STRADA : Jurnal Ilmiah Kesehatan
DOI: https://doi.org/10.30994/sjik.v14i2.1286
ISSN: 2252-3847 (print); 2614-350X (online) Vol.14 No.2 November 2025 Page. 204-215

The Effect of Ethanol Extract of Jicama (*Pachyrhizus erosus*) on Apoptosis Index and Endometrial Thickness in White Rats (*Rattus norvegicus*) Ovariectomy Model

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ABSTRACT

Menopause is the permanent cessation of menstruation due to the loss of ovarian follicular activity, which marks the end of a woman's reproductive function. During menopause, estrogen levels decrease, triggering oxidative stress, leading to cell death (apoptosis) and a decrease in endometrial thickness, which can lead to symptoms such as hot flushes, sleep disturbances, decreased sexual desire, and changes in the menstrual cycle. Menopause-related symptoms are considered natural and are often ignored because they are related to a woman's menstrual cycle. Menopause-related problems increase morbidity and mortality due to inadequate treatment, so interventions are needed to reduce morbidity by alleviating symptoms during menopause. One traditional plant believed to reduce oxidative stress due to menopause is the phytoestrogen found in Jicama (Pachyrhizus erosus). The purpose of this study was to prove that Jicama (Pachyrhizus erosus) ethanol extract can reduce the apoptosis index and increase endometrial thickness in white rats (Rattus norvegicus) in an ovariectomy model. The study design was a true experimental study with a randomized post-only control group design. The experimental animals used were 30 female white rats (Rattus norvegicus) divided into 5 groups: negative control group, positive control group and group with administration of jicama extract dose of 70 mg/200 g BB (P1), dose of 140 mg/200 g BB (P2), dose of 280 mg/200 g BB (P3), Ovariectomy was performed and waited until the 28th day post ovariectomy to check the vaginal pH to ensure that the rats were in menopause and then given exposure to jicama extract for 14 days. The apoptosis index was examined using Tunnel Assay, while the endometrial thickness was stained with Hematoxylin Eosin.

Keywords: apoptosis index, endometrial thickness, ethanol extract of jicama (pachyrhizus erosus), menopause, ovariectomy

Received September 5, 2025; Revised October 10, 2025; Accepted November 10, 2025

DOI: https://doi.org/10.30994/sjik.v14i2.1286
ISSN: 2252-3847 (print); 2614-350X (online) Vol.14 No.2 November 2025 Page. 204-215

BACKGROUND

Menopause is the permanent cessation of menstruation due to the loss of follicle function in the ovaries which results in a sudden decrease in estrogen and an increase in cardiovascular disease and osteoporosis (Scofield, 2024) (Gaten, 2024). Menopause is a natural process of the body where at menopause there is a decrease and eventual cessation of the hormone estrogen which results in symptoms of hot flushes, night sweats, breast tenderness, vaginal dryness, irregular menstruation, mood swings, vaginal atrophy, osteoporosis, heart disease (Rodriquez, 2012) (Doshi .B. Agarwal, 2013). During menopause hormonal changes occur. When estrogen production falls below a critical value, estrogen no longer inhibits the production of gonadotropins FSH and LH. As a result, FSH and LH production increases (Burger H.G, 2017) (Grub, Jessica., et al, 2024). Estrogen is a powerful antioxidant that provides protection against oxidative stress during the reproductive period (Rodriquez, 2012) (Doshi et al, 2013). Estrogen causes cell proliferation in the endometrium so that the glands become more tortuous and excess secretory substances accumulate in the glandular epithelial cells (Elkholi, et al, 2015) (Kapoor, et al, 2023). However, during menopause with a decrease in the hormone estrogen, endometrial epithelial cell proliferation will decrease and cell death will increase so that if this continues continuously, endometrial organ atrophy will occur. Endometrial atrophy during menopause causes the endometrium to thin, making it prone to inflammation, which ultimately triggers postmenopausal bleeding. Of the 3% to 14.2% of reported postmenopausal bleeding, 59% are caused by endometrial atrophy **(**Mor, G. et al, 2011).

