

The Effectiveness of the α -Amylase Enzyme Inhibition by Several Traditional Medicinal Ingredients Black Turmeric, Aromatic Ginger, Black Cumin, Forest Bee Honey and Sweet Leaves in Ready to Drink Formulation

Prayoga Eko Nuryadi¹, Siti Juariah², Serli Alnamira Lorenza³, Susi Endrini⁴

^{1,3,4}Faculty of Medicine, Abdurrah University Jl. Riau Ujung No. 73, Tampan, Air Hitam, Payung Sekaki, Pekanbaru, Riau 28291, Indonesia

²Faculty of Pharmacy and Health Science, Abdurrah University Jl. Riau Ujung No. 73, Tampan, Air Hitam, Payung Sekaki, Pekanbaru, Riau 28291, Indonesia

ABSTRACT

The inhibition of α -amylase enzymes can delay the remaining carbohydrates in the small intestine and reduce the travel of postprandial blood glucose, therefore the content of compounds in the form of tannins, flavonoids, saponins and terpen is believed to be able inhibit the α -amylase enzymes. Knowing the effectiveness of α -amylase enzyme inhibition by some traditional medicine ingredients (*Curcuma caesia*, *Kaempferia galanga*, *Nigella sativa*, *Apis dorsata*, and *Stevia rebaudiana*) and organoleptic properties of the ready to drink formulation. This study is an experimental study using the design "Pretest-Posttest with control Group" on the sample in the form of liquid extracts. Plant liquid extract is tested for its inhibition of α -amylase and used as a formulation of herbal drink ready to drink. The result IC₅₀ values of black turmeric extract (6.46 mg/ml), aromatic ginger (6.10 mg/ml), black cumin (7.36 mg/ml), forest bee honey (2.59 mg/ml) and sweet leaves (4,85 mg/ml). The results of IC₅₀ formulations 1 (8.03 mg/ml) and 2 (14.18 mg/ml) and glibenclamide values are (2.53 mg/ml). Conclusion the effectiveness of α -amylase inhibition of sweet leaf extract is better than other extracts and formulation 1 is better than formulation 2.

ARTICLE DETAILS

Published On:
19 June 2024

Available on:
<https://ijmscr.org/>

1. INTRODUCTION

Diabetes mellitus type 2 is a chronic metabolic disorder where there are disorders of insulin secretion in the pancreatic glands which are characterized by the presence of persistent hyperglycemia[1]. In people with diabetes mellitus type 2 α -amylase levels are higher than in normal people[2]. One of the biggest risk factors associated with the occurrence of diabetes mellitus is obesity. Obesity is a problem that is often found in children to adults, according to [3] Obesity is an excessive accumulation of fat due to an imbalance between energy intake (energy intake) with the energy used (energy expenditure) for a long time[4].

Herbal medicines have been widely used to replace the role of synthetic drugs in dealing with diseases, this is due to lower and more effective side effects, therefore drugs derived from plant extract are expected to be safer and more effective in controlling blood sugar levels[5]. Black turmeric

(*Curcuma caesia*) has the potential as an antioxidant through the inhibition of α -Amylase[6], Next, black turmeric also has potential as an antibacterial inhibiting bacterial growth[7].

Medicinal ingredients that can be used as a blood sugar control through their effect on inhibition of α -amylase enzymes, namely first, black turmeric (*Curcuma caesia*) which has been proven to have an inhibitory effect on the α -amylase enzyme is higher than other types of curcuma with a percentage value of 97, 72% in ethyl acetate solvents[8]. Second, Kencur (*Kaempferia galanga*) delivered in previous studies was conducted by [9] that aromatic ginger has an anti-diabetic effect and is able to repair damage to the albino male rats[5]. Third, black cumin (*Nigella sativa*) which has an IC₅₀ value of 13.91 μ M which is proven to have an effect in inhibiting the work of α -amylase [10]. Fourth, honey (*Apis dorsata*) which has a percentage of resistance of 39.19% and the value of IC₅₀ 872.6 μ g/ml which is still quite good in

