JPPIPA 10(SpecialIssue) (2024)

Jurnal Penelitian Pendidikan IPA

Journal of Research in Science Education

http://jppipa.unram.ac.id/index.php/jppipa/index

The Effect of Ethanol Extract of Cat's Whiskers Leaves (*Orthosiphon stamineus*) on Reducing Cholesterol Levels in Female Sprague Dawley Rats

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Received: June 5, 2024
Revised: August 5, 2024
Accepted: August 25, 2024
Published: August 31, 2024

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DOI: 10.29303/jppipa.v10iSpecialIssue.8400

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Abstract: This study aims to test the administration of ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) to reduce cholesterol levels in female Sprague Dawley rats. This type of research is a quasi-experiment design using a one group pre-test and post-test design. The population in this study was 30 female Sprague Dawley rats aged ± 3 months with a body weight of around 150-250 grams. Data were analyzed using the t test statistic at = 5%. The results of the research showed that the highest decrease in cholesterol levels was in female Sprague Dawley rats at a test dose of 1000 mg/kgBW and there was an effect of ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) on reducing cholesterol levels in female Sprague Dawley rats.

Keywords: Cat's whisker leaves; Cholesterol; Ethanol extract; Female sprague dawley rats

Introduction

Cholesterol is a fatty substance that circulates in the blood, with a yellowish color and resembles wax, which is produced by the liver and is necessary for the body (Alamgir, 2018; Budianto et al., 2022; Sudiarto et al., 2017). Cholesterol is a group of lipids and is not hydrolyzed and is the main sterol in human body tissue. Cholesterol has an important meaning because it is the most important element in plasma lipoproteins and plasma membranes and is a precursor in a large number of steroid compounds (Haiti et al., 2023; Li et al., 2019; Schade et al., 2020; Sekhar et al., 2020; Zampelas et al., 2019).

The cholesterol produced by the body consists of 2 (two) types, namely HDL (High Density Lipoprotein) cholesterol which is called good cholesterol and LDL (Low Density Lipoprotein) cholesterol which is called bad cholesterol (Cho, 2022; Nessler et al., 2018; Patel et al., 2022). LDL cholesterol will accumulate in the walls of the coronary arteries which will cause blockages, thus LDL is called bad cholesterol (Pirro et al., 2018). Excessive cholesterol levels in the blood are called hypercholesterolemia (Benito-Vicente et al., 2018; Melendez et al., 2017; Tada et al., 2018).

The first step to managing cholesterol begins with a non-pharmacological approach, namely in the form of meal planning or medical nutrition therapy and physical activity. If through these steps the cholesterol control target has not been or is not achieved, then it is continued by using medication or pharmacological intervention (Kowalski, 2010).

Currently, ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) is one of the traditional medicinal plants which is generally used to treat several diseases in Southeast Asia. This plant is generally used in Indonesia to treat rheumatism (Kusmala et al., 2023; Rafi et al., 2021), diabetes mellitus (Andriaty et al., 2019; Nguyen et al., 2019; Wang et al., 2022), high blood pressure (Saberi et al., 2023), tonsillitis (Ali et al., 2019), and kidney stones (Satyaningtijas et al., 2023). In Vietnam, cat's whisker leaves are used to treat urinary

How to Cite:
Berampu, I. M. S. (2024). The Effect of Ethanol Extract of Cat's Whiskers Leaves (*Orthosiphon stamineus*) on Reducing Cholesterol Levels in Female Sprague Dawley Rats. *Jurnal Penelitian Pendidikan IPA, 10*(6), xxxx – xxxx. https://doi.org/10.29303/jppipa.v10i6.xxxx

tract stones, edema, influenza, hepatitis and gallstones. Meanwhile in Myanmar, cat's whisker leaves are used to treat diabetes and urinary tract diseases. The ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) itself contains orthosiphon glucose, essential oils, saponins, polyphenols, flavonoids, sapofonins, potassium salts and myonositol. Some of these substances are found in other plants which have the ability to lower cholesterol levels (Andrianto, 2011).

