Effectiveness of Mahkota Dewa (*Phaleria macrocarpa*) Fruit Ethanol Extract on Alloxan-Induced Diabetic Rats (*Rattus norvegicus*)

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ABSTRACT

Diabetes mellitus is one of the chronic diseases in which the management of diabetes mellitus requires multidisciplinary treatment that includes non-drug and drug therapy and is carried out continuously. Mahkota dewa (*Phaleria macrocarpa*) is a plant used in traditional medicine because it contains various bioactive compounds that have been studied for their potential antidiabetic effects. This study aims to determine blood glucose levels, ovarian histopathology, and hematological profile of rats (*Rattus norvegicus*) induced by alloxan after being given ethanol extract of mahkota dewa fruit (*Phaleria macrocarpa*) and Metformin. This study used an experimental method with a completely randomized design (CRD) with 4 treatments and 6 repetitions, namely negative control without being induced by alloxan (K0), positive control only induced by alloxan at a dose of 150 mg/kgBB (K1), induced by alloxan at a dose of 150 mg/kgBB and treated using ethanol extract of mahkota dewa fruit (P1), and induced by alloxan at a dose of 150 mg/kgBB and treated by Metformin (P2). Treatment with ethanol extract and Metformin was done orally using a round. The treatment was done twice a day for 10 days. The data obtained were analyzed with One Way ANOVA and continued with Post Hoc test Duncan test. The treatment of ethanol extract of mahkota dewa fruit can significantly reduce blood glucose levels, improve hematological profiles such as the number of erythrocytes, leukocytes, hemoglobin levels and improvements in ovarian histopathology that experience follicular atresia, pycnosis, cariorexis, cariolysis, and increase the number of antral and preantral follicles.

1. Introduction

Diabetes mellitus is a chronic disease that can be suffered for life (Sihotang, 2017). Diabetes mellitus (DM) is caused by metabolic disorders that occur in the pancreatic organ characterized by an increase in blood sugar or often referred to as hyperglycemia conditions caused by a decrease in the amount of insulin from the pancreas. (Saputri, 2016). According to the International Diabetes Federation (IDF) report, the number of people with type 1 diabetes in Indonesia reached 41.8

thousand people in 2022. This figure makes Indonesia the country with the most type 1 diabetics in ASEAN, as well as 34th out of 204 countries on a global scale (Ahdiat, 2023).

Hyperglycemic conditions in the female reproductive system can affect the function of ovarian follicles. Hyperglycemic conditions affect glucose transport in the ovaries and the production of estrogen produced by ovarian follicles so that estrogen levels in the body become low (Cox et al. 1994). Diabetes mellitus is a chronic disease in which the management of diabetes mellitus requires multidisciplinary treatment that includes non-drug and drug therapy and is carried out continuously. Currently, the Indonesian government encourages people to consume traditional medicine because of its low side effects (Ernawati, 2013).

Mahkota dewa (*Phaleria macrocarpa*) is a plant that contains various bioactive compounds that have been studied for their potential antidiabetic effects. Mahkota dewa contains several active substances such as alkaloids, saponins, flavonoids, and polyphenols (Fatmawati et al., 2019). The saponin content in Mahkota dewa fruit acts as an antibacterial, antiviral, immune system booster and vitality enhancer, blood glucose level controller, and blood clot reducer (Prapti and Desty, 2013).

Research on mahkota dewa fruit peel extract (*Phaleria macrocarpa*) has been conducted by Candrarisna and Kurnianto (2018), which is only the effect of mahkota dewa fruit peel extract on the hematological profile of diabetic rats. Dzannastia et al. 2021 conducted research on the effect of red turi leaf extract (Sesbania grandiflora) on hematological values in diabetic male mice. Research on the effects of hyperglycemic conditions on the reproductive system has been conducted by Mechiavel et al. (2023), that *Cinnamomun burmannii* stem bark can increase testicular weight and the number of spermatozoa in diabetes mellitus mice. Based on the above results, further research was conducted on "Effectiveness of Mahkota Dewa (*Phaleria macrocarpa*) Fruit Ethanol Extract on Alloxan-Induced Diabetic Rats (*Rattus norvegicus*)" especially glucose levels, rat hematology, and ovarian histopathology.