The Effectiveness of the α -Amylase Enzyme Inhibition by Several Traditional Medicinal Ingredients Black Turmeric, Aromatic Ginger, Black Cumin, Forest Bee Honey and Sweet Leaves in Ready to Drink Formulation

reducing blood sugar levels and is able to have the inhibition of α -amylase properties [11]. Fifth, sweet leaf (*Stevia rebaudiana*) which has an IC_{50} value of 13.73 $\mu\text{g/ml}$ on water extract samples and tests carried out on methanol extracts, ethanol, acetone is not possible because almost no inhibition activity found[9].

The five medicinal ingredients above will be made ready to drink which is useful for maintaining blood sugar levels and can prevent obesity. Ready to drink is a term used in describing the type of drink contained in a special packaging so that it can be drunk immediately without having to process it first[12].

2. METHODOLOGY

2.1. Research tool

Equipment used in this study in the form of knives, grinder, glass (pyrex), beaker glass, analytical balance, microplate reader, well plate 96, test tubes, test tube racks, micropipetts, oven, filter paper, blender, spatula, erlenmeyer, pH Meters, cups, drink bottles and plastic wrap.

2.2. Research material.

The ingredients used in this study are black turmeric, (*Curcuma caesia*), kencur (*kaempferia galanga*), black cumin (*nigella sativa*), forest bee honey (*apis dorsata*) sweet leaves (*stevia rebaudiana*), α -amylase enzymes, aluminum foil, Aquades, 1% starch solution, sodium phosphate buffer solution, ka-na tartrate, glibenclamide and reagent of dinitrosalicylic acid (DNS).

2.3. Preparation of plant extract.

The sample in the form of rhizomes from black turmeric and kencur was washed with running water until it was clean from dirt, after that it was cut into small pieces, black cumin (seeds) and sweet leaves, each as much as 500 gr was washed with running water and dried at room temperature, then proceed to the drying process using an oven at a temperature of 50°C for 72 hours[13]. Material that has been dry is mashed using a blender and sifted until it becomes fine powder. Then proceed with the infusa process by taking 10 grams of simplicia and boiled at 100°C with a total of 100 ml distilled water distil houses and dilution[14].

2.4. Inhibition of α -amylase enzyme inhibition in vitro in extracts and formulations.

To conduct testing in inhibition of α -amylase using the modification method described in the study of [15] and [16] In the early stages of 1% starch solution (10 μL), sodium phosphate buffer (20 μL) and sample (10 μL) with various concentrations namely 10 mg/ml, 7.5 mg/ml, 5 mg/ml, 2.5 mg/ml, 1.25 mg/ml which is then put into the holes contained in the well plate 96, distilled water is used as a sample blank, after being completed into the well plate 96 each solution and sample with various concentrations replicated into 3, then incubated at 37°C for 3 minutes. After the preincubation is complete, followed by the addition of 10 μL of the α -amylase enzyme in each well plate 96 hole except in an empty hole. The mixture is incubated at 37°C for 15 minutes. The enzymatic reaction is stopped by adding a 50 μL of dinitrosalicylic acid reagent (DNS) to each well plate 96 hole containing an enzymatic reaction. Absorbance is measured using a microplate reader at a wavelength of λ 490 nm. Control is prepared to be testing using the same procedure only replacing extracts with distilled water for negative controls and 5 mg Glibenclamide tablets for positive control and 3 replication of each control[15][16]. α -amylase inhibition activity will be calculated as a percentage of inhibition:

$$\% \text{inhibition} = \frac{(A_0 - A_t) \times 100\%}{A_0}$$

A_0 = Absorbance of control groups (without plant extracts)

A_t = Absorbance of aqueous extract of medicinal plants.

