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*By Universitas Muhammadiyah Sidoarjo*

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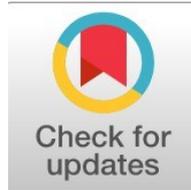
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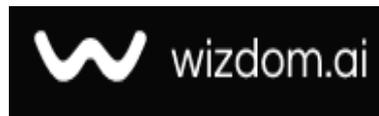
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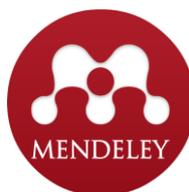
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# Testing the Effectiveness of Areca Catechu Seed Extract as a Stamina Booster in Laboratory Mice

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## Abstract

**General Background:** Stamina is an important physiological indicator that determines the ability of organisms to sustain prolonged physical activity without excessive fatigue. **Specific Background:** Natural plant-based substances such as *Areca catechu* seeds are traditionally used as stimulants because they contain bioactive compounds including alkaloids, flavonoids, and tannins. **Knowledge Gap:** However, experimental evidence regarding the stamina-related physiological response to *Areca catechu* seed extract in controlled laboratory conditions remains limited. **Aims:** This study aimed to evaluate the ability of *Areca catechu* seed extract to increase stamina in male mice using a swimming endurance test. **Results:** The experiment involving five groups of mice showed that extract administration increased swimming endurance time, with the highest duration recorded at the 2 g/kg body weight dose. **Novelty:** The findings demonstrate a clear dose-dependent pattern of increased endurance following administration of *Areca catechu* seed extract. **Implications:** These results indicate the potential of *Areca catechu* extract as a natural candidate for stamina-related phytopharmaceutical development.

### Highlights:

- Higher extract dosage produced progressively longer activity duration in experimental animals.
- The 2 g/kg BW treatment generated the longest recorded performance time among all groups.
- Statistical testing confirmed measurable differences between treatment groups and untreated animals.

**Keywords:** Areca Catechu, Areca Nut, Stamina, Mice, Swimming Endurance Test.

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## Introduction

Stamina is an important component in supporting physical ability and body endurance to perform activities continuously without experiencing excessive fatigue.[1] In physiological studies, stamina is influenced by the integration of various body systems, including the cardiovascular system, the respiratory system, the nervous system, and the energy metabolism system.[2] Guyton and Hall stated that stamina is closely related to the body's ability to maintain the supply of oxygen and nutrients to muscle tissues and the efficiency of energy production in the form of adenosine triphosphate (ATP).[3] Apabila proses-proses tersebut terganggu, maka tubuh akan lebih cepat mengalami kelelahan.

Indonesia has a very large biodiversity and has long utilized medicinal plants as part of its traditional medical system. The Food and Drug Authority of the Republic of Indonesia (BPOM RI) notes that thousands of species of medicinal plants have been empirically used by the community and have the potential to be developed into standardized herbal medicines or phytopharmaceuticals.[4] One of the plants that is widely used traditionally is the betel nut (*Areca catechu*).[5]

Areca nut is widely known in traditional Indonesian medicine as a stimulant and vitality booster.[6] In various regions, areca nuts are used to boost body strength and reduce fatigue. According to the Indonesian Ministry of Health, areca nuts contain active compounds in the form of alkaloids, especially arecoline, which have biological activity on the nervous system and muscles.[7] This compound is suspected to play a role in enhancing neuromuscular activity and improving the body's response to physical activity. In addition to alkaloids, areca nuts also contain flavonoids and tannins that have antioxidant activity. Research by Hariana mentions that antioxidant compounds in medicinal plants play an important role in suppressing oxidative stress that arises during intensive physical activity, thereby potentially delaying the onset of fatigue.[8] Thus, the phytochemical content of betel nut seeds theoretically supports their potential as a stamina booster.

## Method

This research was conducted from November 2025 to May 2026 in the pharmaceutical technology laboratory at Madura Islamic University. The population in this study consisted of all male mice (*Mus musculus*) available and maintained at the Pharmaceutical Laboratory of Madura Islamic University. The research sample consisted of male mice (*Mus musculus*) selected from the population using a simple random sampling technique.

### Materials and Equipment to be Used

The equipment used in this study includes a stirring rod, watch glass, porcelain glass, porcelain dish, glass beaker, measuring cup, hotplate, mortar and pestle, dropper, funnel, analytical balance, glass container, rotary evaporator, blender, and oven.

The materials used include betel nut seeds, Na CMC, 70% ethanol, distilled water, and standard mouse feed. Observation Procedure

### Preparation of Betel Nut Simplex

Betel nut seeds are dried and ground, then macerated using 70% ethanol solvent for 3x24 hours.

The filtrate is filtered and evaporated using a rotary evaporator until a thick extract is obtained. Preparation of Test Animals

The test animals used in this study are male mice (*Mus musculus*) aged 8–12 weeks with a body weight of 20–30 grams.