Oxidative stress plays an integral part of the aging process. Oxidative stress occurs when there is an imbalance between free radicals and antioxidants. During menopause, decreased estrogen levels result in increased oxidative stress (Burger, H.G.,,2017). Oxidative stress causes DNA damage, lipid peroxidation, and protein peroxidation. This triggers the release of released mitochondrial proteins into the cytoplasm. This release of mitochondrial proteins into the cytoplasm initiates the apoptosis program (Sato, .T, et al.,.2013). An increase in the apoptosis index in the endometrium after ovariectomy occurs in luminal epithelial cells, glandular cells, and endometrial stroma. In luminal epithelial cells, an increase in the apoptosis index occurs on days 1-2 after ovariectomy, while in glandular cells it increases 5 days after ovariectomy and in stromal cells it increases but experiences a significant decrease on day 5 after ovariectomy Sato, .et al.,2013). Apoptosis in the menopausal endometrium can occur through the intrinsic pathway or the extrinsic pathway. The intrinsic pathway involves the release of mitochondrial proteins into the cytoplasm. The mitochondrial protein released in the endometrium of ovariectomized rats is Smac/diablo, which neutralizes the cytoplasmic protein that functions as a physiological inhibitor of apoptosis (XIAP) (Leblanc, 2003) (Song, .J., Rutherford, et al, 2002). Apoptosis via the extrinsic pathway in the endometrium of ovariectomized rats is characterized by an increase in TNFα and Fas.

One way to reduce oxidative stress is by administering antioxidants. One bioactive ingredient that can be used as an antioxidant in reducing oxidative stress is phytoestrogen. In a study on puerarin in Pueraria roots (Radix puerarieae) as a phytoestrogen, it was shown that puerarin has an effect of inhibiting the growth of cancer cells which is associated with the work of estrogen in the body through binding to ERβ (Primiani, C.N., 2018) (Wang, D. et al., 2011**)**. One of the phytoestrogen compounds that has a structure similar to estrogen is daidzein and ganestein which can cause estrogenic and anti-estrogenic effects (Morán, J. et al., 2013). Daidzein and ganestein are found in many legume products. Asian people themselves consume legume products in various processed forms and are used as a source of high-quality protein. However, in a study on legume products, it was found that soy protein increases serum uric acid levels in humans, thereby increasing the risk of gout (Messina, M., Messina, V.L. and

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Chan, P., 2015). Women with hypoestrogen conditions can experience increased uric acid levels compared to women with normal estrogen levels (Mumford, S.L. et al., 2016) (Tai, Yu, et al, 2023). Therefore, alternative phytoestrogen supplements are needed to replace endogenous estrogens other than legumes. One such alternative is jicama. Jicama has been used by our ancestors for decades as a traditional cosmetic ingredient, and its use is still common today. The content of essential skin compounds such as vitamin C, vitamin A, and AHA in jicama has never been studied, although information on its secondary metabolites has been reported. Several secondary metabolites in jicama and their activities have been studied. (Lukitaningsih and Holzgrabe ,2014) stated that jicama contains isoflavone compounds, namely daidzein, daidzein-7-o-β-glucopyranose, 5-OH-daidzein-7-o-β-glucopyranose, and 8,9-furanyl-pterocarpan-3ol. The compound has also been studied for its activity including UV light absorption, antioxidant and tyrosinase enzyme inhibition (Lukitaningsih et al., 2013). The main phytosterol content in yam tubers has also been known to contain β-sitosterol and stigmasterol with a concentration of about 0.02% per dry weight of yam or about 2.76% in yam petroleum ether extract (Lukitaningsih et al., 2012). The activity of yam extract as a chemo-preventive agent has also been reported by (Nurrochmad et al, 2013). The formulation of the problem in this study is How is the Effect of Yam Ethanol Extract (Pachyrhizus erosus) on the Apoptosis Index and Endometrial Thickness in White Rats (Rattus norvegicus) Ovariectomy Model?.

METHODS

This study used a true experimental design with a post-test only control group design. The study involved ovariectomizing Rattus norvegicus, which were then administered with jicama extract at doses of 70 mg/200 g body weight, 140 mg/200 g body weight, and 280 mg/200 g body weight. Subjects were randomly assigned to a negative control group for comparison, and a treatment group. The parameters measured in this study were the apoptotic index and endometrial thickness.

The population used in this study were white rats (Rattus norvegicus). Thirty white rats, aged 9-12 weeks and weighing 200-250 grams, were used as samples. The dependent variables were the apoptotic index and endometrial thickness of the ovariectomized white rats. The independent variable was the jicama extract (Pachyrhizus erosus). The tools and materials in this study include a cage for keeping mice in the form of a plastic box measuring 20x30x40 cm, an electric scale, a 1 mL syringe and a gastric tube with a tip made of tin, 1 set of surgical tools including a scalpel knife no. 11, a 1 cc syringe, handscoen, a plastic tray, an operating table, a sterile drape, a corrugation, a bend, a set of minor surgical tools, a temporary tissue storage place before making histopathological preparations, a closed glass jar containing 10% formalin, and a small operating table, Tissue tex processor, microtome, waterbath, incubator, Dot slide microscope lighting Olympus XC 10 and Olyvia software.