2.5. Making a ready to drink beverage formulation.

Powder extract from black turmeric, kencur, black cumin that has been boiled for 5 minutes with a temperature of 100°C on water baths, filtered and left at room temperature and has been through the process of assessing resistance from α -amylase through spectrophotometry and then inserted into a container with a capacity of 100 ml, the appropriate composition of each ingredient can be seen in Table 1, in addition to these ingredients add honey and sweet leaves as natural flavorings. After being inserted all components of the material into the container then stir until homogeneous and filtered again until the formulation is completely clean from the remaining powder of the material and closed tightly. Formulations are made based on modifications from research conducted by [17] so that the formulation is as follows[17]:

Table 1: Formulation of herbal drink ready to drink¹⁴

Ingredient	F 1	F2
Black turmeric extract (ml)	20	20
Aromatic ginger extract (ml)	20	20
Black cumin extract (ml)	20	20
Forest bee honey extract (ml)	40	-
Sweet leaf extract (ml)	-	40
Total	100 ml	100 ml

The Effectiveness of the α -Amylase Enzyme Inhibition by Several Traditional Medicinal Ingredients Black Turmeric, Aromatic Ginger, Black Cumin, Forest Bee Honey and Sweet Leaves in Ready to Drink Formulation

Information :

F1 : Beverage formulation with black turmeric extract, aromatic ginger, black cumin and honey

F2 : Formulation of drinks with black turmeric extract, kencur, black cumin and sweet leaves

3. RESULTS

3.1. *The effectiveness of α -amylase inhibition of black turmeric extract, kencur, black cumin, sweet leaves and forest honey.*

The results obtained from this study descriptively show that Stevia leaves have the effectiveness of α -amylase inhibition better than black turmeric, kencur, black cumin and honey (fig. 1). If sorted from the smallest to large IC₅₀ value then after stevia leaves (2.59 mg/ml) there is forest honey (4.85 mg/ml), kencur (6.10 mg/ml), black turmeric (6.46 mg/ml) and black cumin (7.36 mg/ml).

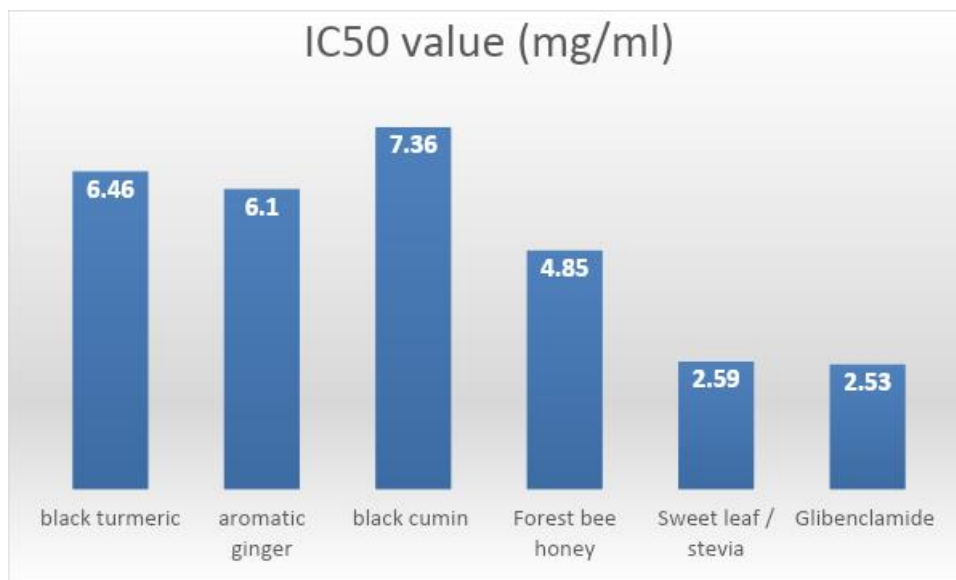


Fig. 1. Comparison of IC₅₀ sample values and positive control

Based on the results of the percentage of absorbance of each sample and positive control listed on (Table 2) the highest percentage of α -amylase inhibitory activity values

found at a concentration of 1.25 mg/ml (black turmeric, kencur, forest bee honey, stevia leaves and Glibenclamide) but in black cumin is found in a concentration of 7.5 mg/ml.

