JOURNAL LA MEDIHEALTICO

VOL. 03, ISSUE 02 (110-115), 2022
DOI: 10.37899/journallamedihealtico.v3i2.579

Sedative Test of Ethanol Extract of Putri Malu Leaves (Mimosa Pudica Linn.) in Mice (Mus Musculus) With Standardized Herbal Medicine Lelap as Comparison

Jefri Naldi[1], Indra Ginting[1], Singgar Ni Rudang[2], Suci Ramadayanti[3]

[1]*Lecturer of Pharmacy, Faculty of Pharmacy and Health, Helvetia Institute of Health Medan, Indonesia*
[2]*Lecturer of Pharmacy, Faculty of Pharmacy, Universitas Sumatra Utara, Indonesia*
[3]*Students of Pharmacy, Faculty of Pharmacy and Health, Institute of Health. Helvetia Medan University, Indonesia*

**Corresponding Author: Jefri Naldi*
Email: jefrinaldi@gmail.com

Article Info

Article history:
Received 2 March 2022
Received in revised form 28 March 2022
Accepted 31 March 2022

Keywords:
Sedative
Sensitive Plants Leaves
Herbal Medicine Asleep

Abstract

Dense activity and with increasing age experienced by each community, is one of the causes of decreased sleep quality and trigger sleep disorders. The use of medicinal plants is increasingly in the community such as Sensistive plant leaves. This study aims to determine the sensitive plants and to know the comparison of the work activities of the sensitive plants leaves with a deep sleep. This research was an experimental study with a complete random design in a unidirectional pattern. Test animals used 15 male mice, divided into 5 groups randomly consisting of 3 animals. First Group was given a 0.5% CMC suspension, Second group was given a deep suspension, Third group was given an sensitive plants leaves extract with 5% concentration, fourth group was given an sensitive plants leaves extract of 10% concentration, fifth group was given an sensitive plants leaves extract of 20%. Data obtained by Anova test, followed by Duncan's different test to see the real difference in each experimental group. Administration of sensitive plants leaves extract of 5% leaves did not make a significant difference with sleep, but at a concentration of 10%, 20% gave a significant difference with sedative test on mice that were given a deep sleep. Suspension of sensitive plants leave has a sedative effect on mice, at a concentration of 10%, 20% has the same sedative effect as deep sleep, and research needs to be done on other parts of the sensitive plants leaves that has a stronger sedative effect.

Introduction

Health is a very important thing to maintain. This is in accordance with the meaning of health in the Republic of Indonesia Law No. 36 of 2009 concerning health which states that health is a healthy state, both physically, mentally, spiritually and socially that allows everyone to live socially and economically productive lives. The need for sleep can be considered as a protection from the organism to avoid the harmful effects of sleep on the body. The sleep center in the brain regulates this physiological function which is very important for the health of the body (Nugroho et al., 2019).

Almost everyone has experienced sleep disorders during their lifetime. This also corresponds to increasing age and various causes. Kaplan and Sadock reported that approximately 40-50% of the elderly population suffer from sleep disorders. Chronic sleep disorders (10-15%) are

caused by psychiatric disorders, drug and alcohol dependence (Wahyuningrum, 2021; Sunarti & Helena, 2018).

Sleep problems such as insomnia can sometimes make daily life more stressful or make a person less productive. Loss of sleep is known to cause imbalance in accepting tasks involving memory, learning, and logical reasoning. People have more difficulty concentrating and become less emotional or irritable. This can be a factor in not achieving work targets, academic tasks, and others (Hallion et al., 2018; Xie et al., 2020).

In addition to using medicines made from synthetic drugs, not a few people use traditional medicines which are believed to be medicines, around 80% of the world's population has used medicinal plants to maintain their primary health, this is reinforced by the back to nature thinking that has contributed to an increase in public awareness of the production of organic food and medicines based on natural raw materials that are healthier, safer and cheaper (Khan & Ahmad, 2019).

Many types of plants grow and are often found and many of them are proven to have medicinal properties. One of the medicinal plants as a sedative is the Putri malu plant (Mimosa pudica Linn.), the parts of the plant that are often used are herbs and roots (Khan & Ahmad, 2019).

The shy daughter plant (Mimosa pudica Linn.) is a wild plant, grows on the side of the road, in the field, breeds quickly, grows sleeping on the ground or sometimes upright. This plant has long been known to have a unique character, namely the leaves that will close when touched by something. This plant comes from South America and Central America, but then spread and grows in Southeast Asia, South Asia, and the Pacific Islands. If you look at the shy daughter plant (Mimosa pudica Linn.) it has round, hairy and thorny stems and small leaves arranged in a compound (Bandara et al., 2021).