2. Methodology

2.1 Time and place of research

The research was conducted from December 2023 to January 2024. The research was conducted in the Experimental Animal Maintenance Room of the Biology Study Program, Faculty of Mathematics and Natural Sciences, Udayana University, Bukit Jimbaran Campus as the maintenance of experimental animals. Preparation and examination of ovarian histology incision preparations were carried out at the Parasitology Laboratory, Denpasar Veterinary Center, Jalan Raya Sesetan No.266, Denpasar, Bali.

2.2 Methods

Research design

This study used a completely randomized design (CRD) with rat samples divided into 4 treatment groups, namely negative control without alloxan induction (K0), positive control only induced alloxan at a dose of 150 mg/kgBB (K1), induced alloxan at a dose of 150 mg/kgBB and treated using ethanol extract of mahkota dewa fruit (P1), and induced alloxan at a dose of 150 mg/kgBB and treated using *Metformin* (P2). Each group consists of 6 repetitions or 6 white rats (*R. norvegicus*).

Preparation of ethanol extract of mahkota dewa fruit (*P. macrocarpa*)

Extraction was carried out by maceration method. To make 50% ethanol extract, thick extract from mahkota dewa fruit weighing 50 mg was measured and mixed with 0.1% Sodium carboxymethyl cellulose (CMC-Na) solution as much as 1 mL, then sterile aquadest vehiculum was added until it reached a volume of 100 mL (Siswandono, 2014).

Dose determination

1. Extract dosage

Determination of the dose in the treatment of ethanol extract of mahkota dewa fruit (*Phaleria macrocarpa*) is a dose of 50%. Determination of the 50% dose is determined by the formula:

$$\%\ extract\ volume\ content: \frac{volume\ of\ viscous\ extract}{Total\ volume} \times 100\%$$

$$\%\ extract\ volume\ content: \frac{50\ mg}{100\ mL} \times 100\% = 50\%$$

2. Metformin dosage

The dose of Metformin used is the result of conversion from human to rat dose The process of converting human doses to rat doses can be calculated using the Laurance and Bacharach formula (1964), namely:

$$Initial\ volume\ \times human\ to\ rat\ conversion\ value = volume\ after\ conversion$$

$$500\ mg \times 0{,}018 = 9\ mg$$

Maintenance of test animals

Prior to the study, rats were acclimatized in laboratory conditions for 7 days and fed. White rats were fed with Hi-Gro 551 pig starter complete feed and drinking water ad libitum. The initial body weight of white rats was weighed before treatment. Weighing was done twice, at the beginning and at the end of the observation (Edward and Yerizel, 2009). After acclimatization, rats were induced with alloxan 150 mg/kgBB intravenously. After induction, the rats were allowed to stand for 3 days. On the last day of alloxan induction, blood glucose levels were measured.

Provision of treatment

The first treatment (P1) rats were treated with 50% ethanol extract of mahkota dewa fruit (*Phaleria macrocarpa*). The volume of each administration is 2 mL. The second treatment (P2) rats were treated with 9 mg dose of Metformin. The administration of treatment with ethanol extract and Metformin was done orally using a round. The treatment was done twice a day for 10 days.

Determination of blood glucose levels

Blood glucose levels were determined by the glucose axidase biosensor method, using the One Touch Ultra device (blood glucose monitoring device, manufactured by Lifescan Johnson & Johnson Company 2002). Blood was taken from the tail of the rat by cutting the tip of the rat's tail.

Glucose measurements were taken 3 times, at the beginning before treatment, after alloxan induction, and at the end after treatment.