Table 2: Comparison of the percentage of absorbance of each sample and positive control with various concentrations

Sampel	Konsentration (mg/ml)				
	10	7,5	5	2,5	1,25
Black Turmeric	7,7483251	21,118555	19,895135	23,71104	28,633848
Aromatic Ginger	18,147393	22,371104	22,80804	23,71104	30,323332
Black Cumin	22,575007	32,158462	22,050685	17,652199	29,886397
Forest Bee Honey	24,61404	32,187591	25,22575	31,371978	33,090591
Sweet Leaves	49,348697	49,766199	49,615898	52,955912	55,527722
Glibenclamide	-30,38159	58,112438	59,976697	61,316633	66,093796

The positive control used is Glibenclamide, testing of anti-diabetic anti-diabetic has been proven and is often used as a blood sugar control drug. The IC₅₀ value of the α -amylase inhibition test in this study is 2.53 mg/ml which is classified as strong in inhibiting the α -amylase enzyme. IC₅₀ results from black turmeric in this study when compared to research conducted by Jain and Parihar (2018) obtained the IC₅₀ value of black turmeric of 92.36 μ g/ml with ethanol solvents and 80.35 μ g/ml with methanol solvents, so the IC₅₀

value In this study that uses water as a solvent shows the results of the black turmeric IC₅₀ value of (6.46 mg/ml)¹⁵. This considerable difference is because the solvent used is different and the form of extraction that is also different. Likewise with aromatic ginger that is not much different from the results of black turmeric, obtained the results of the IC₅₀ value (6.10 mg/ml) and the results in previous studies conducted in one genus *Kaempferia* but different species namely *Kaempferia parviflora* with Using ethanol as a

The Effectiveness of the α -Amylase Enzyme Inhibition by Several Traditional Medicinal Ingredients Black Turmeric, Aromatic Ginger, Black Cumin, Forest Bee Honey and Sweet Leaves in Ready to Drink Formulation

solvent and its inhibition is carried out on α -glucosidase indicating the IC_{50} value ($20.4 \mu\text{g/ml}$)¹⁶. Some things that can affect the differences that are quite far from previous studies related to the type of sample species that are tested differently, different forms of extraction and different types of solvents used.

In this study the black cumin IC_{50} value showed results (7.36 mg/ml) and looked good in inhibiting the α -amylase enzyme compared to previous studies which had an IC_{50} value of $13.91 \mu\text{m}$ by using hexane and acetone in the process of maceration extraction⁶. The results obtained from the test conducted on black cumin there are differences with previous research this is related to the form of extraction used differently and the solvents used are different. The results of the test conducted on forest honey were obtained ($4,85 \text{ mg/ml}$), the results in this study were very different from previous studies conducted by [11] has a resistance percentage value of 39.19% and IC_{50} value ($872.6 \mu\text{g/ml}$)⁷. Stevia leaf results which are the best results compared to other samples of (2.53 mg/ml) when compared to previous studies conducted by [9] the results of IC_{50} ($13.73 \mu\text{g/ml}$) were obtained with a solvent which is used aquades or water, in this study in line with previous studies where using water as a solvent has a strong potential as an α -amylase inhibit, this is related to the presence of steviol glycoside content from *Stevia rebaudiana*[9].

This comparison is carried out because there is no standard unit to assess how strong an extract can inhibit α -amylase when viewed from the IC_{50} value and the absence of similar research conducted on several traditional ingredients tested in this study. The differences that occur from the inhibition of α -amylase can be influenced by the extract components used differently, the compounds contained in the extract samples are different, as well as the method of drying and solvents used when extraction of bioactive compounds has a significant effect on the ability of extract samples in inhibiting enzymes α -amylase[19].

3.2. The effectiveness of α -amylase inhibition from the formulation of black turmeric, kencur, black cumin, sweet leaves and forest honey.