The research procedures included: (1) Acclimatization of Experimental Animals. White mice were allowed to adapt to their environment for 1 week and fed a standard diet. After this process, the mice were randomly assigned to five research groups. (2) Maintenance of Experimental Animals. White mice were kept in 40x30x20 cm plastic cages, covered with wire mesh and lined with 1.5-2 cm of rice husks, which were changed every two days. Food and water were provided in the cages. White mice were fed a standard diet of 40 grams per day per mouse. Water was provided in a special container ad libitum. The room temperature was maintained at a constant 20-25°C. (3) Ovariectomy Procedure. The ovariectomy procedure was performed after the acclimatization process, on the 8th day, using ketamine 40-80 mg/kgBW (intraperitoneal) and xylazine 5-10 mg/kgBW (intraperitoneal) anesthesia. (4) Procedure for Administering Jicama Extract (Pachyrhizus erosus) to Experimental Animals Jicama extract was administered in three dosage categories to each treatment group: P1 at a dose of 70 mg/200

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g BW, P2 at a dose of 140 mg/200 g BW, and P3 at a dose of 280 mg/200 g BW. The jicama extract was administered for 14 days, adapted to the human reproductive cycle, which was then converted to the reproductive cycle of mice. (5) Sampling Procedure. The surgical procedure for organ sampling was performed after 14 days of treatment. Mice were anesthetized with 1% ketamine at a dose of 0.2 mL per mouse, administered intramuscularly, and the anesthetic was allowed to take effect. After ensuring the mice were immobile, the uterus was carefully removed by cutting the isthmus of the left and right fallopian tubes (the part closest to the uterus) and the caudal portion (the boundary between the cervix and uterus). The uterus was then cleaned using a 0.9% NaCl solution, then dried using filter paper with one pressure. Once the uterus had begun to dry, it was weighed and then separated into left and right uterine grafts. The uterus was immediately placed in a bottle containing 10% buffered formalin fixative and soaked for 12-24 hours. (6) Tissue Processing. The uterine tissue was then cut into 2-3 mm thick sections using 10% buffered formalin. The uterine tissue was then placed in 10% formalin and processed in a Tissue Tex Processor for 90 minutes. Paraffin-embedding of the uterine tissue was carried out according to the tissue code. The uterine tissue was then sectioned using a microtome at a thickness of 3-5 microns. Deparaffinization was performed by placing the uterine tissue in an oven for 30 minutes at a temperature of 70-80°C. The hydration process was carried out by placing it in two tubes of xylene solution for 3 minutes each, and finally, immersing it in running water for 15 minutes. Apoptosis index examination using Tunnel Assay which includes several stages, namely the rehydration process, specimen permeabilization, inactivation of endogenous peroxidation, equilibration, reaction labeling process, labeling reaction termination process, blocking, detection, development, and counterstain and storage.

Data analysis in this study used the first statistical test carried out, namely the Normality test using Shapiro Wilk because the number of samples <50. If the significance value or p-value > 0.05 then it can be concluded that the data is normally distributed so that it meets the rules for parametric testing. Next, a homogeneity test is carried out using Levene's test (F test). If the significance value or p value > 0.05 then it can be concluded that the research group is homogeneous then the next test can be carried out, namely a comparative test using the One Way Anova test. The entire process in this study complied with ethical principles in accordance with the permission of the ethical committee of Research Institut Universitas STRADA Indonesia, with a letter No. 0823411/EC/KEPK/I/06/2025.

RESULTS

The requirements for conducting parametric tests are that the data must be normally distributed and homogeneous. The Shapiro-Wilk test for normality was used because the sample size was <50. The decision criterion was that if the significance value or p-value >0.05, the data would be normally distributed. The results of the data normality test are presented in the table below:

Table 1. Normality Test of Apoptosis Index and Endometrial Thickness Data of White Rats (Rattus norvegicus)

Variable	Group	*p-value*	Distribution
Apoptosis Index	Negative Control (KN)	0,608	Normal
	Positive Control (KP)	0,502	Normal
	Jicama Ekstract 1 dosis 70 mg/200 g BB (KP1)	0,456	Normal
	Jicama Ekstract 2 dosis 140 mg/200 g BB (KP2)	0,201	Normal
	Jicama Ekstract 2 dosis 280 mg/200 g BB (KP3)	0,105	Normal

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	Negative Control (KN)	0,678	Normal
	Positive Control (KP)	0,123	Normal
	Jicama Ekstract 1 dosis 70 mg/200 g BB (KP1)	0,367	Normal
Endometrial Thickness	Jicama Ekstract 2 dosis 140 mg/200 g BB (KP2)	0,489	Normal
	Jicama Ekstract 2 dosis 280 mg/200 g BB (KP3)	0,237	Normal

Description: If the p-value > 0.05 then the data is normally distributed and if the p-value < 0.05 then the data is not normally distributed.