Hematology examination

Mouse blood was taken through the eye as much as 3 mL for hematological examination. Hematology examination included counting the number of erythrocytes, number of leukocytes, hemoglobin levels using Rayto RT-7600® Auto hematology analyzer (Rayto ltd, Shenzen, China).

Observation of ovarian histology incision

After making preserved preparations of ovarian histology incisions, observations were made under a light microscope with a magnification of 400 and 1000x. then observed the entire follicle by distinguishing between antral and preantral follicles and counting the total number of follicles in one field of view. In addition, it was observed by distinguishing the shape of cells and cells that underwent necrosis or cell death.

2.3 Data analysis

The data obtained were analyzed using one-way analysis of variance (One Way ANOVA) with Statistical Product and Service Solutions (SPSS) 26. If the results of the analysis of variance showed a significant difference, it was continued with the Duncan's test (BNT). Qualitative data were analyzed descriptively by observing changes in ovarian histology structure.

3. Result and Discussion

3.1 Result

Blood glucose

The results showed that the average final blood glucose level of K0 was 93.67 mg/dL, K1 was 197.17 mg/dL, P1 was 101.33 mg/dL, and P2 was 83.50 mg/dL. After that, Duncan's post hoc test was continued to find out which group had a significant difference. The results obtained were that there was a significant difference in blood glucose levels ($P < 0.05$) in the positive control group (K1) with K0, P1, and P2. Groups that did not have significant differences ($P > 0.05$) were between the K0 group with the P1 and P2 groups (Table 1).

Table 1. Mean blood glucose levels in each treatment group

Treatment	Blood glucose (mg/dL)		
	Initial	Induction	End
K0	93.67 ± 7.763^{a}	93.67 ± 7.763^{a}	93.67 ± 1.767^{a}
K1	86.83 ± 12.734^{a}	218.50 ± 61.773^{b}	197.17 ± 2.404^{b}
P1	94.33 ± 16.342^{a}	324.17 ± 136.813^{b}	101.33 ± 7.659^{a}
P2	83.00 ± 23.048^{a}	294.00 ± 85.133^{b}	83.50 ± 6.504^{a}

Notes: Different letter notations in the same column indicate significant differences ($P \leq 0.05$) Numbers behind the sign (±) indicate standard deviation. Without induced alloxan, negative control (K0); only induced alloxan with a dose of 150 mg/kgBB (K1), induced alloxan with a dose of 150 mg/kgBB and treated using ethanol extract of mahkota dewa fruit (P1), and induced alloxan with a dose of 150 mg/kgBB and treated using Metformin (P2).

Hematology

Hematology values of white rats include parameters of hemoglobin (Hb) levels, erythrocyte counts (RBC), and leukocyte counts (WBC). Based on Table 4 hemoglobin (HB) levels in K0 12.7 g/dL, K1 11.1 g/dL, P1 13.5 g/dL, and P2 12.25 g/dL. In the Duncan test, the groups that had significant differences ($P < 0.05$) were between K1 with K0, P1, and P2; P2 with K1 and P1; P1 with K1 and P2. While P1 with K0 and P2 with K0 showed no significant difference ($P > 0.05$). This means that the administration of mahkota dewa fruit extract significantly increases hemoglobin levels (Table 2).

The number of leukocytes K0 is 7.65 x 103 cells/µL, K1 is 13.8 x 103 cells/µL, P1 is 6.05 x 103 cells/µL, and P2 is 7.4 x 103 cells/µL. after statistical analysis with Duncan's test, treatment groups that differ significantly ($P < 0.05$) are K1 with K0, P1, and P2. While K0, P1, and P2 are not significantly different ($P> 0.05$). The administration of mahkota dewa fruit extract can significantly reduce the number of leukocytes (Table 2).

The average number of erythrocytes in K0 is 8.55 x 106 cells/µL, K1 is 7.9 x 106 cells/µL, P1 is 7.94 x 106 cells/µL, and P2 is 8.44 x 106 cells/µL. After statistical analysis with Duncan's test, there was no significant difference ($P > 0.05$) in all treatment groups (K0, K1, P1, and P2). However, there was an increase in the number of erythrocytes after being treated with mahkota dewa fruit extract (Table 2).