The formulation used in this study consists of 2 groups, namely for formulation 1 made of black turmeric, kencur, black cumin and forest bee honey and for formulation 2 made of black turmeric, kencur, black cumin and stevia leaves which then from this formulation will be made into A ready to drink herbal drink so that consumers who consume no longer need to process these ingredients to get their properties. Currently herbal plants extract is increasingly used for making food and drinks compared to products made from chemicals, with the development of technology and science there have been many studies on herbal beverage products for health[19].

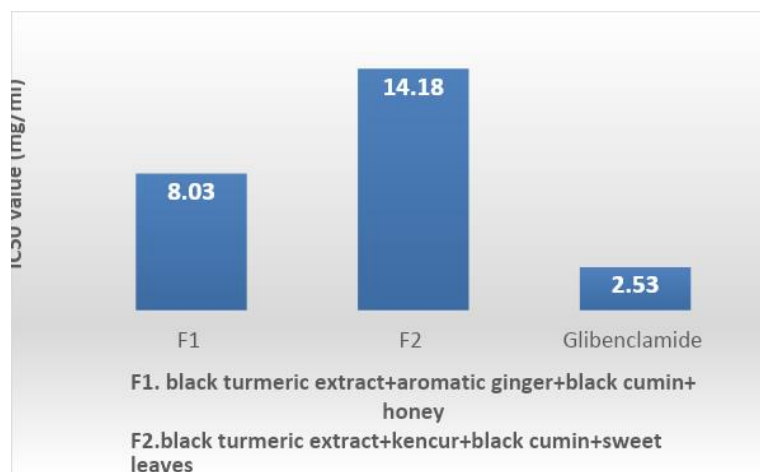


Fig. 2 Comparison of IC_{50} Formulation Values 1 and 2

Based on the results of the IC_{50} obtained from the two ready to drink herbal formulations above, IC_{50} formulation 1 (8.03 mg/ml) is smaller than the formulation 2 (14.18 mg/ml), so that formulation 1 effectiveness in inhibiting α -amilase enzymes better than formulation 2.

The results of the percentage of absorbance listed on (Fig.3) can be seen that formulation 1 has the highest inhibition activity of the percentage at a concentration of 1.25 mg/ml but formulation 2 is concentrated 5 mg/ml .

The Effectiveness of the α -Amylase Enzyme Inhibition by Several Traditional Medicinal Ingredients Black Turmeric, Aromatic Ginger, Black Cumin, Forest Bee Honey and Sweet Leaves in Ready to Drink Formulation

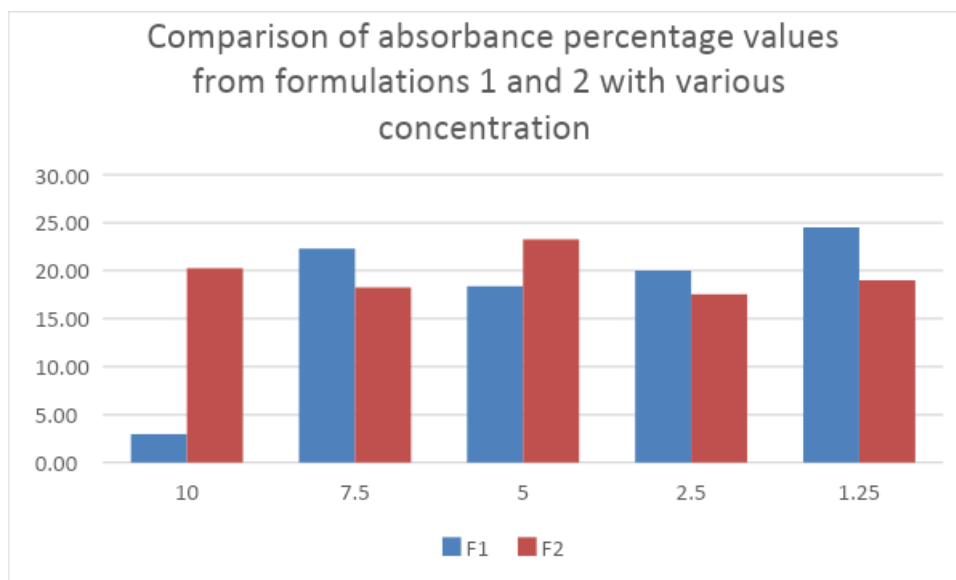


Fig. 3 Comparison of absorbance percentage values from formulations 1 and 2 with various concentrations

