Oral administration of clabet (Trigonella foenum graecum) seeds ethanol extracts improved testosterone level in old-male wistar (Rattus norvegicus) mice

Bissi Herliane*, Wimpie Pangkahila1,2, IGM Aman1,3

ABSTRACT

Introduction: Aging can cause decreased testosterone levels in men which is known as andropause. Some herbs are known to increase the levels Testosterone. One of them is Klabet seed that contain phyto-testosterone. The purpose of this study was to prove that the administration of ethanol extract of Klabet seeds increased testosterone levels in old male Wistar rats.

Methods: A pretest-posttest control group design study was conducted using 20 male white wistar rats that were divided into 2 (two) groups: control group that received 2 cc of distilled water and treatment group that received 2 cc ethanol extract of Klabet seeds 250mg/250gBB rats/day by sonde for 14 days. The testosterone level was measured on day 1 and 15.

Results: The analysis showed that there was no significant difference in the pre-test and post-test testosterone level in P0 (Pre-test: 16.4±0.553 ng/ml; posttest: 15.9±1.51ng/ml) while increased level of testosterone could be observed in treatment group (P1) (pre-test: 16.6±0.476ng/ml; post-test: 19.4±0.828ng/ml). Comparative analysis between post-test results (P0 vs P1) showed that the difference was statistically significant (p <0.001).

Conclusion: Oral administration of Klabet seeds ethanol extract can significantly increase testosterone level in old male wistar mice.

Keywords: Testosterone, Andropause, Klabet seeds ethanol extract

INTRODUCTION

Every living thing, sooner or later will experience the aging process. Women want to always look beautiful and youthful; while men want to stay handsome and strong in old age.1,2 Aging is a natural process marked by a decline in cognitive function, cell function and various body organs so that all body components cannot function properly. Aging does not occur in a short time but through several stages and many factors could affect aging including hormones. Hormone is a substance that contributes greatly in slowing down the aging process and maintaining the quality of organ functions. But as humans grow older, the body produces a lot of hormones that are unable to maintain normal hormone levels. One hormone that almost always decreased in aging is testosterone. Decreased testosterone in men due to aging is called andropause which is usually caused by hypogonad.1,2

Andropause is a symptom due to aging which is usually closely related to loss of sexual desire, decreased sexual ability, decreased sexual activity, impotence, disorders of spermatogenesis and erectile dysfunction.3 This situation will affects sexual life, which in turn can reduce the quality of life. Normal testosterone ranges from 300-1000 ng/dl but the number of Leydig cells reduced in about 6 million-7 million per year. Testosterone tends to decrease rapidly after the age of 30 and will continue to decline. At the age of 50, testosterone decreases around 20 to 50 percent.4

In recent years there have been many progresses in research to find an herbal ingredient that can be used as an alternative to hormone therapy that could increase testosterone level.5,6,7,8 Traditional medicine is still very popular because of its preferable side effects, tends to be safer and considered as inherited knowledge from ancestors which is passed from generation to generation.8 Some natural materials including plants can be used as traditional medicine, one of which is Klabet seeds which is known to improve sexual performance and ability by increasing testosterone.5,6,9

Klabet seeds contain phytotestosterone compounds that have similar physiological effects with natural testosterone. Klabet seeds also contain many flavonoids, vitamin C, antioxidants, tannins, phenols, alkaloids, steroids and saponins. The entire active compounds contained in Klabet seeds