Table 2. Mean hemoglobin (HB), erythrocyte count (RBC), and leukocyte count (WBC)

Treatment	Variables		
	HB (g/dL)	RBC (x 10^6/µL)	WBC (x 10^3/µL)
K0	12.7 ± 0.282^{bc}	8.55 ± 0.388^{a}	7.65 ± 1.767^{a}
K1	11.1 ± 0.707^{a}	7.9 ± 0.141^{a}	13.8 ± 2.404^{b}
P1	13.5 ± 0.141^{c}	7.94 ± 0.084^{a}	6.05 ± 0.212^{a}
P2	12.25 ± 0.212^{b}	8.44 ± 0.629^{a}	7.4 ± 0.424^{a}

Notes: (Hb) hemoglobin level; (RBC) erythrocyte count; (WBC) leukocyte count. Different letter notations in the same column indicate significant differences ($P \leq 0.05$) Numbers behind the sign (±) indicate standard deviation. Without induced alloxan, negative control (K0); only induced alloxan with a dose of 150 mg/kgBB (K1), induced alloxan with a dose of 150 mg/kgBB and treated using ethanol extract of mahkota dewa fruit (P1), and induced alloxan with a dose of 150 mg/kgBB and treated using Metformin (P2).

Ovarian Histopathology

The observation of ovarian histopathology of the negative control group (K0) which was not induced by alloxan and not treated with ethanol extract of mahkota dewa fruit and Metformin did not show any damage to the ovaries. While in the observation of ovarian histopathology of the positive control group (K1) which was induced by alloxan and not treated, damage was found in a number of follicles that were abnormal (atresia) and found pycnosis and reduction of granulosa cells. After being treated with mahkota dewa fruit extract (P1) and Metformin (P2), the ovary shape improved back to normal but the core of the oocyte or granulosa experienced pycnosis, karyolysis, and karyorrhexis. Comparison of the histopathological picture of rats for each treatment can be seen in Figures 1 and 2.

