Test of the Aphrodisiac Effect of Ethanol Extract of Roots, Stems and Leaves of Packaged Plants (*Smilax rotundifolia*) on Male White Rats (*Sprague Dawley*)

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Abstract

Developing countries use medicinal plants for aphrodisiac treatment, one of which is a pack plant (*Smilax rotundifolia*) which is commonly used to enlarge male genitalia in Papua Indonesia. Because these claims are not scientifically tested and proven, at this time This study aims to determine the effect of the ethanol extract of *S. rotundifolia* stem and leaf tubers on the parameters of sexual behavior in male Sprague Dawley rats. As well as fear the most effective plant parts have aphrodisiac activity. Thirty rats were divided into 5 groups (A-E) consisting of group A given 0.5% Na CMC colloid solution (normal control), group B given X-gra® suspension dosage 51.37 mg/kgBW (positive control), group C was given root ethanol extract dosage 200 mg/kgBW, group D was given stem ethanol extract dosage 200 mg/kgBW and group E was given leaf ethanol extract dosage 200 mg/kgBW. The aphrodisiac test was observed on the 31st day with a ratio of male and female rats (1: 2). All parameters tested in the extract group showed significant differences with the normal group. This showed an aphrodisiac effect in all extract groups. The 200 mg/kgBW stem extract group reduced latent rates and intromission mounts as well as increased copulation rates, showing a marked increase in the number of intromissions, reduced ejaculatory latency and inter-intromission interval, and increased ejaculation frequency. which was not significant in the X-gra group (positive control) (p <0.05). As an extract of the roots, stems and leaves of *S. rotundifolia*, it shows aphrodisiac effects, but the stem extract of *S. rotundifolia* is the most effective in stimulating sexual behavior parameters in male rats.

Keywords: *S. rotundifolia*, Aphrodisiac, sexual behavior, Sprague Dawley rats

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1. Introduction

The United State National Health and Social Life Survey (NHSLS) estimates that approximately 31% of men suffer from sexual dysfunction. Complaints experienced by male patients with sexual dysfunction are erectile dysfunction and premature ejaculation [1]. Medical treatment for erectile dysfunction includes administration of type 5 phosphodiesterase inhibitor drugs (PDE-5 such as sildenafil, local therapy with intracavernous and intrarectal alprostadil. Patients with premature ejaculation require psychosexual counseling and sometimes administration of drugs such as dapoxetine [2]. The use of aphrodisiac chemical drugs, still causes several problems including causing serious side effects [3].

Reviewing the side effects caused by chemical drugs, people in several developing countries use a lot of medicinal plants for treatment [4]. Historical and current use of medicinal plants or also known as Ethnobotany [5]. Ethnobotany is important for the conservation of resources of traditional medicinal plants [6]. The largest contribution of traditional medicinal plants in Indonesia is from the island of Papua, more than 50% of medicinal plants are in the Papua region [7]. One of the traditional Papuan treatments is the use of a pack plant (Smilax rotundifolia) which is commonly used to enlarge male genitals [8].

The pack plant (Smilax rotundifolia) is a plant that is known to have high efficacy as a male medicine [9]. The literature on packaged plants (S. rotundifolia) is still lacking. Plants with the Smilax genus such as Smilax myosotiflora contain alkaloids, saponins, flavonoids, tannins, and coumarin [10]. Saponin compounds (steroid glycosides), flavonoids and alkaloids are active compounds that have potential as aphrodisiacs [11-13].

Aphrodisiac is described as a substance that can increase sexual arousal [3]. Several previous studies concluded that plants with the genus Smilax can have aphrodisiac activity. Concluded that oral administration of S. myosotiflora extract could increase fertility and sexual stimulation in male rats [14].

Based on the above background, research on the aphrodisiac effect of ethanol extract of roots, stems and leaves of packaged plants (S. rotundifolia) on male white rats (Sprague dawley) needs to be done, because there are differences in active compounds in each part of the plant, namely roots, stems, and leaf. Aphrodisiac effects were observed through parameters of mount latency (ML), intromission latency (IL), ejaculation latency (EL), mount frequency (MF), intromission frequency (IF), ejaculation frequency (EF), post-ejaculatory interval (PEI), intromission rate (IR), inter-intromission interval (III), copulatory rate (CR) [10]. Another parameter observed was the increase in testicular weight and seminal vesicles of male rats [15].