The tests carried out on the inhibition of α -amylase enzymes aims to determine the ability of the sample in binding and activating the work of the α -amylase enzyme so that no bonds are formed with starch and with the inhibition of the α -amylase enzymes are expected absorbance of sugar and can suppress increased postprandial glucose[19].

The sample material used in this study has the ability to inhibit the activity of α -amylase enzymes. This is thought to be related to the secondary metabolic content in black turmeric, kencur, black cumin, forest bee honey and stevia leaves that are in line with research from M. Daud AK *et al* (2019) which states that active compounds contained in plant extracts have inhibitory properties good on the activity of the α -amylase enzymes and α -glucosidases such as tannins, flavonoids, polysaccharides, tercent and saponins so that plants that contain active substances are often used as herbal medicines in overcoming high blood sugar levels or preventing diabetes mellitus[21]. This study is in line with research conducted by [22] also said that the α -amylase inhibition mechanism of active biochemical compounds contained in the drug ingredients such as flavonoids, tannins, saponins and terpenes cannot be explained scientifically, But in several studies that have been carried out explains that these compounds are related to the bonds formed between α -amylase and the active biochemistry[22].

Sample testing in this study has the principle that is more active from the extract compound used, the hydrolyzed starch will be less so that the less glucose used. Extracts that have the biggest % inhibition are the most active extracts[23]. According to [24] the level of hydrolysis that occurs in starch by α -amylase is related to several conditions that can affect such as the concentration of substrate, pH, temperature, activator and inhibitor. In this study, the largest inhibition % inhibition value of each sample was obtained at a concentration of 1.25 mg/ml for black turmeric, kencur,

honey and stevia leaves while for black cumin at a concentration of 7.5 mg/ml[24].

Based on the results of the IC_{50} value in (Table 2) shows that Glibenklamide as a positive control has the smallest IC_{50} value compared to the results of the extract sample, but the difference in the glibenklamide IC_{50} value and the extract samples are not much different, especially the Stevia leaves which only have a difference of 0.06 mg /ml compared to glubenklamide. Glibenclamide is one of the type 2 DM drugs that is often used and entered into the sulfonylurea group that works by stimulating insulin expenditure by binding to receptors on the β cells of Langerhans Island²⁴. This small IC_{50} value can be influenced by the correct inhibition process of the variation of the sample concentration carried out in the test, on the contrary the large IC_{50} value is influenced by the effect resulting from the content of the sample compound is smaller in inhibiting the activity of the α -amylase enzyme.

4. CONCLUSIONS

Based on the results of the research conducted the following conclusions were obtained:

1. The smallest to largest IC_{50} value, after stevia leaves have forest honey (4.85 mg/ml), kencur (6.10 mg/ml), black turmeric (6.46 mg/ml) and black cumin (7.36 mg/ml). So that Stevia leaves have an effectiveness in α -amylase inhibition which is better than other extract samples.
2. The highest percentage of α -amylase inhibition activity is found at a concentration of 1.25 mg/ml (black turmeric, kencur, forest bee honey, stevia leaves and glibenclamide) but in black cumin is found in a concentration of 7.5 mg/ml.
3. IC_{50} results obtained from the two ready to drink herbal drink formulations above are found IC_{50} Formulation 1 is smaller (8.03 mg/ml) compared to

The Effectiveness of the α -Amylase Enzyme Inhibition by Several Traditional Medicinal Ingredients Black Turmeric, Aromatic Ginger, Black Cumin, Forest Bee Honey and Sweet Leaves in Ready to Drink Formulation

formulation 2 (14.18 mg/ml), so that formulation 1 effectiveness in inhibiting the α -enzyme amylase is better than formulation 2.