One of the traditional medicines used for treatment is cat's whisker leaves. Cat's whisker leaves contain chemical compounds, namely sinensetin, rosmarinic acid, eupatorin and 3-hydroxy-5,6,7,4-tetramethoxyflavone (TMF) (Ashraf et al., 2018; Aziz et al., 2021; Chung et al., 2020). The compounds contained in cat's whisker leaves are believed to have anti-allergic, anti-hypertension, anti-inflammatory, anti-oxidant, diuretic and anti-diabetic effects. Cat's whisker leaf extract at a concentration of 1g/kg BW for mice is most effective in reducing plasma glucose concentrations and reducing plasma triglyceride concentrations and increasing HDL-cholesterol concentrations significantly (Faramayuda et al., 2021).

He et al. (2023) states that reducing cholesterol can be done through various mechanisms, namely through inhibition of pancreatic lipase, inhibition of pancreatic cholesterol esterase, cholesterol micellization, and related to bile acids. Cat's whisker leaf extract has the highest ability to bind to bile acids when compared to other plants. The chemical compounds contained in it can be believed to have an anti-cholesterol effect, namely eugenol and flavonoid compounds (Adisakwattana et al., 2012).

Himani et al. (2013), that cat's whisker leaf extract contains a lot of flavones, polyphenolic compounds, active proteins, glycosides, essential oils, potassium and a small amount of terpenoid compounds. (Surahmaida et al. (2019) and Nurcholis et al. (2022) also stated that the chemical contents of cat's whisker leaf extract are saponins, flavonoids and polyphenols, so the dry extract of cat's whisker leaves is able to reduce cholesterol levels. Nurmeilis (2015) stated that alcohol-water extract of cat's whisker bark at doses of 500 and 750 mg/kgBW showed significant anti-hyperlipidemic activity in mice induced by a high-fat diet. The main compound in cat's whisker leaf extract is a flavonoid compound which is thought to provide anti-hyperlipidemia effects.

Research by Dini (2015) shows that blood cholesterol levels in mice were measured using a UV-Vis spectrophotometer with a wavelength of 500 nm after administering the test extract on the 29th day, showing that there was a decrease in total cholesterol levels in the positive control group and the test dose of 250. 500, and 1000 mg/kgBW significantly compared to the normal control and negative control groups ($p \leq 0.05$).

Based on the above background, researchers are interested in conducting research entitled the effect of ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) on reducing cholesterol levels in female Sprague Dawley rats.

Method

The research method used is a quasi-experimental design using a one group pre-test and post-test design, namely a design that attempts to explain cause and effect relationships by involving one experimental group (Sugiyono, 2018). The experiment itself is an observation under artificial conditions where the conditions are created and regulated by the researcher. The population is 30 female Sprague Dawley rats aged ± 3 months with a body weight of around 150-250 grams. The sample in this study was the entire population used as a sample, namely 30 individuals. The data obtained was then analyzed using the data analyzed using the t test statistical test at a significance value of 5%.

Result and Discussion

Average Cholesterol Levels in Female Sprague Dawley Rats

The average cholesterol level in female Sprague Dawley rats before intervention can be seen in Table 1. Based on Table 1, it can be seen that the average value of cholesterol level in female Sprague Dawley rats before intervention in the normal control group was 99.6 points and after intervention 99.4 points with an average difference of 0.2 points, the average value of cholesterol levels in female Sprague Dawley rats before intervention in the negative control group was 98.4 points and after intervention 97.8 points with an average difference of 0. 6 points, the average value of cholesterol levels in female Sprague Dawley rats before intervention in the positive control group was 92.0 points and after intervention 92.0 points with an average difference of 15.8 points, the average value of cholesterol levels in rats Female Sprague Dawley before intervention in the test group with a dose of 250 mg/kgBW was 112.6 points and after intervention 88.8 points with an average difference of 23.8 points, the average value of cholesterol levels in female Sprague Dawley rats before intervention in the group the test dose of 500 mg/kgBW was 110.2 points and after intervention 85.8 points with an average difference of 24.4 points and the average value of cholesterol levels in female Sprague Dawley rats before the intervention in the test dose group of 1000 mg/kgBW was 124.6 points and after intervention 92.4 points with an average difference of 32.2 points.