Next, a data homogeneity test was performed to determine the variance of the data across all groups. The decision criterion for the homogeneity test is that if the significance value or p-value is > 0.05, the data is considered homogeneous. The results of the data homogeneity test are presented in the following table:

Table 2. Homogeneity Test of Apoptosis Index and Endometrial Thickness Data of White Rats (Rattus norvegicus)

Variable	p-value	Information
Apoptosis Index	0,166	Homogen
Endometrial Thickness	0,320	Homogen

Description: If the p-value > 0.05 then the data is homogeneous and if the p-value < 0.05 then the data is not homogeneous.

Based on the results of the normality and homogeneity tests of the data in the table above, the two research variables, namely the apoptosis index and endometrial thickness, all have normally distributed and homogeneous data, thus fulfilling the requirements for conducting parametric tests.

The Effect of Jicama Extract on the Apoptosis Index of Endometrial Epithelial Cells in Ovariectomized White Rats

The apoptosis index was observed using the TUNNEL method on endometrial epithelial cells, which appeared brown. Observations were made using an Olympus microscope at 1000x magnification. The average number of apoptotic cells was calculated from 20 fields of view.

The following is an overview of the TUNNEL Assay results from various groups:

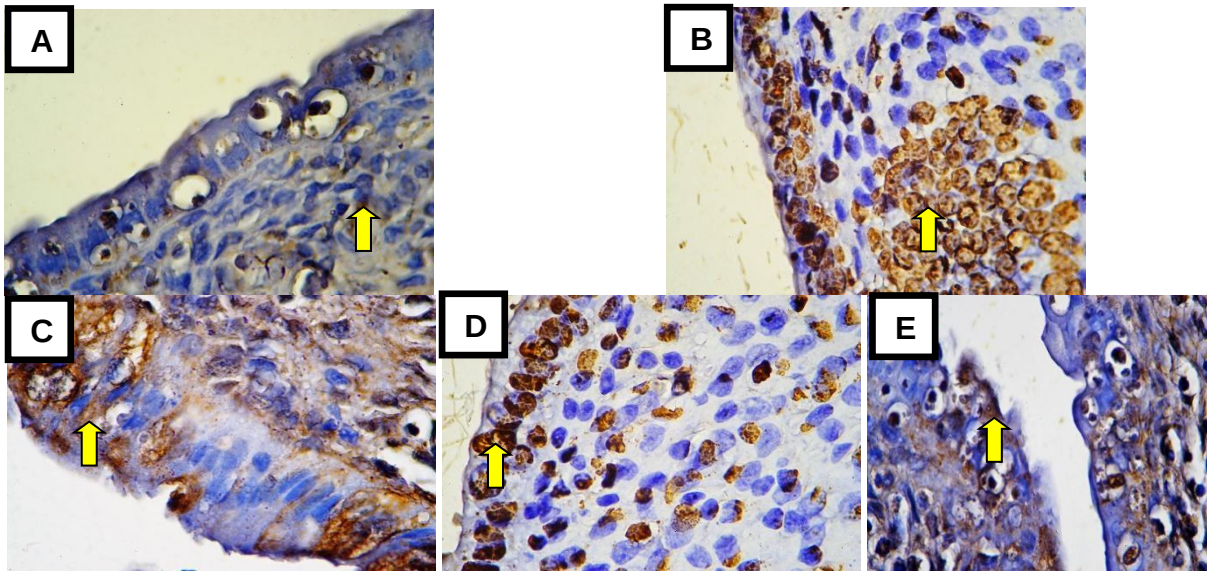

Figure 5.1 Apoptosis in Endometrial Epithelial Cells

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Description: **A**. Endometrial epithelial cells of the negative control group showed the least apoptosis compared to all groups; **B**. The positive control group (ovariectomy without jicama extract) experienced the most apoptosis compared to all groups; **C**. The jicama extract group with a dose of 70 mg/200 g BB experienced less apoptosis than the positive control group and more than the jicama extract group with a dose of 140 mg/200 g BB and 280 mg/200 g BB; **D**. The jicama extract group with a dose of 140 mg/200 g BB experienced less apoptosis than the positive control group and the jicama extract group with a dose of 70 mg/200 g BB and more than the control group with a dose of 280 mg/200 g BB; E. The group with a dose of 280 mg/200 g BB of jicama extract experienced less apoptosis compared to the positive control group, the group with a dose of 70 mg/200 g BB of jicama extract and the group with a dose of 280 mg/200 g BB of jicama extract.