4. Formulation 1 has the highest inhibitory activity of its percentage at a concentration of 1.25 mg/ml but formulation 2 is concentrated 5 mg/ml.

ACKNOWLEDGEMENT

This research was not funded by any grant.

REFERENCES

- I. G. P. Suryaputra, H. Apriningsih, and M. M. Wardani, "Hubungan Komorbid dengan Mortalitas dan Lama Rawat Inap pada Pasien COVID-19 di Rumah Sakit UNS Surakarta," *Plex. Med. J.*, vol. 1, no. 1, pp. 32–41, 2022.
- II. N. Malathi, S. Mythili, and H. R. Vasanthi, "Salivary diagnostics: a brief review," *Int. Sch. Res. Not.*, vol. 2014, 2014.
- III. Kementerian Kesehatan RI, "Panduan Pelaksanaan Gerakan Nusantara Tekan Angka Obesitas (GENTAS)," [Http://P2Ptm.Kemkes.Go.Id/Dokumen-Ptm/Panduan-Gentas](http://P2Ptm.Kemkes.Go.Id/Dokumen-Ptm/Panduan-Gentas). pp. 6–16, 2017.
- IV. R. I. Kemenkes, "Panduan Pelaksanaan Gerakan Nusantara Tekan Angka Obesitas (GENTAS)," *Direktorat Pencegah. dan Pengendali. Penyakit Tidak Menular Direktorat Jenderal Pencegah. dan Pengendali. Penyakit Kementeri. Kesehat. RI*, 2017.
- V. P. Jain, T. Satapathy, and R. K. Pandey, "Acaricidal activity and biochemical analysis of Citrus limetta seed oil for controlling Ixodid Tick Rhipicephalus microplus infesting cattle," *Syst. Appl. Acarol.*, vol. 26, no. 7, pp. 1350–1360, 2021.
- VI. S. Kartini, S. Juariah, D. Mardhiyani, M. F. A. Bakar, F. I. A. Bakar, and S. Endrini, "Phytochemical Properties, Antioxidant Activity and α -Amilase Inhibitory of *Curcuma caesia*," *J. Adv. Res. Appl. Sci. Eng. Technol.*, vol. 30, no. 1, pp. 255–263, 2023.
- VII. S. Juariah, F. I. A. Bakar, M. F. A. Bakar, and S. Endrini, "Antibacterial potential of *Curcuma caesia* Roxb ethanol extract against nosocomial infections," *Bali Med. J.*, vol. 12, no. 2, pp. 1959–1963, 2023.
- VIII. N. W. Sudatri, N. Warasiti, N. M. Suartini, and N. I. Bidura, "Anti-diabetic and anti-cholesterol activity of *Kaempferia galanga* L. herbal medicine rhizome in albino rats," *Int. J. Fauna Biol. Stud.*, vol. 6, no. 5, pp. 13–17, 2019.
- IX. U. H. Zaidan, N. I. Mohamad Zen, N. A. Amran, S. Shamsi, and S. S. A. Gani, "Biochemical evaluation of phenolic compounds and steviol glycoside from *Stevia rebaudiana* extracts associated with in vitro antidiabetic potential," *Biocatal. Agric. Biotechnol.*, vol. 18, no. February, p. 101049, 2019, doi: 10.1016/j.bcab.2019.101049.
- X. S. Tiji *et al.*, "Linking the phytochemicals and the α -glucosidase and α -amylase enzyme inhibitory effects of *Nigella sativa* seed extracts," *Foods*, vol. 10, no. 8, p. 1818, 2021.
- XI. V. Krishnasree and P. Mary Ukkuru, "In vitro antidiabetic activity and glycemic index of bee honeys," *Indian J. Tradit. Knowl.*, vol. 16, no. 1, pp. 134–140, 2017.