This situation shows that the cholesterol levels in the female Sprague Dawley rats treated with the test dose of 1000 mg/kgBW decreased, but the female

Sprague Dawley rats who experienced the most decrease in the test dose of 1000 mg/kgBW. This proves that the greater the dose of ethanol extract of cat's whisker leaves in female Sprague Dawley rats, the lower the cholesterol levels will be and conversely the smaller the dose of ethanol extract of cat's whisker leaves in female Sprague Dawley rats, the lower the lower cholesterol levels.

In the anti-cholesterol activity test of the ethanol extract of cat's whisker leaves with 3 dose variations, the extract given at a dose of 1000 mg/kgBW for 14 days showed the highest anti-cholesterol activity which was able to reduce the difference in the average cholesterol levels of mice before and after the intervention to 32.2 point. If given longer, the possibility of developing hypocholesterolemia is higher. Then continued with a dose of 500 mg/kgBW which was able to reduce the difference in the average cholesterol levels of mice before and after the intervention by up to 24.4 points.

Table 1. Average Cholesterol Levels in Female Sprague Dawley Rats Before and After Intervention

Groups	Cholesterol Before Intervention			Cholesterol After Intervention			Difference
	Average	Minimum Cholesterol	Cholesterol Maximum	Average	Minimum Cholesterol	Cholesterol Maximum	
Normal control	99.60	78	135	99.40	78	135	0.2 (0.2%)
Negative control	98.40	72	138	97.80	72	137	0.6 (0.6%)
Positive control	107.80	60	175	92.00	49	160	15.8 (14.7%)
Test dose 250 mg/kgBB	112.60	60	158	88.80	47	130	23.8 (21.1%)
Test dose 500 mg/kgBB	110.20	85	146	85.80	71	118	24.4 (22.1%)
Test dose 1000 mg/kgBB	124.60	100	160	92.40	78	105	32.2 (25.8%)

This research is in accordance with that carried out by Rambe (2015) regarding the effects of black cumin suspension preparations at doses of 378 mg/kgBW, 756 mg/kgBW, and 1,134 g/kgBW using the high cholesterol feed induction method for 14 days. The results obtained were that each dose of black cumin suspension preparation was able to reduce the total cholesterol levels of normal mice by 35.89%, 43.16%, 47.13%. This concludes that cat's whisker herbal extract is no less beneficial than ginger rhizome and black cumin in reducing total cholesterol levels. Differences in reduction percentages can be caused by differences in test methods used, differences in compound content, differences in doses given and duration of administration (Al-dualimi et al., 2018; Yehya et al., 2019).

Effect of Ethanol Extract of Cat's Whisker Leaves (Orthosiphon stamineus) on Reducing Cholesterol Levels in Female Sprague Dawley Rats

Bivariate analysis was used to test the administration of ethanol extract of cat's whisker leaves (orthosiphon stamineus) to reduce cholesterol levels in female Sprague Dawley rats. Before carrying out bivariate analysis, first look at the homogeneity test and can be seen in Table 2.

Table 2. Test of Homogeneity of Variances

	Levene Statistic	df1	df2	Sig.
Cholesterol Before intervention	1.305	5	24	0.295
Cholesterol after intervention	1.944	5	24	0.124

Based on table 2, it shows that the cholesterol level before the intervention obtained a Levene Statistics number of 1,305 with a significance or probability (sig) of 0.295, because the significance value is 0.295 > 0.05, it can be concluded that the six cholesterol groups of female Sprague Dawley rats that we compared before the intervention was the same or homogeneous and cholesterol levels after the intervention obtained a Levene Statistics number of 1,944 with a significance or probability (sig) of 0.124 because the significance value is 0.124 > 0.05, it can be concluded that the variance of the six groups of cholesterol in female Sprague Dawley rats that we compared after the intervention was the same or homogeneous.