This plant is believed to have a calming effect and is used to treat insomnia. Which contains tannins, mimosin, pipecholinic acid. In previous studies, the Putri malu plant (Mimosa pudica Linn.) had activity as a sedative, decreased blood glucose levels, decreased uric acid levels, anthelmintic, purti malu plant with species (Mimosa microphylla D.) having sedative activity (Singh et al., 2021; Parvathy et al., 2021; Rajendiran et al., 2019). The purpose of this study was to determine the sedative test of the Ethanol Extract of Putri Malu Leaves (Mimosa pudica Linn.) in Mice (Mus musculus) with standardized herbal medicine Lelap as a comparison.

Methods

The research used in this study was an experimental method, namely by testing the sedative effect of Putri malu leaf extract (Mimosa pudica Linn.) in mice with Lelap Standardized Herbal Medicine as a comparison.

Making Ethanol Extract of Putri Malu Leaves

Putri malu leaves are used by the community as a sedative, which is 20 g in 100 ml. So, in the manufacture of Putri malu leaf extract it takes as much as 1 kg, the dose of Putri malu leaf extract tested as a sedative based on previous research is 5%, 10%, 20%. So in this study the doses tested were the same, namely 5%, 10%, 20% with the comparison of Standardized Herbal Medicines.

Putri malu leaf extract was started by selecting the fresh Putri malu leaves and the leaves were still intact as much as 1 kg, then washed with running water until clean. Cut the leaves of the shy princess into several parts, then let them dry without being exposed to direct sunlight. After the leaves of the shy princess are dry, then powdered using a blender until smooth and sieved using a mess, then weighed and stored in a clean and airtight container.

Putri malu leaf extract was made using a maceration solvent with 70% ethanol as a solvent. The method of making ethanol extract of Putri malu leaves is as much as 320 grams of Putri

malu leaf powder, then put it into a container and poured with 2400 ml of 70% ethanol (75 parts), then stir the lid with plastic. Let stand for 5 days protected from the sun, do the stirring at least 3 times. Filter with flannel using a wooden filter. The dregs were rinsed with ethanol until 3200 ml . was obtained (100 parts). Put in a tightly closed container, let stand for 2 days in a dark place. Pour it over and put it in the container provided, then vaporize the ethanol using a rotary evaporator until you get a thick extract of daunputri malu. The thick extract was weighed and dissolved with 0.5% CMC to obtain the desired concentration.

Manufacturing of 0.5% CMC Suspension

Weigh as much as 0.5 g of CMC (Carboxyl Methyl Cellulose) sprinkled into a mortar that already contains 25 ml of hot aquadest, leave for 15 minutes to obtain a transparent mass, after it expands it is crushed and then diluted with a little aquadest. Then put into a container, enough with aquadest to 100 ml.

Determination of Dosage of Putri Malu Leaf Ethanol Extract

From the results of maceration of 320 g of dry powder of Putri malu leaves, it resulted in 58.19 g of thick extract of Putri malu leaves. The thick extract of the Putri malu leaves was given in 3 doses, namely 5%, 10%, 20%.

Sleep Dose Determination

The dose of sleep therapy for humans is 600 mg. Then the dose conversion was carried out between humans weighing 70 kg to 20 g mice which according to Laurence & Bacharach was 0.0026, (14) so that a deep dose was obtained, namely 600 mg x 0,0026 x 1000 /20= 78 mg/kgBB. The dose used was 78 mg/kgBW, then diluted with 0.5 ml according to the capacity of the mice and administered orally using an oral probe.

Determination of Timing or Sleeping Habits and Activities of Mice

Observations were made for 3x24 hours to see the habits of the mice, such as being active, scratching, silent and resting (asleep). In this study, the size of the cage used was 100 cm x 30 cm x 30 cm aimed at facilitating the movement of 4 mice. obtained when the mice start to be active, then at that time they are given treatment with the aim that the material given causes the expected effect.

Test Subject Treatment

The test animals used in this study were 15 male mice aged 2-3 months weighing 19-30 g. Before being used, the mice were adapted to the environment for one week and made observations by looking at the habits of the mice for 3x24 hours and being fasted for about 16 hours by being given drinking water. Determination of Sleep Time in Mice.

After observing the habits of mice, treatment can be carried out on mice by calculating the sleep time of each mouse in minutes for each group. Sleep time in mice is from the moment the mice lose consciousness, which is marked by a state where the mice are still in an oblique position until the reflex returns to restore the body position to normal. The normal position is characterized by the mice being able to stand on all fours and the head can be lifted perfectly.