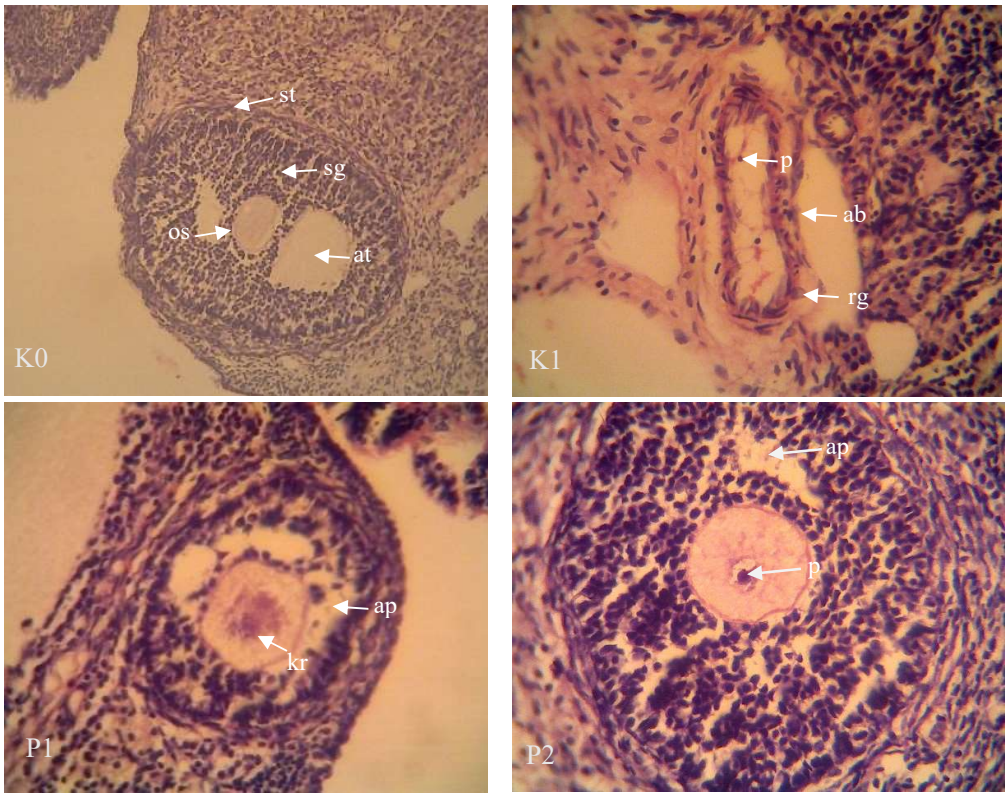

Figure 1. Tertiary follicle or antral follicle at 400x magnification with HE staining. Normal tertiary follicle (K0); atresia follicle (K1); tertiary follicle has karyorrhexis of oocyte nucleus (P1); follicle has pyknosis of oocyte nucleus (P2). Note: oocyte (os); antrum (at); granulosa cell (sg); teka cell (st); pycnosis (p); abnormal follicle (ab); reduction granulosa (rg); cariorexis (kr); apoptosis (ap).

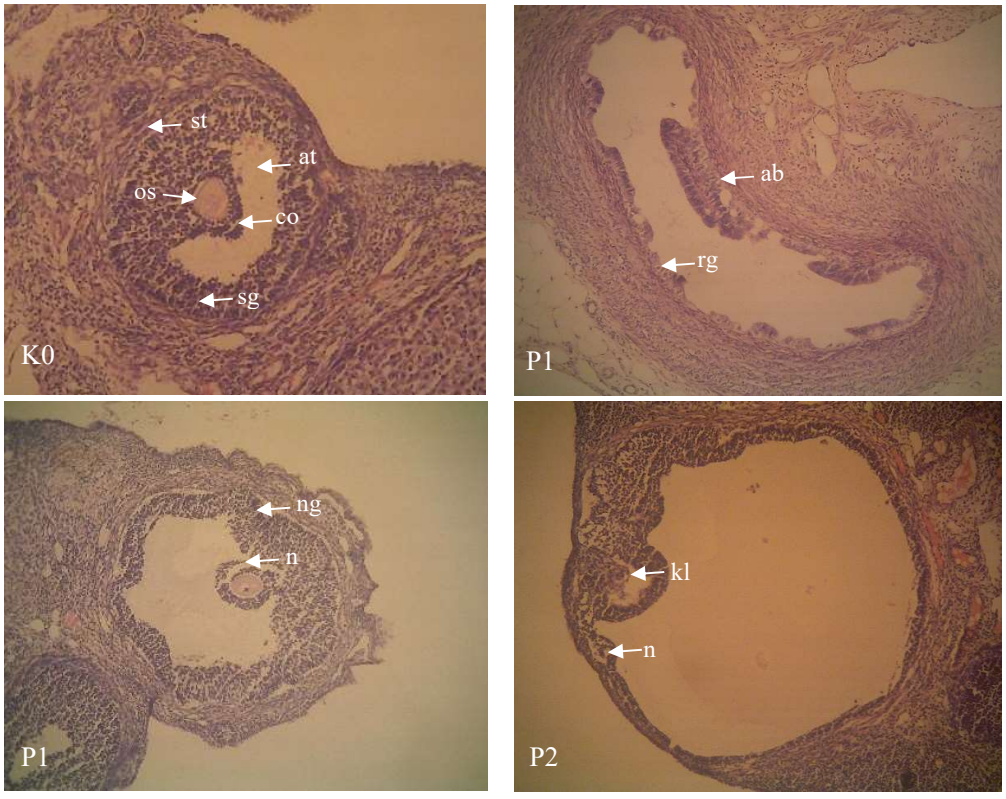

Figure 2. Normal de Graaf follicle at 100x magnification with HE staining. (K0); de Graaf follicle abnormal (K1); de Graaf follicle with repair (P1); de Graaf follicle with cariolysis in the oocyte nucleus (P2). Note: teka cell (st); granulosa cell (sg); oocyte (os); cumulus oophorus (co); antrum (at); cariolysis (kl); necrosis (n); abnormal follicle (ab); reduction granulosa (rg); necrosis (n).