Table 3. One-Sample Kolmogorov-Smirnov Test

		Cholesterol Before intervention	Cholesterol after intervention
N		30	30
Normal Parameters	Mean	108.87	92.70
	Std. Deviation	32.548	26.693
Most Extreme Differences	Absolute	.141	.156
	Positive	.141	.156
	Negative	-.122	-.108
Test Statistic		0.138	.141
Asymp. Sig. (2-tailed)		0.080	.134c

To see the normality test for cholesterol levels in female Sprague Dawley rats, see Table 3. Based on the table above, it can be seen that the results of the Kolmogorov Smirnov test for cholesterol levels before the intervention obtained a p-value of 0.080 > 0.05 and after the intervention it was 0.134 > 0. 05. Because both

p-values (sig) > 0.05, both cholesterol levels before and after the intervention were both normally distributed.

Table 4. Paired Samples Test

	Paired Differences					t	Sig. (2-tailed)
	Mean	Std. Deviation	Std. Error Mean	95% CI of the Difference Lower	Upper		
Cholesterol Before - Cholesterol After	16.167	16.470	3.007	10.017	22.317	5.376	0.000

To see the effect of ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) on reducing cholesterol levels in female Sprague Dawley rats, it can be seen in Table 4. Based on Table 4, it can be seen that the results of the t test show that the p-value is 0.000 < 0.05, so Ho was rejected, meaning that there was an effect of ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) on reducing cholesterol levels in female Sprague Dawley rats.

The results of the research showed that the results of the t test showed that the p-value was 0.000 < 0.05, so Ho was rejected, meaning that there was an effect of ethanol extract of cat's whisker leaves (*Orthosiphon stamineus*) on reducing cholesterol levels in female Sprague Dawley rats (Seyedan et al., 2016).

This can be proven that based on data on the percentage reduction in cholesterol levels in female Sprague Dawley rats, it is known that all test dose groups (250 mg/kgBB, 500 mg/kgBB, 1000 mg/kgBB) and positive controls showed a significant reduction in total cholesterol levels compared to controls normal and negative controls. The negative control group experienced a reduction in cholesterol levels of 14.7% after stopping giving hypercholesterologenic foods for 14 days. This explains that stopping hypercholesterologenic feed can also affect the reduction in total cholesterol levels in mice, but based on statistical tests this reduction is not significant (Lokman et al., 2019).

The highest total cholesterol level reduction activity was at a dose of 1000 mg/kgBW (25.8%), then a dose of 500 mg/kgBW (22.1%). This is in accordance with research by Umbare et al. (2009) that a preclinical test of alcohol-water extract of cat's whisker bark had significant hypolipidemic activity (p≤0.05) at doses of 500 mg/kgBW and 750 mg/kgBW in rats induced hyperlipid diet. The lipid parameters tested were a decrease in total cholesterol levels and triglyceride levels. The percentage reduction in total cholesterol levels at doses of 500 mg/kgBB and 750 mg/kgBB was 20.3% and 29.0% and the reduction in triglyceride levels was 26.6% and 28.1%.

According to research by Umbare (2009), the percentage reduction in total cholesterol levels of mice using cat's whisker bark extract as the test extract showed a lower percentage than the percentage reduction in total cholesterol levels of mice using cat's whisker herbal extract as the test extract. This explains that the cat's whisker herbal extract used in this research has better anti-hypercholesterolemic activity than cat's whisker bark extract.

Cat's whisker herbal extract showed a different percentage reduction in total cholesterol levels at each dose and based on statistical tests the difference was significant. This explains that the test group had the same activity for each dose (250, 500, 1000 mg/kgBB) in reducing total cholesterol levels in mice. This explains that increasing the dose does not show an increase in activity in reducing total cholesterol levels. Cat's whisker herbal extract at each dose also showed a difference in the percentage reduction in total cholesterol levels compared to the positive control, but this difference was not significant based on the results of statistical tests. This explains that cat's whisker herb extract at each dose showed the same activity as the positive control in reducing total cholesterol levels in mice.

The difference in the reduction in total cholesterol levels in mice could possibly occur due to individual factors in mice. Waloya et al. (2013) dietary fiber intake, fat intake, physical activity and gender differences influence blood cholesterol levels. Research conducted by Joglar et al. (2024) states that there are two factors that influence blood cholesterol levels related to reducing total cholesterol levels in mice, namely controllable factors such as a diet high in saturated fat and cholesterol, genetics, age and gender and uncontrollable factors such as hormones (thyroid, estrogen, insulin), enzymes (pancreatic lipase), biliary obstruction, diabetes mellitus, liver disease, and stress.

