

## EFFECT OF TRIGONA LAEVICEPS HONEY ON DECREASED GLUCOSE LEVELS IN MALE RATS (RATTUS NOVERGICUS)

Sri Ratna Dewi \*

Clinical Pathology Departement, Faculty of Medicine and Health Sciences, Warmadewa University,  
Jl. Terompong No.24, Sumerta Kelod, Kec. Denpasar Timur, Kota Denpasar

\*Correspondence: Sri Ratna Dewi [email: ratnasamuh86@gmail.com]

### ABSTRACT

Diabetes mellitus (DM) is a metabolic disease characterized by increased blood sugar levels (hyperglycemia) which occurs due to abnormalities in insulin secretion, insulin action or both. People use many herbal remedies, one of which is Trigona laeviceps honey. This study aims to determine the effect of giving Trigona laeviceps honey on reducing glucose levels in male rats (*Rattus norvegicus* L). This research was an experimental study using 28 Wistar rats (*Rattus norvegicus*) which were then analyzed by the Kruskal Wallis test to determine differences in glucose levels in the groups. The results showed that there was a decrease in glucose levels in the group given Trigona laeviceps honey and a significant difference in mean glucose levels was obtained based on the treatment group (p 0.000).

**Keywords :** *Trigona laeviceps* honey., DM., blood glucose.

### INTRODUCTION

Hyperglycemia or increased blood sugar caused by abnormalities in insulin secretion, insulin action or both is characteristic of diabetes mellitus (DM). This chronic hyperglycemia condition can cause complications of microangiopathy and macroangiopathy which lead to dysfunction or failure of multiple organs, especially the eyes, kidneys, nerves, heart and blood vessels.

These complications are based on inflammatory reactions triggered by oxidative stress which then induces oxidative tissue damage. This oxidative damage will cause an inflammatory reaction that induces the release of pro-inflammatory cytokines such as TNF- $\alpha$  which can induce kidney microvascular damage, causing progression of glomerular damage.<sup>2,3</sup>

There has been an increase in the prevalence of DM in Indonesia from 2013 by 2% based on doctors' diagnoses in residents aged  $\geq 15$  compared to 2018. To follow up on this, the government recommends efforts to control DM that has been diagnosed using several methods, including by regulating diet, exercise and using herbal alternatives.<sup>4</sup>

Honey is an alternative herbal that contains phenolics, namely caffeic, ellagic, ferulic and p-coumaric acids; flavonoids such as apigenin, chrysin, galangin, hesperetin, kaempferol, pinocembrin and quercetin; as well as antioxidants such as tocopherols, ascorbic acid, superoxidodismutase (SOD), catalase

(CAT), and reduced glutathione (GSH). Several studies show that honey can be used as an antioxidant, anti-inflammatory, antibacterial, antiviral, antiulcer, antihyperlipidemic, anti-diabetic and anticancer.<sup>5</sup>

Previous research shows that honey from stingless bees (*Trigona laeviceps*) can act as an anti-inflammatory, anti-cancer agent, antimicrobial and has antioxidant properties.<sup>6</sup> However, the benefits of "kele" honey (*Trigona laeviceps*) are not widely known and are considered as a dietary alternative for patients, including type 2 diabetes patients (having high inflammatory factors) due to the lack of systematic scientific studies to support its medical properties. On the other hand, the quality and efficacy of the honey produced is also influenced by the type of bee species, the location where the bees live, the season, and also the type of flower and the condition of the flowers pollinated by the bees. This study aims to determine the effect of giving Trigona laeviceps honey on reducing glucose levels in male rats (*Rattus norvegicus* L).

### MATERIALS AND METHODS

This research is a true experiment research with a "One-groups pretest-posttest design". This research will use Wistar rats (*Rattus norvegicus*) with the criteria being male rats, 2-3 months old, healthy and with normal activities, and weighing approximately 150-200 grams. The research has

been approved by the Animal Ethics Committees on Faculty of Veterinary Medicine, Udayana University with registration number B/166/UN14.2.9/PT.01.04/2021, August 16<sup>th</sup> 2021.

The instruments used in this research were sonde, cage, drinking water container, digital scale, 1 cc syringe, glucose examination tool (Easy touch GCU). The materials used are mineral water, rat food, STZ (streptozotocin), and glucose strips

The mice were divided randomly into four groups according to the treatment group, namely the control group with mice without hyperglycemia, the hyperglycemia group, the hyperglycemia group with a honey dose of 0.4ml/kg and the hyperglycemia group with a honey dose of 0.8 ml/kg. The number of male Wistar rats that will be used in this research is 28 male Wistar rats.

