of A. paniculata leaf at a dose of 125 mg / kg bw, 250 mg / kg bw, and 500 mg / kg bw for 18 consecutive days has affected the spermatozoa quality of male mice (Mus musculus), as it causes the decrease of count, motility, and viability of spermatozoa while increasing the number of abnormal spermatozoa.



THANK YOU FOR YOUR ATTENTION