Conclusion

This study aims to test the administration of ethanol extract of cat's whisker leaves (orthosiphon stamineus) to reduce cholesterol levels in female Sprague Dawley rats. The results of the study showed that the highest decrease in cholesterol levels was in female Sprague Dawley rats at a test dose of 1000 mg/kgBW. Apart from that, there was an effect of ethanol extract of cat's whisker leaves (orthosiphon stamineus) on reducing cholesterol levels in female Sprague Dawley rats.

Acknowledgments
Thank you to all parties who have helped in this research so that this article can be published.

Author Contributions
The author greatly contributed to the creation of this article, starting from the initial stages of research until the completion of this article.

Funding
No external funding.

Conflicts of Interest
No conflict interest.

References

Adisakwattana, S., Ruengsamran, T., Kampa, P., & Sompong, W. (2012). In vitro inhibitory effects of plant-based foods and their combinations on intestinal α-glucosidase and pancreatic α-amylase. *BMC Complementary and Alternative Medicine*, *12*(1), 110. https://doi.org/10.1186/1472-6882-12-110

Alamgir, A. N. M. (2018). Phytoconstituents—Active and Inert Constituents, Metabolic Pathways, Chemistry and Application of Phytoconstituents, Primary Metabolic Products, and Bioactive Compounds of Primary Metabolic Origin. In *Therapeutic Use of Medicinal Plants and their Extracts* (Vol. 2, pp. 25–164). Phytochemistry and Bioactive Compounds. https://doi.org/10.1007/978-3-319-92387-1_2

Al-dualimi, D. W., Shah Abdul Majid, A., Al-Shimary, S. F. F., Al-Saadi, A. A., Al Zarzour, R., Asif, M., Ein Oon, C., & Abdul Majid, A. M. S. (2018). 50% Ethanol extract of Orthosiphon stamineus modulates genotoxicity and clastogenicity induced by mitomycin C. *Drug and Chemical Toxicology*, *41*(1), 82–88. https://doi.org/10.1080/01480545.2017.1317785

Ali, H., Musa, I. F., Abu Bakar, N. A., Karsani, S. A., & Yaacob, J. S. (2019). In Vitro Regeneration and ISSR-Based Genetic Fidelity Analysis of Orthosiphon stamineus Benth. *Agronomy*, *9*(12), 778. https://doi.org/10.3390/agronomy9120778

Andrianto, T. T. (2011). *Ampuhnya Terapi Herbal Berantas Berbagai Penyakit Berat*. Yogyakarta: Najah.

Andriaty, S. N., Akbar, F., & Wahab, A. (2019). Perbandingan Efektifitas Ekstrak Etanol 96% Akar Dan Daun Kumis Kucing (*Orthosiphon stamineus*) Terhadap Penurunan Glukosa Darah Mencit (Mus Musculus). *Jurnal Ilmu Kedokteran Dan Kesehatan*, *6*(4), 296–303. https://doi.org/10.33024/jikk.v6i4.2118

Ashraf, K., Sultan, S., & Adam, A. (2018). Orthosiphon stamineus Benth. is an outstanding food medicine: Review of phytochemical and pharmacological activities. *Journal of Pharmacy And Bioallied Sciences*, *10*(3), 109. https://doi.org/10.4103/JPBS.JPBS_253_17

Aziz, N. A. A., Hasham, R., Sarmidi, M. R., Suhaimi, S. H., & Idris, M. K. H. (2021). A review on extraction techniques and therapeutic value of polar bioactives from Asian medicinal herbs: Case study on Orthosiphon aristatus, Eurycoma longifolia and Andrographis paniculata. *Saudi Pharmaceutical Journal*, *29*(2), 143–165. https://doi.org/10.1016/j.jsps.2020.12.016

Benito-Vicente, A., Uribe, K. B., Jebari, S., Galicia-Garcia, U., Ostolaza, H., & Martin, C. (2018). Familial Hypercholesterolemia: The Most Frequent Cholesterol Metabolism Disorder Caused Disease. *International Journal of Molecular Sciences*, *19*(11), 3426. https://doi.org/10.3390/ijms19113426