Results and Discussion

The Result of Making Ethanol Extract of Putri Malu Leaves

The leaves of the shy daughter were taken from the stabat area, Langkat Regency, North Sumatra. In the morning, the leaves of the shy princess still look fresh and have not closed, so taking the leaves of the embarrassed princess is done in the morning.

In this study, the extraction method chosen was maceration because it was the simplest and easiest method to do. Maceration was carried out by immersing 320 grams of the powder of

Putri malu leaves in a liquid filter. The obtained maserate was then concentrated. The process of concentrating the powder of the Putri malu leaves resulted in 58.19 g of thick extract with a yield of 18.18%.

Results of Making and Determining Dosages and Treatment of Test Subjects

100 ml of 0.5% CMC has been made which is used as a suspending agent, which avoids homogeneity when suspending the Putri malu leaf extract and sleeping pills. In the administration of sleeping medicine as a positive control, dose conversion was carried out between humans weighing 70 kg to 20 g mice according to (Laurence & Bacharach).

The mice were divided into 5 groups, each group consisted of 3 mice, then the mice were fasted for 16 hours before given treatment. Each group of mice were given 0.5% CMC (negative control), lelap (positive control), Putri malu leaf extract with doses of 5%, 10%, 20% by oral route. Then observe and note how many minutes the mice began to fall asleep and the reappearance of the reflex to restore normal body position.

Results of Observation of Time or Habits and Activities of Mice

Mice have the same habits, what can generally be seen is the habit of eating, drinking, resting (sleeping), staying still, scratching (body care) and moving normally. These are the types of habits that mice do.

In this case the researchers observed the habits of mice that were active, silent, and resting (asleep), and were carried out for 3x24 hours where 4 mice were used and on the 4th morning the mice had the same active properties, when compared to other times.

Sedative Test Results in Mice

From the results of the sedative test on mice given CMC 0.5% as a negative control, sleep as a positive control, and P1 5%, P2 10%, P3 20% had a significant difference seen from the 10th minute to the 150th minute. After the data was processed using One-Way ANOVA, there was a significant difference in the sedative test ($\propto = 0.05$). This can be seen in Duncan's average difference as below.

Table 1. Duncan's Average Difference Test Results Against Sedative Test at 10, 20 and 30 minutes

Treatment	Subset for alpha = 0.05			Treatment	Subset for alpha = 0.05	Treatment	Subset for alpha = 0.05
	1	2	3		1		1
CMC 0,5%	1.0000			CMC 0,5%	1.3333	Lelap	2.0000
Lelap	1.3333	1.3333		Lelap	2.0000	P1	2.0000
P1	1.6667	1.6667	1.6667	P1	2.0000	P2	2.0000
P3		2.0000	2.0000	P2	2.3333	P3	2.3333
P2			2.3333	P3	3.0000	CMC 0,5%	3.0000
Sig.	.112	.112	.112	Sig.	.081	Sig.	.253

At the 10th minute there was no significant difference between the administration of 0.5% CMC and sleep but it was significantly different from the sedative test given P1 5%, P2 10%, P3 20%. This shows that 0.5% CMC has the same effect as sleep, while P1 5%, P2 10%, P3 20% has not seen a sedative effect. At the 20th and 30th minutes of all mice there was no significant difference ($\propto = 0.05$), this means that there is uniformity of mice responding to the

treatment given.

Table 2. Duncan's Average Difference Test Results Against Sedative Tests In The 40th, 50th, 60th and 70th Minutes

Treatment	Subset for alpha = 0.05		Treatment	Subset for alpha = 0.05	Treatment	Subset for alpha = 0.05	Treatment	Subset for alpha = 0.05
	1	2		1		1		1
P1	2.0000		Dark	2.6667	P1	2.0000	P1	2.0000
Dark	2.3333	2.3333	CMC 0.5%	3.0000	P3	2.3333	CMC 0.5%	3.0000
P3	2.3333	2.3333	P1	3.0000	Dark	3.0000	P3	3.0000
CMC 0.5%	3.0000	3.0000	P2	3.0000	CMC 0.5%	3.3333	Dark	3.3333
P2		4.0000	P3	3.0000	P2	3.3333	P2	4.0000
Sig.	.212	.050	Sig.	.693	Sig.	.200	Sig.	.073

At the 40th minute there was no difference in the sedative test of mice that were given sleep and P1 5%. This shows that giving P1 5% has a sedative effect, P3 20% and P2 10% at 40 minutes have not had a sedative effect. At the 50th, 60th and 70th minutes of all mice there was no significant difference ($\propto$ = 0.05), this means that there is uniformity of mice responding to the treatment given.