Observation of ovarian histopathology on the number of preantral follicles and antral follicles. The criteria for preantral follicles are follicles that do not yet have an antrum. Starting from primordial, primary or secondary follicles that do not yet have an antrum, while antral follicles are follicles that have an antrum ranging from tertiary follicles to de Graaf follicles.

Calculation of the number of preantral and antral follicles is done by looking at the entire ovarian field of view. Then enlarged to ensure the criteria corresponding to preantral and antral follicles. Based on Table 5, the average number of preantral follicles in each treatment group is 8.83 follicles (K0), 4.83 follicles (K1), 7.50 follicles (P1), and 6.00 follicles (P2). After statistical analysis, the treatment groups that were significantly different were K0 with K1, P1, and P2; K1 with K0, P1, and P2, P1 with K0, K1, and P2; and P2 with K0, K1, and P1. The data above shows that ethanol extract of mahkota dewa fruit can increase the number of preantral follicles significantly better than Metformin, although it is still significantly different from the negative control group (K0) (Table 3).

The average number of antral follicles in each treatment group is K0 6 follicles, K1 1.83 follicles, P1 3.83 follicles, and P2 2.67 follicles. After the Duncan test was conducted to see which groups had significant differences, it was found that the groups that were significantly different ($P < 0.05$) were K0 with K1, P1, and P2; K1 with P1; and K0 with P1. While there are groups that are not significantly different ($P > 0.05$), namely groups K1 with P2 and P1 with P2. The data above shows that ethanol extract of mahkota dewa fruit can increase the number of antral follicles significantly better than Metformin, although it is still significantly different from the negative control group (K0) (Table 3).

Table 3. Mean and One Way Anova test results of the number of preantral and antral follicles

Treatment	Variables	
	Preantral Follicles	Antral Follicles
K0	8.83 ± 0.753^{d}	6.00 ± 0.894^{c}
K1	4.83 ± 0.753^{a}	1.83 ± 0.753^{a}
P1	7.50 ± 1.225^{c}	3.83 ± 1.427^{b}
P2	6.00 ± 0.894^{b}	2.67 ± 1.211^{ab}

Notes: Different letter notation in the same column indicates significant difference ($P \leq 0.05$) while the same letter notation indicates no significant difference ($P \geq 0.05$). Numbers behind the sign (±) indicate standard deviation. Without induced alloxan, negative control (K0); only induced alloxan with a dose of 150 mg/kgBB (K1), induced alloxan with a dose of 150 mg/kgBB and treated using ethanol extract of mahkota dewa fruit (P1), and induced alloxan with a dose of 150 mg/kgBB and treated using Metformin (P2).

3.2 Discussion

Blood glucose

Normal rat blood sugar levels are 50- 135 mg/dl and said to be hyperglycemic if the blood sugar levels of rats > 135 mg/dl (Rahman, 2014). In K1, P1, and P2 after alloxan induction there is an

increase in blood glucose levels until hyperglycemic conditions occur. Alloxan reacts by damaging essential components in pancreatic β-cells, which results in reduced insulin-transporting granules in these cells (Mursiany, 2019). Mahkota dewa fruit extract in this study can reduce blood glucose levels of alloxan-induced diabetic rats, hyperglycemic blood glucose levels from 324.17 to 101.33 mg/dL. Arjadi and Susatyo (2010) stated that the decrease in blood glucose levels due to the administration of mahkota dewa can be explained through two main mechanisms, namely intra pancreatic and extrapancreatic mechanisms. The intra-pancreatic mechanism works by repairing (regenerating) damaged pancreatic β-cells, protecting β-cells from damage, and stimulating insulin release through active compounds of alkaloids and flavonoids. Meanwhile, extra-pancreatic mechanisms can occur in various ways (Prameswari and Widjanarko, 2014).