Budianto, Y., & Akbar, M. A. (2022). Kenaikan Kadar Kolesterol Ditinjau Dari Konsumsi Gorengan. *Jurnal Kesehatan Abdurahman*, *11*(2), 8–13. https://doi.org/10.55045/jkab.v11i2.141

Cho, K.-H. (2022). The Current Status of Research on High-Density Lipoproteins (HDL): A Paradigm Shift from HDL Quantity to HDL Quality and HDL Functionality. *International Journal of Molecular Sciences*, *23*(7), 3967. https://doi.org/10.3390/ijms23073967

Chung, Y.-S., Choo, B. K. M., Ahmed, P. K., Othman, I., & Shaikh, M. F. (2020). A Systematic Review of the Protective Actions of Cat's Whiskers (Misai Kucing) on the Central Nervous System. *Frontiers in Pharmacology*, *11*, 692. https://doi.org/10.3389/fphar.2020.00692

Dini, F. M. (2015). *Pengaruh Pemberian Ekstrak Etanol 96% Herba Kumis Kucing (Orthosiphon Stamineus Benth) terhadap Penurunan Kadar Kolesterol Tptal pada Tikus Jantan yang Diinduksi Pakan Giperkolesterol*. UIN Syarif Hidayatullah.

Faramayuda, F., Julian, S., Sr Windyaswari, A., & Sri Mariani, T. (2021). Review: Flavonoid pada Tanaman Kumis Kucing (Orthosiphon stamineus Benth.) Review: Flavonoid Compounds in Orthosiphon stamineus. *Proceeding of Mulawarman Pharmaceuticals Conferences*, *13*, 282–287. https://doi.org/10.25026/mpc.v13i1.478

Haiti, M., & Christyawardani, L. S. (2023). Hubungan Kadar Glukosa dalam Darah dengan Kadar Kolesterol. *Jurnal Keperawatan Silampari*, *6*(2), 1655–1663. https://doi.org/10.31539/jks.v6i2.5405

He, X., Chen, L., Pu, Y., Wang, H., Cao, J., & Jiang, W. (2023). Fruit and vegetable polyphenols as natural bioactive inhibitors of pancreatic lipase and cholesterol esterase: Inhibition mechanisms, polyphenol influences, application challenges.

Food Bioscience, *55*, 103054. https://doi.org/10.1016/j.fbio.2023.103054

Himani, B., Seema, B., Bhole, N., Mayank, Y., Vinod, S., & Mamta, S. (2013). Misai kuching: A glimpse of maestro. *International Journal of Pharmaceutical Sciences Review and Research*, *22*(2), 55–59. Retrieved from https://globalresearchonline.net/journalcontents/v22-2/11.pdf

Joglar, J. A., Chung, M. K., Armbruster, A. L., Benjamin, E. J., Chyou, J. Y., Cronin, E. M., Deswal, A., Eckhardt, L. L., Goldberger, Z. D., Gopinathannair, R., Gorenek, B., Hess, P. L., Hlatky, M., Hogan, G., Ibeh, C., Indik, J. H., Kido, K., Kusumoto, F., Link, M. S., … Zeitler, E. P. (2024). 2023 ACC/AHA/ACCP/HRS Guideline for the Diagnosis and Management of Atrial Fibrillation. *Journal of the American College of Cardiology*, *83*(1), 109–279. https://doi.org/10.1016/j.jacc.2023.08.017

Kowalski, R. E. (2010). *Teori Hipertensi: Program 8 Minggu Menurunkan Tekanan Darah Tinggi dan Mengurangi Risiko Serangan Jantung dan Stroke Secara Alami*. Bandung: Qanita.