The endometrial epithelial cell apoptosis index was calculated by dividing the number of brown endometrial epithelial cells, indicating DNA fragmentation, by the total number of cells observed in each field of view. The number of apoptotic cells was counted in 20 fields of view at 1000x magnification. The number of counted cells was marked using raster image software.

Furthermore, the apoptosis index calculation data was analyzed using the SPSS 20 program. Based on the results of the One Way ANOVA test, a significant difference was obtained in the average apoptosis index of endometrial epithelial cells in the five treatment groups, this is indicated by the p-value = 0.000 < α. Next, a post-hoc LSD (Least Significant Difference) test was conducted to determine which groups had significant differences. The results of the One Way ANOVA and LSD tests are presented in the table below:

Table 3. Effect of Jicama Extract on the Apoptosis Index of Endometrial Epithelial Cells in Ovariectomized White Rats

Treatment Group	N	Apoptosis Index	
		Mean ± Stand.dev	*p-value*
Negative Control (KN)	6	13,0 ± 4,69[a]	0,000 < α
Positive Control (KP)	6	38,56 ± 6,53[b]	
Jicama Ekstract 1 dosis 70 mg/200 g BB (KP1)	6	25,47 ± 3,67[c]	
Jicama Ekstract 2 dosis 140 mg/200 g BB (KP2)	6	19,0± 2,45[c]	
Jicama Ekstract 2 dosis 280 mg/200 g BB (KP3)	6	15,09 ± 3,01[a]	

In table 5.3 based on the results of the LSD test, it shows that there is a significant difference in the average apoptosis index between the positive control group (ovariectomy without jicama extract) with treatment group 1 (ovariectomy + jicama extract dose 70 mg/200 g BB), treatment group 2 (ovariectomy + jicama extract dose 70 mg/200 g BB) and treatment group 3 (ovariectomy + jicama extract dose 140 mg/200 g BB). However, there was no significant difference between treatment group 3 (ovariectomy + jicama extract dose 280 mg/200 g BB) and treatment group 2 (ovariectomy + jicama extract dose 140 mg/200 g BB). In addition, there was no significant difference between the negative control group and treatment group 3 (ovariectomy + jicama extract dose 280 mg/200 g BB).

The Effect of Jicama Extract on Endometrial Thickness in Ovariectomized White Rats

Endometrial thickness was measured using the HE (Hematoxylin Eosin) method, where the HE-stained slides were then scanned using an OLYMPUS XC10 microscope. Endometrial thickness was measured using a dot slide, taking 10 points (5 points with the highest thickness and 5 points with the lowest thickness) and then averaged at 200x magnification. The endometrial thickness measurement results can be accessed using Olyvia software.

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The following image shows the endometrial thickness of all treatment groups:

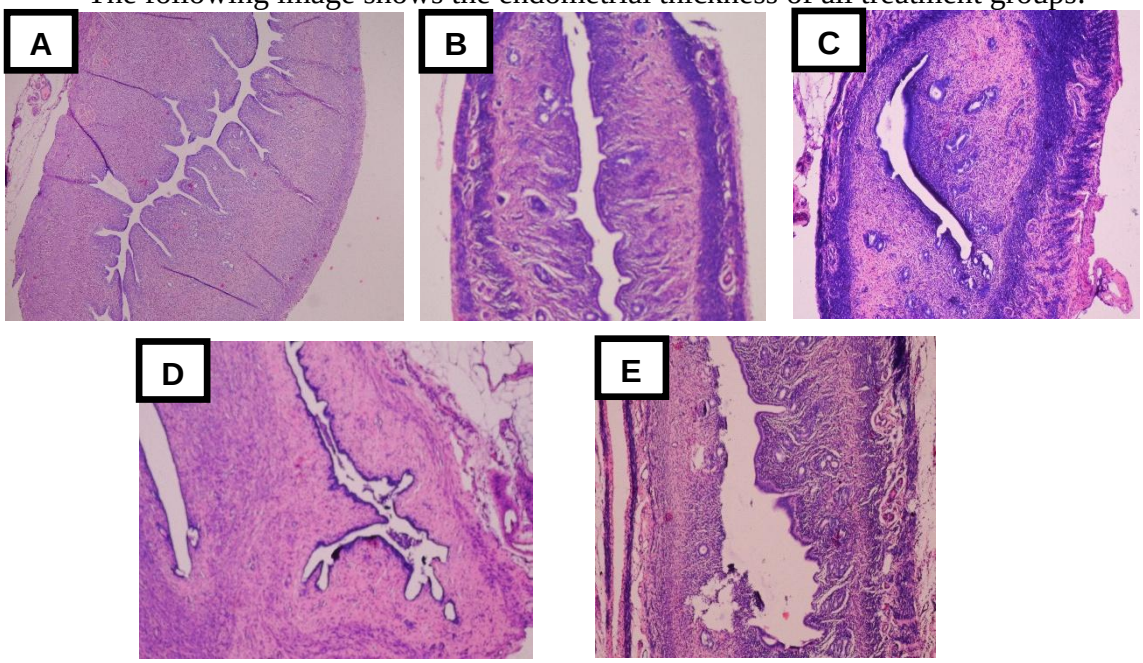

Figure 5.2 Endometrial Thickness

Description: **A**. Endometrial Thickness of Negative Control Group (not ovariectomized); **B**. Endometrial Thickness of Positive Control Group (diovariectomized without giving jicama extract); **C.** Endometrial Thickness of Treatment Group 1 (diovariectomized + jicama extract dose 70 mg/200 g BB); **D**. Endometrial Thickness of Treatment Group 2 (diovariectomized + jicama extract dose 140 mg/200 g BB); **E.** Endometrial Thickness of Treatment Group 3 (diovariectomized + jicama extract dose 280 mg/200 g BB)

The results of the endometrial thickness calculation were analyzed using SPSS 20. Based on the One Way ANOVA test, it was found that there was a significant difference in the average endometrial thickness of the five treatment groups, this was indicated by the p-value = 0.000 < α. Then, a post-hoc LSD (Least Significant Difference) test was conducted to determine which groups had significant differences.

The results of the One Way Anova and LSD tests are presented in the table below:

Table 4. Effect of Jicama Extract on Endometrial Thickness of Ovariectomized White Rats

Treatment Group	N	Ketebalan Endometrium	
		Rerata ± Stand.dev	*p-value*
Negative Control (KN)	6	345,63 ± 4,67[a]	0,000 < α
Positive Control (KP)	6	156,77± 3,43[b]	
Jicama Ekstract 1 dosis 70 mg/200 g BB (KP1)	6	167,89 ± 7,56[c]	
Jicama Ekstract 2 dosis 140 mg/200 g BB (KP2)	6	179,81 ± 7,52[c]	
Jicama Ekstract 2 dosis 280 mg/200 g BB (KP3)	6	356,57 ± 5,13[a]	

Description: LSD test results are shown in the mean ± Std column. Deviation if it contains different letters means there is a significant difference (p-value < 0.05) and if it contains the same letters it means there is no significant difference (p-value> 0.05).

Based on table 5.4 above, the results show that there is a significant difference in the average endometrial thickness between the positive control group (ovariectomy without jicama extract) and treatment group 1 (ovariectomy + jicama extract dose 70 mg/200 g BB), treatment

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group 2 (ovariectomy + jicama extract dose 140 mg/200 g BB) and treatment group 3 (ovariectomy + jicama extract dose 280 mg/200 g BB). However, there is no significant difference between treatment group 1 (ovariectomy + jicama extract dose 70 mg/200 g BB) and treatment group 2 (ovariectomy + jicama extract dose 140 mg/200 g BB). In addition, there is no significant difference between the negative control group and treatment group 3 (ovariectomy + jicama extract dose 280 mg/200 g BB).

DISCUSSION

Based on the results of the One Way Annova test calculation, the p-value = 0.000 <0.05 was obtained, so it can be concluded that there is a significant difference in the average apoptosis index of the five sample groups. Furthermore, a multiple comparison test with the LSD test was carried out, resulting in a significant difference in the average apoptosis index between the positive control group and treatment group 1 (ovariectomy + dose of 70 mg / 200 g BB), treatment group 2 (ovariectomy + dose of 140 mg / 200 g BB) and treatment group 3 (ovariectomy + dose of 280 mg / 200 g BB) with the lowest average apoptosis index value in the jicama group 3 (ovariectomy + dose of 280 mg / 200 g BB). In addition, there was no significant difference between the negative control group and treatment group 3 (ovariectomy + dose of 280 mg / 200 g BB). Thus, it can be concluded that there is an effect of giving jicama extract on the apoptosis index of ovariectomized white rats (Rattus norvegicus). At a dose of 280 mg/200 g BW, the average apoptosis index was close to normal, suggesting that anthocyanin administration at a dose of 280 mg/200 g BW was the optimal dose in this study. This study used jicama, which is an easily available, inexpensive, and affordable estrogen. Therefore, the hypothesis in this study was proven that administration of jicama extract can reduce the apoptosis index at the optimal dose of 280 mg/200 g BW, so it is possible that no further dose increase is necessary. The limitations of this study are that it used experimental animals, so an appropriate dosage for use in humans has not yet been established.