Hematology

Diabetes can cause high oxidative stress in the body. This oxidative stress can damage red blood cells, causing red blood cells to break down faster and experience hemolysis (Dzannastia, 2023). Hyperglycemia also changes the properties of the erythrocyte membrane, causing an increase in erythrocyte osmotic fragility. This fragility causes the erythrocytes to break more easily and eventually lysed prematurely. Lysed erythrocytes cause the number to decrease so that the hemoglobin content in erythrocytes also decreases (Hanifa et al., 2023). The decrease in hemoglobin levels is influenced by an increase in glycated hemoglobin. Glycation of hemoglobin occurs when glucose in the blood binds to the hemoglobin protein. As a result of increased glucose levels in the blood, HbA1c levels in the blood become high (Eyth and Naik, 2022). Bindayel's research (2021) states that there is an inversely proportional relationship between HbA1c levels and Hb, so people with diabetes who have high HbA1c levels will have low Hb concentrations. HbA1c is the glycated form of hemoglobin, which is formed through a non-enzymatic pathway when hemoglobin is exposed to plasma glucose. HbA1c is a measure of the beta-N-1deoxy fructosyl component of hemoglobin. Glycation is a spontaneous nonenzymatic reaction that occurs when glucose is covalently linked to hemoglobin at the amino terminus of the b-globin chain. The result of the non-enzymatic reaction between reducing sugars, proteins, and other products will produce Advance glycation end products (AGEs). The accumulation of AGEs is associated with the progression and complications of diabetes (Chauhan, 2017).

Ethanol extract of mahkota dewa fruit in this study can increase hemoglobin levels in alloxaninduced diabetic rats. Anthocyanins contained in mahkota dewa fruit have benefits as antioxidants, anti-hyperglycemia, and anti-glycation. Anthocyanins can inhibit the glycation reaction between glucose and free amino. The inhibited amadori products will also inhibit the formation of AGEs. Anthocyanins can also inhibit some enzymes, such as aldose reductase which plays a role in the polyol pathway that converts glucose into sorbitol, which can increase the formation of AGEs. (Nugrahini et al., 2022).

The results showed that the number of leukocytes above normal was in the positive control group (K1) which was induced by alloxan without treatment. The increased number of leukocytes is caused by hyperglycemia. According to Gushiken et al. (2020) Hyperglycemia causes an increase in Reactive Oxygen Species (ROS) resulting in chronic inflammation with the mechanism of activating excessive cytokine release and stimulating leukocyte migration to inflammatory areas. In addition, during the inflammatory phase there is a release of cytokines which causes an increase in the number of leukocytes in the blood and tissue circulation (Ellis et al., 2018). Excessive

cytokine release causes leukocyte levels to increase. Increased leukocyte levels will stimulate the production of ROS. Excess ROS production can cause damage to surrounding cells (Gushiken et al., 2021). Mahkota dewa fruit extract in this study can reduce the number of diabetic rat leukocytes to normal. This is because the mahkota dewa fruit extract contains terpenoid compounds. Terpenoid compounds have anti-inflammatory activity by inhibiting cytokine activity. This will reduce the increase in leukocyte counts in blood circulation and tissues and suppress leukocyte production, keeping leukocyte counts within normal limits (Park et al., 2019).

Ovarian histopathology

Based on Figures 1 and 2, observation of ovarian histopathology in rats with 400x magnification shows that there are signs of necrosis (cell death) due to hyperglycemia from alloxan induction at a dose of 150 mg/kgBB. Based on the histopathology of the positive control group (K1) there are oocyte cells that experience the process of cell death. This is as shown that necrosis consists of three types, namely pycnosis, karyorexis, and karyolysis. Necrosis is usually characterized by a pycnotic nucleus which is a shrinkage of the nucleus as a result of cytoplasmic homogenization and eosinophilic increase. After pycnotic nucleation, the nucleus may disintegrate and leave fragments of chromatin substance scattered within the cell, this process is called karyorrhexis. Dead cell nuclei lose the ability to be colored and disappear, this process is called karyolysis (Hakimah et al., 2021).