Several tests were carried out in this research, namely Qualitative and Quantitative Phytochemical Tests on honey with component analysis using the Fourier Transform Infra Red (FTIR) method. Next, the mice's body weight was measured, the hyperglycemia mouse model was induced, and they were given treatment according to the group. The data was then processed using statistical techniques, namely the data normality test was carried out using the Shapiro-Wilk test and the Kruskal Wallis test to determine significant differences.

## RESULTS

The data in table 1 shows that *Trigona laeviceps* honey contains alkaloids, flavonoids, phenols, tannins, saponins and steroids. With the highest phenol content being 456.431 mg GAE/100 g.

Table 1. *Trigona* Honey Phytochemical Test Results

No	Parameter	Result Test
Phytochemical screening		
1	Alkaloid	+
2	Flavonoid	+
3	Fenol	+
4	Tannin	+
5	Saponin	+
6	Steroid	+
Quantitative Test		
7	Flavonoid (mg QE/100 g)	94,414

8	Tannin (mg TAE/100 g)	23,707
9	Fenol (mg GAE/100 g)	456,431

Table 2. Average Rat Blood Glucose Levels Based on Day (mg/dl)

Group Treatment	Average Glucose Levels (days)		
	0	6	21
I	97,67	97,67	79
II	400,83	383	314,5
III	209,67	100,17	73,3
IV	197,5	114,83	77,67

Information:

I: Negative/normal control (given sterile distilled water orally)

II: Positive control (diabetes) (STZ 50 mg/kg BW subcutaneously)

III: *Trigona laeviceps* honey dose 0.27 ml/200mgBW

IV: *Trigona laeviceps* honey dose 0.54 ml/200mgBW

Table 3. Differences in Mean Blood Glucose Levels of Rats in the Honey Treatment Group (mg/dl)

Group Treatment	Average Levels	Normality	Kruskall Wallis Test
I	91,44±2,85	0,037	0,000*
II	371,67±15,4	0,264	
III	127,72±14,68	0,004	
IV	130±12,49	0,13	

Based on table 3, the mean blood glucose levels in the honey treatment group show that there is a difference in glucose levels in the control and honey treatment groups with a significance value of 0.000 ( $p < 0.005$ ).

## DISCUSSION

In this study, the initial blood glucose levels of 50 mg/200 mg Wistar rats which were induced before any treatment was given were normal (70-110 mg/dl). Then, a rat model of diabetes mellitus was created by administering subcutaneous doses of streptozotocin (STZ) to groups II, III and IV. After being adapted for 3 days by administering STZ, blood sugar measurements were carried out again in the mice and it was found that there was an increase in the blood sugar of the mice compared to the initial level of glucose or on day 0 which occurred in each treatment group given honey III and IV, as well as positive controls in the category diabetic blood glucose levels 150-200 mg/dl.

The male Wistar rat diabetes model was created using streptozotocin which can cause damage to  $\beta$  cells in the pancreas gland which can cause the rats to experience increased glucose levels. Streptozotocin works by binding to GLUT-2 which facilitates the entry of streptozotocin into the cytoplasm of pancreatic  $\beta$  cells. This causes increased depolarization in the mitochondria due to the influx of  $Ca^{2+}$  ions which is followed by the use of excess energy resulting in an energy shortage in the cell.

The data in table 2 shows the average blood glucose levels of male mice in each group which were measured on day 0 after being made diabetic, day 6, and day 21. Group I was the negative control group (normal), group II was the positive control, group III was treated with *Trigona laeviceps* honey at a dose of 0.27 ml/200mgBW and group III was given *Trigona laeviceps* honey treatment 0.54 ml/200mgBW.

From table 1 it can be seen that *Trigona laeviceps* honey contains a lot of phenols and flavonoids. Polyphenols contain antioxidants that can reduce oxidative stress by inhibiting the change in superoxide (O<sub>2</sub>) chains into hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>). Polyphenols can also inhibit and treat several degenerative diseases, one of which is diabetes. Polyphenols in honey can reduce hydroxyl and peroxy alcohol radicals because they act as hydrogen donors.<sup>7,8</sup>

The decrease in blood glucose levels in mice given *Trigona laeviceps* honey could also be caused by the influence of the flavonoid compounds contained in honey. In the quantitative phytochemical test of *Trigona laeviceps* honey in this study, data on the flavonoid content was quite high, namely 94.414 mg QE/100 g (table 1). In other research on the effect of methanol extract of leafy flower root (*Phyllanthus niruri* L.) on hyperglycemic rats, it also showed the occurrence of a hypoglycemic mechanism.<sup>9</sup> This is possibly because flavonoid glycoside compounds can increase the solubility of blood glucose so that it is easily excreted in the urine.<sup>10</sup>