Oxidative stress plays an integral part of the aging process. Oxidative stress occurs when there is an imbalance between free radicals and antioxidants. Oxidative stress causes DNA damage, lipid peroxidation, and protein peroxidation. This triggers the release of mitochondrial proteins into the cytoplasm. This release of mitochondrial proteins into the cytoplasm initiates apoptosis (Monroe et al., 2002). One way to reduce oxidative stress is by administering antioxidants extracted from tropical plants. One bioactive ingredient that can be used as an antioxidant to reduce oxidative stress is jicama extract.

Jicama has a wide variety of uses, both for food and other purposes. In culinary terms, jicama is often consumed as a fresh fruit, in rujak (traditional fruit salad), or in salads due to its crunchy texture and distinctive sweet flavor. Jicama tubers have a high water content and are often used as a natural coolant, making them popular as a refreshing snack in hot tropical regions. Furthermore, jicama is also processed into flour, which is used as a base ingredient in processed foods such as cakes and chips. In the world of health, jicama is known as a source of natural phytoestrogens, compounds that have a chemical structure and function similar to the hormone estrogen (Lukitaningsih & Holzgrabe, '2013). This phytoestrogen content makes jicama often studied as a natural ingredient to treat various hormone-related health problems, such as menopausal symptoms and the side effects of hormonal contraceptives. Research shows that jicama contains isoflavones, especially daidzein and genistein, which play an important role in modulating the activity of estrogen receptors in the body (Harris, 2005). Besides being an easily accessible food source, jicama is also frequently used in both traditional and modern medicine due to its unique nutritional content. One of the active compounds of primary interest in this research is isoflavones, particularly daidzein and genistein, which act as primary phytoestrogens. Phytoestrogens are plant compounds with chemical structures and activities similar to the human hormone estrogen, making them a natural alternative in various hormonal

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therapies.

Based on research, jicama contains various important nutrients such as fiber, vitamin C, and several minerals that play a vital role in body health (Lukitaningsih and Holzgrabe, 2013). However, the main focus in this study is its bioactive compound content, specifically isoflavones. Isoflavones in jicama have a chemical structure similar to the estrogen hormone, known as 17-estradiol. Isoflavones in jicama, particularly daidzein and genistein, have estrogenic activity that can interact with estrogen receptors in the body. The daidzein content in jicama is reported to be 108.831 mg/100 grams, while genistein reaches 163.079 mg/100 grams (Lukitaningsih, 2009). This content is lower than that of soybeans, but has its own advantages because jicama does not contain high levels of purines, which can cause diseases such as gout in certain individuals (Nurhayati et al., 2023). Isothoflavones work by binding to estrogen receptors in the body, namely ERα and ERβ. In studies involving hypoestrogenous rat models, genistein and daidzein demonstrated different abilities to modulate the expression of these two receptors. Daidzein preferentially interacts with ERβ, an estrogen receptor closely linked to reproductive health and endometrial function (Harris, 2005). Genistein, on the other hand, has a broader ability to influence both, although its effects are highly dependent on concentration and the individual's hormonal status.

Research by Mayasari Putri Ardela (2024) using mice as a model animal showed that administration of ethanol extract of jicama significantly increased estrogen levels and ER~ expression. This study indicates that phytoestrogens in jicama not only have estrogenic activity, but also help improve endometrial function by improving the hypoestrogen condition induced by DMPA. In the hypoestrogen model mice, estrogen levels increased significantly along with the dose of jicama extract administered. The best results were achieved with a dose of 280 mg/200 g body weight of mice, which produced the highest estrogen levels and the most dominant ER~ expression (Ardela et al., 2024). Other studies have shown that the aging process increases oxidative stress, resulting in cell dysfunction or cell death in various organs, including Leydig cells in the testes. These changes lead to decreased testosterone production, which can lead to prostate atrophy by activating the critical apoptosis signaling pathway. Furthermore, decreased circulating testosterone levels increase oxidative stress, which in turn induces cell death or apoptosis. This apoptosis can lead to prostate weight loss (Jang, Hoon, et.al, 2013). Due to the accumulation of free radicals while the body is not protected by sufficient endogenous antioxidants, additional exogenous antioxidants are needed to maintain optimal cell function to help protect the body from oxidative stress and suppress aging (Rahman.K., 2007). A study by Jang (2013) found that administering an anthocyanin supplement derived from black soybean seed extract to the prostate of andropause-model rats at a dose of 160 mg/kg body weight reduced oxidative stress, as indicated by increased serum SOD activity and a decreased apoptosis index in the prostate. However, the decrease in the apoptosis index after anthocyanin administration in this study was not accompanied by an increase in prostate weight (Jang, Hoon, et.al, 2013).