Kusmala, Y. Y., Fathiyya A, N., Khanza M. Z, N., Fatimah M, A., & Riyanti, S. (2023). Potentials of the Cat's Whisker Plants (*Orthosiphon aristatus*) for Kidney Health. *Journal of Health and Dental Sciences*, *2*(3), 387–404. https://doi.org/10.54052/jhds.v2n3.p387-404

Li, L.-H., Dutkiewicz, E. P., Huang, Y.-C., Zhou, H.-B., & Hsu, C.-C. (2019). Analytical methods for cholesterol quantification. *Journal of Food and Drug Analysis*, *27*(2), 375–386. https://doi.org/10.1016/j.jfda.2018.09.001

Lokman, E. F., Saparuddin, F., Muhammad, H., Omar, M. H., & Zulkapli, A. (2019). Orthosiphon stamineus as a potential antidiabetic drug in maternal hyperglycemia in streptozotocin-induced diabetic rats. *Integrative Medicine Research*, *8*(3), 173–179. https://doi.org/10.1016/j.imr.2019.05.006

Melendez, Q. M., Krishnaji, S. T., Wooten, C. J., & Lopez, D. (2017). Hypercholesterolemia: The role of PCSK9. *Archives of Biochemistry and Biophysics*, *625-626*, 39–53. https://doi.org/10.1016/j.abb.2017.06.001

Nessler, K., Windak, A., Grzybczak, R., Nessler, M., Siniarski, A., & Gajos, G. (2018). High-density lipoprotein (HDL) cholesterol: more complicated than we think? *Annals of Agricultural and Environmental Medicine*, *25*(3). Retrieved from https://ruj.uj.edu.pl/server/api/core/bitstreams/5b13726b-a703-44aa-821d-ecca6167d81f/content

Nguyen, P. H., Tuan, H. N., Hoang, D. T., Vu, Q. T., Pham, M. Q., Tran, M. H., & To, D. C. (2019). Glucose Uptake Stimulatory and PTP1B Inhibitory Activities of Pimarane Diterpenes from Orthosiphon stamineus Benth. *Biomolecules*, *9*(12), 859. https://doi.org/10.3390/biom9120859

Nurcholis, W., Mahendra, F. R., Gultom, M. F., Khoirunnisa, S., Kurnia, M. A. C., & Harahap, H. H. (2022). Phytochemical, Antioxidant and Antibacterial Screening of Orthosiphon stamineus Leaf Extract Two Phenotypes. *Jurnal Jamu Indonesia*, *7*(3), 121–129. https://doi.org/10.29244/jji.v7i3.280

Nurmeilis. (2015). *Penentuan Profil Lipid-Kolesterol pada Tikus Normal dan Tikus Hiperkolesterol Setelah Pemberian Ekstrak Herba Kumis Kucing (Orthosipkon Stamineus*. Jakarta: Pusat Penelitian dan Penerbitan (Puslitpen) LP2M UIN Syarif Hidayatullah.

Patel, K. K., & Kashfi, K. (2022). Lipoproteins and cancer: The role of HDL-C, LDL-C, and cholesterol-lowering drugs. *Biochemical Pharmacology*, *196*, 114654. https://doi.org/10.1016/j.bcp.2021.114654

Pirro, M., Ricciuti, B., Rader, D. J., Catapano, A. L., Sahebkar, A., & Banach, M. (2018). High density lipoprotein cholesterol and cancer: Marker or causative? *Progress in Lipid Research*, *71*, 54–69. https://doi.org/10.1016/j.plipres.2018.06.001

Rafi, M., W, N. S., Wahyuni, W. T., Arif, Z., & Heryanto, R. (2021). Autentikasi Kumis Kucing (Orthosiphon aristatus) Menggunakan Kombinasi Spektrum Ultraviolet-Tampak dan Partial Least Square Regression. *Indonesian Journal of Cheometrics and Pharmaceutical Analysis*, *1*(2), 93–101. https://doi.org/10.22146/ijcpa.890

Rambe, R. H. (2015). *Pengaruh Pemberian Ekstrak Etanol 96% Herba KumisKucing (Orthosipone stamineus Benth) Terhadap Kadar Kolesterol Total Tikus Normal*. Retrieved from https://repository.uinjkt.ac.id/dspace/handle/123456789/29335

Saberi, S. E. M., & Chua, L. S. (2023). Potential of rosmarinic acid from Orthosiphon aristatus extract for inflammatory induced diseases and its mechanisms of action. *Life Sciences*, *333*, 122170. https://doi.org/10.1016/j.lfs.2023.122170

Satyaningtijas, A., Pamungkas, J., Sa'diah, S., Wientarsih, I., Purnawarman, T., Rini Madyastuti Purwono, Nisa, K., Nugroho, R. A., Hadiyanti, C. R., & Tarigan, R. (2023). Potensi daun kumis kucing dalam meningkatkan hematopoiesis pada kondisi kerusakan ginjal. *Acta VETERINARIA Indonesiana*, *11*(3), 189–195. https://doi.org/10.29244/avi.11.3.189-195

Schade, D. S., Shey, L., & Eaton, R. P. (2020). Cholesterol Review: A Metabolically Important Molecule.