Atretic follicles form during the reproductive period of mammals, the process is known as atresia. Atretic follicles are normal follicles that have degenerated. Atretic follicles are usually accompanied by pycnosis, reduction of granulosa cells due to proliferation and damage to the basement membrane (Lee et al. 2000). The attrition process can take place at any stage of follicular development characterized by the cessation of mitotic division of granulosa cells, detachment of granulosa cells from the basement membrane and oocyte death (Junquiera & Carneiro 1998). Follicular development requires interactions between granulosa cells as well as with germ cells involving endocrine, autocrine, paracrine and gap junction pathways. Thus the two-way relationship between oocytes and granulosa cells is very important for the process of follicle development to occur. According to Colton et al. (2003) reduced communication between granulosa cells causes changes in paracrine communication between oocytes and granulosa cells. In addition, follicle cells require nutrient supply through gap junctions for oocyte development. Gap junctions are specialized regions on the membranes of adjacent cells that allow communication between these cells (Granot & Dekel, 1998). Gap junctions play an important role in the development of granulosa cells. The types of gap junction proteins are known as connexins. Granulosa cells are known to express connexin-43 while in oocytes it is connexin-37. Hyperglycemia conditions cause a decrease in connexin-43 protein expression which affects intercellular communication in granulosa cells and increases the occurrence of apoptosis in these granulosa cells.

In this study, the administration of mahkota dewa fruit extract can regenerate granulosa cells that experience necrosis so that follicles do not experience atresia. The content of alkaloids, flavonoids, and vincristine (polyphenols) contained in mahkota dewa fruit is also an antioxidant that can capture hydroxy and superoxide radicals and then neutralize free radicals so as to protect cells and maintain the integrity of cell and tissue structures/cell regeneration against unwanted reactions (Redha, 2010). Hyperglycemic conditions in the female reproductive system can affect the number of ovarian follicles. Hyperglycemic conditions affect glucose transport in the ovaries and the production of estrogen produced by ovarian follicles so that estrogen levels in the body become low

(Cox et al. 1994). The proestrus phase is a phase of increasing estrogen hormones in the blood along with the development of primary follicles into de Graff follicles stimulated by the hormone FSH (Follicle Stimulating Hormone). FSH is a hormone secreted by the anterior pituitary that functions in the growth of ovarian follicles. FSH induces the secretion of estrogen and progesterone from the ovaries by activating aromatase and P450 enzymes, inducing granulosa cell proliferation and LH receptor expression and directing negative feedback on GnRH secretion (Wikayanti and Panjaitan, 2019). Mahkota Dewa fruit contains polyphenolic compounds including epicatechin (EC), epigalo catechin (EGC), epicatechin gallate (ECG), epigalocatechin gallate (EGCC), flavonoids, gallic acid and chlorogenic (Herawati and Hidayah 2012). Based on table 5, the administration of ethanol extract of mahkota dewa fruit in diabetic rats induced by alloxan can increase the number of follicles in the ovary both antral and preantral follicles. Catechins in mahkota dewa play a major role in inhibiting superoxide, hydrogen peroxide, hydroxyl radicals and nitric acid derived from various chemicals that enter the body. Catechins also cause catechol structures that inhibit the formation of free radicals, and cause an increase in LH and FSH. When the amount of FSH increases, the number of follicles in the ovaries will also increase (Sari et al., 2018).

4. Conclusion

Mahkota dewa (*Phaleria macrocarpa*) fruit extract can reduce hyperglycemic blood glucose levels to normal, improve hematological profiles in the form of hemoglobin levels and leukocyte counts, and increase the number of preantral and antral follicles in alloxan-induced diabetic rats.

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