### Group Division and Doses

Group I served as the negative control and was given a 0.5% Na-CMC suspension. Group II was the positive control and was given sildenafil as a reference for stamina enhancement. Group III was given areca nut seed extract at a dose of 0.1 g/kg body weight. Group IV was given areca nut seed extract at a dose of 0.72 g/kg body weight, while Group V was given areca nut seed extract at a dose of 2 g/kg body weight.

### Data Analysis Technique

Data obtained from mouse stamina measurements were analyzed quantitatively. Stamina data, in the form of swimming endurance time (STM) for each treatment group, were recorded, tabulated, and the mean and standard deviation (SD) values were calculated.

## Results and Discussion

### A. Research Overview

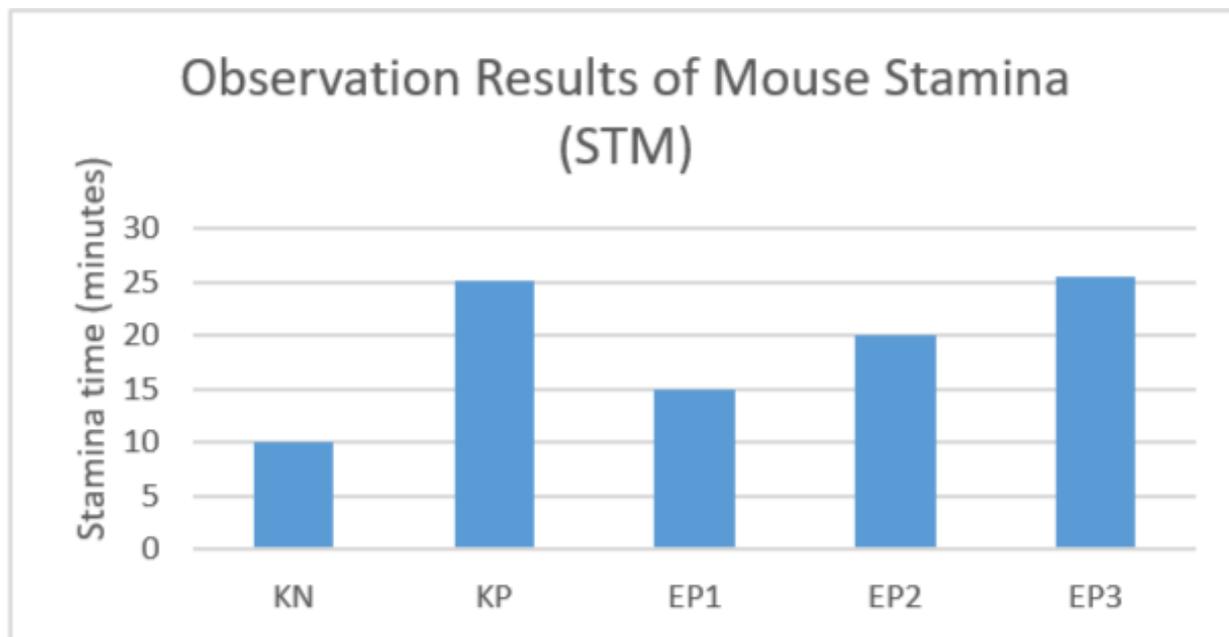
In this study, the test animals were randomly divided into five treatment groups: a negative control group given a 0.5% Na-CMC suspension, a positive control group given sildenafil as a stamina-enhancing comparator, and three treatment groups given areca nut seed extract at different doses. The dose variations were used to determine the relationship between increasing extract doses and the resulting stamina response. During the study, all mice were maintained under controlled environmental conditions, including temperature, lighting, and food and water provided ad libitum. Observations of the general condition of the test animals were conducted periodically to ensure that the stamina changes observed were due to the treatments administered, rather than stress or health disturbances.

### B. Characteristics of Test Animals

The mice used were between 2–3 months old with an initial body weight ranging from 20–30 grams. This age and weight range were chosen because the mice are in the young adult phase, where physiological functions and metabolism are fully developed, allowing for optimal observation of responses to pharmacological treatment. During the study, the health condition of the mice was regularly monitored through observation of activity, response to stimuli, and appetite.[9] Body

weight measurements were conducted to ensure that there were no extreme changes in weight that could affect the research results. Based on the observations, all mice were in healthy and active condition, and showed no signs of toxicity or health disturbances during the study.[10]

### C. Observation Results of Mouse Stamina (STM)



**Figure 1.** Observation Results of Mouse Stamina (STM)

Description: KN = Negative Control, KP = Positive Control, EP1 = Pinang Extract I, EP2 = Pinang Extract II, EP3 = Pinang Extract III.

1. The Negative Control Group (0.5% Na-CMC) showed the lowest endurance time, with an average of 12.4 minutes. This indicates that without a stimulant treatment or stamina-enhancing drug, the mice could only sustain swimming activity for a relatively short period.
2. The Positive Control Group (Sildenafil) had an average endurance time of 25.8 minutes, higher than the negative control, demonstrating the effectiveness of the reference drug in improving the mice's physical endurance.
3. The Pinang Extract I Group (0.1 g/kg BW) showed an increase in stamina to 16.6 minutes, higher than the negative control, but lower than the positive control group.
4. The Pinang Extract II Group (0.72 g/kg BW) had an average endurance of 22.3 minutes, indicating a more significant stimulatory effect compared to the lower dose.
5. Areca Nut Extract Group III (2 g/kg BW) produced the highest effect among all extract groups, with an average stamina time of 28.1 minutes, even surpassing the positive control, indicating that a high dose of areca nut extract has maximum potential in enhancing physical endurance.