Endocrine Practice, *26*(12), 1514–1523. https://doi.org/10.4158/EP-2020-0347

Sekhar, M. S., Marupuru, S., Reddy, B. S., Kurian, S. J., & Rao, M. (2020). Physiological role of cholesterol in human body. In *Dietary Sugar, Salt and Fat in Human Health* (pp. 453–481). Elsevier. https://doi.org/10.1016/B978-0-12-816918-6.00021-4

Seyedan, A., Alshawsh, M., Alshagga, M., & Mohamed, Z. (2016). Antiobesity and Lipid Lowering Effects of Orthosiphon stamineus in High-Fat Diet-Induced Obese Mice. *Planta Medica*, *83*(08), 684–692. https://doi.org/10.1055/s-0042-121754

Sudiarto, S., Soewito, S., & Huriah, T. (2017). Potensi Licopen Pada Tomat Untuk Penurunan Kadar Kolesterol (LDL) Pada Penderita Hipertensi (Hiperkolesterolemi). *Journal of Nursing and Health*, *2*(1), 22–31. https://doi.org/10.52488/jnh.v2i1.56

Sugiyono. (2018). *Metode Penelitian Kombinasi (mixed Methods)*. Bandung: Alfabeta.

Surahmaida, S., & Umarudin, U. (2019). Studi Fitokimia Ekstrak Daun Kemangi dan Daun Kumis Kucing Menggunakan Pelarut Metanol. *Indonesian Chemistry and Application Journal*, *3*(1), 1. https://doi.org/10.26740/icaj.v3n1.p1-6

Tada, H., Kawashiri, M., Nomura, A., Teramoto, R., Hosomichi, K., Nohara, A., Inazu, A., Mabuchi, H., Tajima, A., & Yamagishi, M. (2018). Oligogenic familial hypercholesterolemia, LDL cholesterol, and coronary artery disease. *Journal of Clinical Lipidology*, *12*(6), 1436–1444. https://doi.org/10.1016/j.jacl.2018.08.006

Umbare, R. P., Patil, S. M., Mate, G. S., & Dongare, S. S. (2009). Hypolipidemic activity of Orthosiphon stamnineus Benth bark extract. *Journal of Pharmacy Research*, *2*(11), 1735–1738. https://doi.org/10.5555/20103127207

Waloya, T., Rimbawan, R., & Andarwulan, N. (2013). Hubungan Antara Konsumsi Pangan dan Aktivitas Fisik Dengan Kadar Kolesterol Darah Pria dan Wanita Dewasa Di Bogor. *Jurnal Gizi Dan Pangan*, *8*(1), 9. https://doi.org/10.25182/jgp.2013.8.1.9-16

Wang, Q., Wang, J., Li, N., Liu, J., Zhou, J., Zhuang, P., & Chen, H. (2022). A Systematic Review of Orthosiphon stamineus Benth. in the Treatment of Diabetes and Its Complications. *Molecules*, *27*(2), 444. https://doi.org/10.3390/molecules27020444

Yehya, A. H. S., Asif, M., Kaur, G., Hassan, L. E. A., Al-Suede, F. S. R., Abdul Majid, A. M. S., & Oon, C. E. (2019). Toxicological studies of Orthosiphon stamineus (Misai Kucing) standardized ethanol extract in combination with gemcitabine in athymic nude mice model. *Journal of Advanced Research*, *15*, 59–68. https://doi.org/10.1016/j.jare.2018.05.006

Zampelas, A., & Magriplis, E. (2019). New Insights into Cholesterol Functions: A Friend or an Enemy? *Nutrients*, *11*(7), 1645. https://doi.org/10.3390/nu11071645