Through this research, information on Indonesian medicinal plants that can be used as an aphrodisiac material will be obtained. It is hoped that the development of traditional medicines derived from native Indonesian plants can contribute to the world of health and provide treatment with lower side effects.

2. Experimental section

2.1. Plant Identification

Identification at UPT. Tadulako University Sulawesi Biological Resources

2.2. Extract Making

Fresh samples are wet sorted, washed, drained, chopped, dried, then powdered, sieved, and weighed. 1.43 kg of macerated simplicia powder (96% ethanol solvent; 3×24 hours). The filtrate in rotary (40°C), concentrated, the thick extract is weighed [16].
2.3. Manufacture of test and comparison preparations

Extract preparation: the extract was suspended using 0.5% NaCMC solution in 100 ml ad, homogenized. X-gra® preparation: mixed with X-gra® added 0.5% NaCMC, crushed homogeneously, put into a volumetric flask, added 0.5% NaCMC ad 100 ml, homogenized [3].

2.4. Aphrodisiac Effect Testing

Provision of test and comparison preparations for 30 days [14]. 2 days before testing, female rats were given estradiol valerate (16.00 WIITA) [3]. Male rats were placed in cages (quiet room & 75W red light), and adapted (5 minutes). Entered by female rats, the ratio of male: female (1: 2). 30 minutes of observation using a recording device [14]. Observed and calculated according to standard methods [10], namely:

a. Mount latency (ML): Time (seconds) of introduction of the female mouse to the first mount.

b. Intromission latency (IL): Time (seconds) from introduction of the female mouse to the first intromission.

c. Ejaculation latency (EL): Time (minutes) of first intromission to first ejaculation.

d. Mount frequency (MF): Total mounts observed (30 min)

e. Intromission frequency (IF): Total intromissions observed (30 minutes)

f. Ejaculation frequency (EF): Total ejaculations observed (30 minutes)

g. Post-ejaculatory interval (PEI): Time (minutes) from ejaculation to intromission.

h. Intromission ratio (IR): Total intromissions divided by total mounts plus intromissions

i. Inter-intromission interval (III): Ejaculation latency divided by total intromissions.

j. Copulatory rate (CR): Number of mounts plus intromissions divided by time from first mount to ejaculation (not ejaculation latency)

2.5. Analisis Data

Using the SPSS statistical data processing program including normality, homogeneity, parametric (One-Way ANOVA) or non-parametric (Kruskal Wallis) tests

3. Results and Discussion

Aphrodisiac is described as a substance that can increase sexual arousal [3]. Several previous studies concluded that plants with the genus Smilax can have aphrodisiac activity. Concluded that oral administration of S. myosotiflora extract can increase fertility and sexual stimulation in male rats [14]. The pack plant (Smilax rotundifolia) is a plant that is known to have high efficacy as a male medicine [9].

ML and IL are considered as a mirror of sexual motivation of the rats and it normally acts inversely proportional to sexual motivation or desire [17]. In the administration of S. rotundifolia stem extract showed a reduction in ML and IL which statistically showed no significant difference with the X-Gra group (positive control). Compared with the provision of root and leaf extracts which showed a significant difference with X-gra, this shows that in giving stem extracts the hesitation time for the male rats to move towards receptive females was reduced. Hence, it might suggest that intake stem extract groups were extremely aroused and increased in sexual eagerness. Any alterations in MF or IF are considered as a reflection of libido, strength, potency, sexual performance and vigor in male rats [18]. Treatment of S. rotundifolia stem extract showed a remarkable increase in the number of complete intromissions. Where the S. rotundifolia stem extract was not significant different with the X-gra group. In addition, the S. rotundifolia stem extract showed an increase that was not significantly different from the X-gra group which experienced an increase in IF value. Such elevation in the number of intromissions by male rats suggests that the penile tumescence and rigidity as well as
accessory muscle that help in sustaining erection of the male sexual organs were fully functioning [19].