The role of flavonoids and alkaloids as hypoglycemic agents was also reported in research by Arjadi and Susatyo (2007). Alkaloid and flavonoid compounds can repair (regenerate) damaged pancreatic  $\beta$ -cells and protect  $\beta$ -cells from damage and stimulate insulin release. Meanwhile, alkaloids have the ability to regenerate damaged pancreatic  $\beta$ -cells. Alkaloids can also stimulate sympathetic nerves (sympathomimetics) so that they can increase insulin secretion.<sup>11</sup>

Flavonoids are antioxidants that protect pancreatic cells due to the effects of free radicals. Alkaloids can inhibit glucose absorption in the intestine, increase glucose transport in the blood, stimulate glycogen synthesis and inhibit glucose synthesis by inhibiting the enzymes glucose 6-phosphatase, fructose 1,6-bisphosphatase which can reduce the formation of glucose from substrates other than carbohydrates.<sup>12</sup>

## CONCLUSION

This research showed that there was a decrease in glucose levels in the group of mice given *Trigona laeviceps* honey at a dose of 0.27 ml/200mgBW and 0.54 ml/200mgBW. In addition, significant differences in mean glucose levels were obtained based on treatment groups (p 0.000).

## REFERENCES

1. American Diabetes Association. Diagnosis and Classification of Diabetes Mellitus. *Diabetes Care*. 2012; 35(Supplement\_1):S64–S71

2. Rahman I, Biswas SK, Kirkham PA. Regulation of inflammation and redox signaling by dietary polyphenols. *Biochem Pharmacol*. 2006;72(11):1439-1452.
3. Ozougwu JC, Obimba KC, Belonwu CD, Unakalomba CB. The Pathogenesis and Pathophysiology of Type 1 and Type 2 Diabetes Mellitus. *Academic Journals*. 2013;4(4): 46-57
4. Kementerian Kesehatan Republik Indonesia. Riset Kesehatan Dasar. Badan Penelitian dan Pengembangan Kesehatan. 2018.
5. Rao P, Kumarathevan, Salleh N, Gan SH. Biological and therapeutic effects of honey produced by honey bees and stingless bees : a comparative review. *Brazilian Journal of Pharmacognosy*. 2016;1(26):1-8.
6. De Sousa JM, de Souza EL, Marques G, Meireles B, de Magalhães Cordeiro ÂT, Gullón B, and Magnani M. Polyphenolic profile and antioxidant and antibacterial activities of monofloral honeys produced by Meliponini in the Brazilian semiarid region. *Food Research International*. 2016;84,61–68.
7. Eliasson L, Abdul KF, Braun MG, Alvanovskis J, Hoppa MB and Rorsman P. “Novel aspects of the molecular mechanism controlling insulin secretion”. *J. Physiol*. 2008; 586(14):13-24
8. Biluca FC, da Silva B, Caon T, Mohr ETB, Vieira GN, Gonzaga LV, et al. Investigation of phenolic compounds, antioxidant, and anti-inflammatory activities in stingless bee honey (Meliponinae). *Food Res. Int.*, 2020;129
9. Fahri C, Sutarno, Listyawati S. Kadar Glukosa dan Kolesterol Total Darah Tikus Putih (*Rattus norvegicus* L.) Hiperglikemik setelah Pemberian Ekstrak Metanol Akar Meniran (*Phyllanthus niruri* L.). *Biofarmasi*. 2005;3(1):1-6.
10. Chairul Y, Jamal Z, Zainul. Efek Hipoglikemik Ekstrak Herba Meniran (*Phyllanthus niruri* L.) pada Kelinci Putih Jantan. *Berita Biologi*. 2000;5(1):93-100.
11. Arjadi F, Susatyo P. Regenerasi Sel Pulau Langerhans Pada Tikus Putih (*Rattus norvegicus*) Diabetes yang Diberi Rebusan Daging Mahkota Dewa (*Phaleria macrocarp* (scheff.) Boerl.). 2007;2(2): 118-122
12. Larantukan SVM, Setiasih NLE, Widyastuti SK. Pemberian Ekstrak Etanol Kulit Batang Kelor Glukosa Darah Tikus Hiperglikemia. *Indonesia Medicus Veterinus*. 2014;3(4):292-299