The hypothesis in this study was proven, namely that the administration of jicama extract was able to reduce the apoptosis index in the endometrium of ovariectomized white rats. In this study, jicama acted as an endogenous antioxidant that was able to reduce oxidative stress that arises due to aging factors. The administration of anthocyanin at a dose of 70 mg/200 g BW in this study was able to reduce the apoptosis index in the endometrial epithelial cells of ovariectomized white rats, but the optimal dose that produced the lowest average apoptosis index to near normal conditions in this study was a dose of 280 mg/200 g BW.

Based on the results of statistical analysis using the One Way Annova test, the p-value = 0.000 was obtained, which means <α was obtained so that it can be concluded that there was a significant difference in the average endometrial thickness of the five treatment groups. Then continued with a multiple comparison test with the LSD test showed that there was a significant

DOI: https://doi.org/10.30994/sjik.v14i2.1286
ISSN: 2252-3847 (print); 2614-350X (online) Vol.14 No.2 November 2025 Page. 204-215

difference in the average endometrial thickness between the positive control group (diovariectomy without anthocyanin administration) with treatment group 1 (ovariectomy + dose 70 mg / 200 g BB), treatment group 2 (ovariectomy + dose 140 mg / 200 g BB) and treatment group 3 (ovariectomy + dose 280 mg / 200 g BB) with the highest average endometrial thickness value in treatment group 3 (ovariectomy + dose 280 mg / 200 g BB). However, there was no significant difference between the negative control group and treatment group 3. This shows that there is an effect of administering bengkoang extract on the endometrial thickness of ovariectomized white rats (Rattus norvegicus), especially at the highest dose in this study, namely 280 mg/200 g BB, which was able to increase the endometrial thickness to near normal conditions.

This is consistent with research conducted by Nur Laili (2015), which found that administering purple eggplant ethanol extract, which also contains anthocyanins, to white mice can increase endometrial thickness. The optimal dose in this previous study, which demonstrated increased endometrial thickness, was at a high dose of 99 mg/200 g of mouse body weight. Fernandez's (2015) study on the effect of dragon's tail leaf extract (Rhaphidophora pinnata, Schott), which is a flavonoid compound in the same group as jicama, on the uterine development of ovariectomized female mice (Mus musculus), showed a 38.7% increase in endometrial thickness and a 30.3% increase in uterine diameter. The optimal dose used in this study was 100 mg/kg body weight. During menopause, a state of hypoestrogenism occurs, resulting in increased oxidative stress (Doshi et al., 2013). In this study, the oxidative stress studied was in the endometrium, namely changes in endometrial thickness due to the regulation of estrogen hormone changes. Estrogen is a powerful antioxidant present in the body. Estrogen is a second-order antioxidant. Estrogen and its metabolites have antioxidant properties due to the presence of OH at C3 of the phenolic ring in the A position. The antioxidant activity of estrogen is carried out by estrogen molecules acting as free radical scavengers, neutralizing excess ROS, and increasing the number of antioxidant molecules such as thioredoxin and SOD. In addition, estrogen can act to trap transition metals, such as Fe2+ and Cu+, reducing the formation of Fe2+ and Cu+ into oxidants again (Rodriguez, 2015).

This study demonstrated that administering jicama extract increased endometrial thickness in ovariectomized white rats. In this study, jicama extract acted as an antioxidant, capable of scavenging free radicals, particularly in the endometrium of ovariectomized rats. The optimal dose resulting in the highest average endometrial thickness in this study was 280 mg/200 g body weight.

CONCLUSION

Based on the research results, it can be concluded that there is an effect of administering bengkoang extract on reducing the apoptosis index and increasing endometrial thickness in white rats in the ovariectomy model. The optimal dose used in this study was 280 mg/200 BW, which was proven to reduce the apoptosis index and endometrial thickness. Jicama can be used as an alternative natural ingredient containing phytoestrogens to overcome complaints that occur in menopausal women. It is hoped that the results of this study can be used as a guideline for making bengkoang capsules so that they can be consumed directly by menopausal women.

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