Ejaculation in male rats is achieved after a certain number of intromissions, approximately 8 to 12. EL is a useful indicator of sexual pleasure and performance, whereas EF represents strength, vigor and stamina [20]. In this experiment, giving S. rotundifolia stem extract showed insignificant differences with X-gra. Similar to the EF parameter, the S. rotundifolia stem extract also showed insignificant differences with X-gra. It is clear evidence that sexual pleasure and performance were enhanced in extract-treated rats, particularly at the stem extract of S. rotundifolia.

| Parameters | Normal | Control groups | Positive | X-gra® (51.37 mg/kg BW) | Roots (200 mg/kgBW) | Stems (200 mg/kgBW) | Leaves (200 mg/kgBW) | p value |
|------------|--------|----------------|----------|------------------------|---------------------|---------------------|---------------------|---------|
| ML (Sec)   | 31.60±7.27| 31.60±7.27c | 31.60±7.27c | 31.60±7.27c | 31.60±7.27c | 31.60±7.27c | 31.60±7.27c | p 0.05 |
| IF (Min)   | 37.40±6.54| 37.40±6.54c | 37.40±6.54c | 37.40±6.54c | 37.40±6.54c | 37.40±6.54c | 37.40±6.54c | p 0.05 |
| MF         | 28.87±6.46| 28.87±6.46c | 28.87±6.46c | 28.87±6.46c | 28.87±6.46c | 28.87±6.46c | 28.87±6.46c | p 0.05 |
| MF         | 9.60±2.07| 9.60±2.07c | 9.60±2.07c | 9.60±2.07c | 9.60±2.07c | 9.60±2.07c | 9.60±2.07c | p 0.05 |
| IF         | 6.60±2.07| 6.60±2.07c | 6.60±2.07c | 6.60±2.07c | 6.60±2.07c | 6.60±2.07c | 6.60±2.07c | p 0.05 |
| EF         | 1.00±0.00| 1.00±0.00c | 1.00±0.00c | 1.00±0.00c | 1.00±0.00c | 1.00±0.00c | 1.00±0.00c | p 0.05 |
| PEI (Min)  | 5.02±0.96| 5.02±0.96c | 5.02±0.96c | 5.02±0.96c | 5.02±0.96c | 5.02±0.96c | 5.02±0.96c | p 0.05 |
| PEI (Min)  | 4.00±0.03| 4.00±0.03c | 4.00±0.03c | 4.00±0.03c | 4.00±0.03c | 4.00±0.03c | 4.00±0.03c | p 0.05 |
| CR         | 5.41±0.80| 5.41±0.80c | 5.41±0.80c | 5.41±0.80c | 5.41±0.80c | 5.41±0.80c | 5.41±0.80c | p 0.05 |

Note: (a) Kruskal-wallis test. (b) One-way ANOVA test. ± = Values are median. p < 0.05= Significant level. The values with different superscript letters in a column are significantly different (p < 0.05). ML= Mount latency. IL= Intromission latency. EL= Ejaculation latency. MF= Mount frequency. IF= Intromission frequency. EF= Ejaculation frequency. PEI= post-ejaculatory interval. IR= Intromission rate. III= Inter-intromission interval. CR= Copulatory rate.

The prosexual effect of S. rotundifolia was further manifested by the significant increase in CR. Such enhancement in CR indicates a sustained increase in interest, stamina, focus and agility in the sexual acts [21]. Last but not least, the administration of S. rotundifolia also pronounced a significant decrease in III. The reduction of III values is an indication that is significant and sustained of penile erection was activated [22]. Both CR and III parameters showed that all extracts showed insignificant differences with positive controls.

Earlier report showed that plants with the genus smilax such as S. myosotiflora contained stigmasterol, sitosterol and campesterol. Stigmasterol has been demonstrated to be acting as an intermediate in the biosynthesis of androgens, corticoids and estrogens [22]. Sexual behavior and penile erection are critically depended on androgens which may act through central and peripheral mechanisms [23]. Thus, the improvement of sexual behaviors was observed in this study could be due to the alteration of androgens in male rats.

4. Conclusion

This study shows that the ethanol extract of packaged plant stems (smilax rotundifolia) can stimulate sexual behavior parameters in male rats. Compared to the ethanol extract of leaves and roots, ethanol extract of packaged plant stems (smilax rotundifolia) at a dose of 200 mg / KgBW has an aphrodisiac effect that is not significantly different from X-gra.
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Conflict of Interest

The authors declare there is no conflict of interest.

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