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METHODS

The Klabet seeds used in this study were obtained from Purworejo, Central Java, which was extracted using 96% ethanol in the Biopepticide Laboratory of the Faculty of Agriculture, Udayana University for phytochemical testing. The dose of ethanol extract of Klabet seeds used is 250 mg / 250 g BB / day while 2 cc distilled water was used as placebo. The study used 20 old male wistar rats (Rattus norvegicus) aged 18 months with a body weight of 250-260 grams which then divided into 2 groups: the control group and the treatment group. 1 ml of blood was taken from each rat through orbital medial canthus for testosterone evaluation before treatment (pretest). The rats were anesthetized with ketamine 10% dose 50mg/kgBB and zylazine 2% dose 20mg/kgBB injected intramuscularly in the thighs. The control group was given 2 cc of aquadest with a frequency of 2 times a day which administered by sonde for 14 days. The treatment group was received ethanol extract of Klabet seeds with a dose of 250 mg / 250 g BB rats / day with a frequency of twice a day for 14 days. On the 15th day, post-test testosterone level was assessed by similar protocol. After the treatment was completed, all mice were left alive and properly maintained at the Laboratory Animal Unit of the Pharmacology Laboratory, Faculty of Medicine, Udayana University, Denpasar-Bali.

RESULTS

This study proves that the administration of Klabet ethanol extract of 250 mg / 250g BB rat / day for 14 days of treatment can increase testosterone levels in old male wistar rats. Ethanol extract of Klabet seeds with an active compound which has the potential to increase testosterone. Prior to analysis, the normality and homogeneity of the data in all groups were assessed (Table 1 and Table 2). Descriptive analysis data showed mean testosterone pretest in the control group (PO) 16.4 ± 0.553 ng / ml and the treatment group (P1) 16.6 ± 0.476. The mean testosterone posttest control group (P0) was 15.9 ± 1.51 ng / ml and the treatment group (P1) was 19.4 ± 0.828 ng / ml (Table 3).

The results of the significance analysis with unpaired T-test (Independent sample T-test) mean testosterone (pretest) between groups showed no significant difference because the value of p = 0.341, while the average testosterone (posttest) between groups showed a significant difference because the p value <0.001 (Table 3).

Table 1 Normality test result in every group

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>P</th>
<th>Interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td>P0 Pre-test</td>
<td>10</td>
<td>0.313</td>
<td>Normal</td>
</tr>
<tr>
<td>P1 Pre-test</td>
<td>10</td>
<td>0.378</td>
<td>Normal</td>
</tr>
<tr>
<td>P0 Post-test</td>
<td>10</td>
<td>0.333</td>
<td>Normal</td>
</tr>
<tr>
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</tbody>
</table>

Table 2 Inter-group homogeneity test result

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<tr>
<th>Group</th>
<th>N</th>
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<tbody>
<tr>
<td>Testosteron Pre-test</td>
<td>20</td>
<td>0.426</td>
<td>Normal</td>
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<tr>
<td>Testosteron Post-test</td>
<td>20</td>
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Table 3 Comparative analysis between groups as well as before and after treatment

<table>
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<tr>
<th>Group</th>
<th>Testosterone (ng/ml)</th>
<th>P*</th>
<th>Before</th>
<th>After</th>
<th>Mean Difference</th>
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<tr>
<td>Control</td>
<td>164.4±0.553</td>
<td>0.45</td>
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<td>0.553</td>
<td>0.456</td>
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<td>&lt;0.001</td>
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DISCUSSION

Neuroendocrine theory is one of the theories about aging which explains that the aging process is related to hormone levels. The hypothalamus plays an
important role in coordinating and controlling the secretion of hormones by various organs of the body. When it gets old, decreased hormone production has an impact on the body's function being disrupted. An example is andropause in men. In andropause men there is a decrease in testosterone levels\(^4\). For this reason, hormone therapy is needed which can increase and maintain testosterone levels so that they remain at normal levels, either using therapy with chemical or herbal drugs\(^{7,8,9,10}\). Klabet seeds are one of the ingredients that can be used as medicine as hormone therapy\(^{5,6}\). Klabet seeds contain phytotestosterone, tannins, flavonoids, vitamin C, antioxidants, phenols, alkaloids, saponins and steroids\(^{5,6}\). The entire active compound found in Klabet seeds has the potential to increase testosterone.

**CONCLUSION**

Based on the results of this study it can be concluded that administering ethanol extract of Klabet seeds orally at a dose of 250 mg / 250 g BB / day for 14 days increased testosterone levels in old male Wistar rats.

**REFERENCES**