Table 3. Duncan's Average Difference Test Results Against Sedative Tests In the 80th and 90th Minutes

Treatment	Subset for Alpha = 0.05		Treatment	Subset for alpha = 0.05	
	1	2		1	2
P1	2.0000		P1	1.6667	
Dark	3.0000	3.0000	P2	1.6667	
P2	3.6667	3.6667	Dark		3.3333
P3	3.6667	3.6667	CMC 0.5%		4.0000
CMC 0.5%		4.0000	P3		4.0000
Sig.	.077	.264	Sig.	1.000	.338

At the 80th minute there was no significant difference between the administration of P1 5% and sleep but it was significantly different from the sedative test that was given sleep, P2 10%, P3 20%. This shows that P1 10%, P3 20% has the same effect as sleep and the sedative effect has been seen. At the 90th minute to the 150th minute of giving P1 5%, P2 10% and P3 there was no significant difference with sleep. This shows that giving P1 5%, P2 10% and P3 has no sedative effect.

From the sedative test, Putri malu leaf extract has the same effect as standardized herbal medicine for sleep, it can be seen at 80 minutes. Where the mimosin contained in Putri malu binds to receptors and GABA, the inhibitory effect of neurons by Putri malu leaves causes channels When chloride is open, the inhibitory effect of neurons increases the electronic potential along the cell membrane, so that cells are difficult to excite and a sedative-hypnotic effect occurs.

Conclusion

The conclusion from the results of this study is that the ethanolic extract of the leaves of Putri malu with concentrations of 5%, 10%, 20% has a sedative effect and at concentrations of 10%, 20%, has the same effect as standard herbal medicines.

Thank-You-Note

Acknowledgments are conveyed to the Supervisor I, namely Mrs. Mayang Sari, ST, M.Si and the Supervisor II, namely Mr. Indra Ginting, Drs. MM, Apt guided and provided direction to the authors during this research.

References

Bandara, H. M. N. T., Karunaratne, A. M., Abayasekara, C. L., Panagoda, G. J., Senanayake, M. R. D. M., & Nanayakkara, B. S. (2021). A herbal hand sanitizer with minimal alcohol and Mimosa pudica leaves-a preliminary investigation. *Sri Lankan Journal of Infectious Diseases*, 11.

Hallion, L. S., Steinman, S. A., & Kusmierski, S. N. (2018). Difficulty concentrating in generalized anxiety disorder: An evaluation of incremental utility and relationship to worry. *Journal of anxiety disorders, 53*, 39-45.

Khan, M. S. A., & Ahmad, I. (2019). *Herbal medicine: current trends and future prospects. In New look to phytomedicine (pp. 3-13)*. Academic Press.

Nugroho, R. A., Sari, Y. P., Hardi, E. H., & Aryani, R. (2019). *Myrmecodia: Efek Fisiologi dan Potensi Manfaat*. Deepublish.

Parvathy, P., Murali, V. S., Devi, V. M., Murugan, M., & Jmaes, J. J. (2021). ICP-MS assisted heavy metal analysis, phytochemical, proximate and antioxidant activities of Mimosa pudica L. *Materials Today: Proceedings, 45*, 2265-2269.

Rajendiran, D., Khan, H. B. H., Packirisamy, S., & Gunasekaran, K. (2019). Dose dependent antidiabetic effect of Mimosa pudica leaves extract in type 2 diabetic rat model. *Phar. Innov. J, 8*, 1-04.

Singh, S., Dodiya, T. R., Singh, S., & Dodiya, R. (2021). Topical Wound Healing, Antimicrobial and Antioxidant Potential of Mimosa pudica Linn root Extracted using n-Hexane Followed by Methanol, Fortified in Ointment Base. *International Journal of Pharmaceutical Sciences and Nanotechnology*, *14*(3), 5472-5480.

Sunarti, S., & Helena, H. (2018). Gangguan Tidur pada Lanjut Usia. *Journal of Islamic Medicine, 2*(1), 1-15.

Wahyuningrum, E. (2021). Gangguan Tidur Anak Usia Sekolah. *Jurnal Keperawatan, 13*(3), 699-708.

Xie, C., Xie, Z., & Li, D. (2020). *Association of electronic cigarette use with self-reported difficulty concentrating, remembering, or making decisions in US youth*. Tobacco induced diseases, 18.