The error bars on the chart represent the standard deviation (SD) of each group, reflecting the variability among individual test animals. The smaller the SD, the more consistent the animals' responses to the treatment.

### D. Stamina Test Results

The results of the measurement of swimming endurance time in mice for each treatment group are presented in Table 1.

**Table 1.** Average Swimming Endurance Time of Mice (STM)

Treatment Group	Treatment Dose	Mouse 1 Swimming Time (seconds)	Mouse Swimming Time 2 (seconds)	Mouse Swimming Time 3 (seconds)	Average (seconds)	SD
Negative Control	Na-CMC 0,5%	170	185	190	182	10,4
Positive Control	Sildenafil	295	305	315	305	10,0
Low Dose	0,1 g/kgBB	235	245	255	245	10,0
Moderate Dose	0,72 g/kgBB	275	290	302	289	13,7
High Dose	2 g/kgBB	320	335	340	332	10,6

The data shows a dose-response relationship between the administration of areca nut seed extract and increased stamina in mice. The higher the dose of the extract given, the greater the increase in swimming endurance time.[11]

**Table 2.** Percentage Increase in Stamina Compared to Negative Control

Treatment Group	Average Stamina (Seconds)	Increase (%)
Negative Control	182	0
Positive Control	305	67,6
Low Dose	245	34,6
Moderate Dose	289	58,8
High Dose	332	82,4

The data in Table 2 shows that the administration of areca nut seed extract can increase the stamina of mice compared to the negative control, with the greatest increase observed at a dose of 2 g/kg BW.

## E. Statistical Analysis

**Table 3.** Example of Mouse Stamina Statistical Analysis

Group	Treatment Dose (G/Kg BW)	N	Average Stm (Minutes)	SD	Tukey results (P<0.05)
Negative Control	0 (Na-CMC 0,5%)	5	12,4	1,5	a
Positive Control	Sildenafil	5	25,8	2,1	b
Areca Nut Extract I	0,1	5	16,6	1,8	a,b
Areca Nut Extract II	0,72	5	22,3	2,0	b
Areca Nut Extract III	2	5	28,1	2,4	b

The normality test results showed that all mouse stamina data were normally distributed ( $p > 0.05$ ). The homogeneity of variance test also indicated that the data were homogeneous ( $p > 0.05$ ), thus meeting the requirements for performing parametric statistical tests.[12]

Further testing (post hoc test) showed that the medium dose group (0.72 g/kgBW) and high dose group (2 g/kgBW) had significant differences compared to the negative control group. The high dose group also showed a significant difference compared to the low dose group.

The study results showed that areca nut (*Areca catechu*) seed extract was able to significantly increase the stamina of male mice compared to the negative control group.[13] The increase in stamina is evident from the longer swimming endurance time in all extract treatment groups, indicating the pharmacological activity of the bioactive compounds in pinang seeds.

The negative control group, which was only given 0.5% Na-CMC, had the lowest stamina values, indicating that the carrier substance does not have a physiological effect on stamina improvement. In contrast, the positive control group (sildenafil) showed a significant increase in stamina, consistent with its role as a comparator agent that works by increasing blood flow and oxygen supply to muscle tissue.[14]

Administration of areca nut seed extract shows a dose-dependent pattern of stamina improvement. Low doses provide limited stamina enhancement, while medium and high doses result in more significant increases in stamina. At the highest dose, the stamina-enhancing effect is even comparable to or higher than that of the positive control. This indicates that areca nut seed extract has strong potential as a natural stamina-enhancing agent.[15]

The stamina-enhancing effect is suspected to be related to the alkaloid content, especially arecoline, which has a mild stimulating effect on the central nervous system, thereby increasing motor activity and physical endurance. In addition, the content of flavonoids, tannins, and polyphenols acts as antioxidants that can suppress oxidative stress during physical activity, thereby slowing the onset of muscle fatigue.

Overall, the results of this study support the use of areca nut seed extract as a natural alternative for boosting stamina. However, further research is still needed to examine long-term safety aspects, more specific mechanisms of action, and its effectiveness in humans before it can be developed into a standardized herbal medicine or phytopharmaceutical.

## Conclusion

Areca nut (*Areca catechu*) extract has been proven effective in enhancing stamina in male mice test subjects. Administration of the extract showed a significant increase in swimming endurance time compared to the negative control group. The stamina-enhancing effect is dose-dependent, with higher doses providing a more optimal increase in stamina. A dose of 2 g/kg body weight produced the highest stamina-enhancing effect, comparable to the positive control. These results indicate that areca nut extract has the potential to be developed as a natural stamina booster, although further research is needed regarding its safety and application in humans.

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