- XII. T. D. A. Mukmina, R. L. Prameswari, R. I. Hapsari, I. Muflihati, A. R. Affandi, and J. S. T. No, "Karakteristik minuman ready to drink dengan variasi konsentrasi cmc dan rasio kacang tunggak dan kacang hijau," *J. Pangan dan Gizi*, vol. 9, no. 1, pp. 74–82, 2019.
- XIII. A. Chandra and J. R. B. Witono, "Pengaruh berbagai proses dehidrasi pada pengeringan daun *Stevia Rebaudiana*," in *Seminar Nasional Teknik Kimia "Kejuangan"*, 2018, p. 11.
- XIV. S. Yuningtyas and D. S. Artianti, "Aktivitas Inhibisi Enzim? -Glukosidase Ekstrak Air Dan Etanol Umbi Lapis Bawang Merah (*Allium Ascalonicum*)," *Fitofarmaka J. Ilm. Farm.*, vol. 5, no. 1, pp. 1–7, 2015.
- XV. M. I. Kazeem, J. O. Adamson, and I. A. Ogunwande, "Modes of inhibition of α -amylase and α -glucosidase by aqueous extract of morinda lucida benth leaf," *Biomed Res. Int.*, vol. 2013, 2013, doi: 10.1155/2013/527570.
- XVI. D. Pujimulyani *et al.*, "Amylase inhibition and free radical scavenging activities of white turmeric extract and fractions," *J. Teknol. dan Ind. Pangan*, 2018.
- XVII. M. Mardiah, R. Noor, M. Suprayatmi, and E. Listianti, "MINUMAN FUNGSIONAL READY TO DRINK BERBAHAN BAKU REMPAH LOKAL," *J. Ilm. PANGAN HALAL*, vol. 1, no. 2, pp. 48–54, 2019.
- XVIII. A. Jain and D. K. Parihar, "In-vitro antidiabetic potential of endangered *Curcuma caesia* rhizome extracts and its characterization by FTIR and GC / HR-MS analysis," *Int. J. Adv. Sci. Eng. Technol.*, vol. 6, no. 4, pp. 16–22, 2018.
- XIX. *et al.*, "Effect of drying methods and extraction solvents on anti-amylase activity of selected medicinal plants," *J. Ayurvedic Herb. Med.*, vol. 7, no. 1, pp. 24–29, 2021, doi: 10.31254/jahm.2021.7106.
- XX. M. A. M. Daud AK, Juliani, Sugito, "1. α -Amylase and α -Glucosidase Inhibitors from Plant Extracts," *J. Med. Vet.*, vol. 13, no. 2, pp. 151–158, 2019, doi: 10.21157/j.med.vet.v13i2.13819.

The Effectiveness of the α -Amylase Enzyme Inhibition by Several Traditional Medicinal Ingredients Black Turmeric, Aromatic Ginger, Black Cumin, Forest Bee Honey and Sweet Leaves in Ready to Drink Formulation

- XXI. M. D. AK, J. Juliani, S. Sugito, and M. Abrar, "21. α -Amylase and α -Glucosidase Inhibitors from Plant Extracts," *J. Med. Vet.*, vol. 13, no. 2, 2019.
- XXII. A. I. Martinez-Gonzalez, G. Díaz-Sánchez, L. A. de la Rosa, I. Bustos-Jaimes, and E. Alvarez-Parrilla, "Inhibition of α -amylase by flavonoids: Structure activity relationship (SAR)," *Spectrochim. Acta - Part A Mol. Biomol. Spectrosc.*, vol. 206, pp. 437–447, 2019, doi: 10.1016/j.saa.2018.08.057.
- XXIII. W. Wirasti and T. Lestari, "Penghambatan Ekstrak Daun Kremah (*Alternanthera sessilis*) Terhadap Enzim α -amilase secara In-Vitro," *Pharmakon J. Farm. Indones.*, vol. 18, no. 1, pp. 68–74, 2021.
- XXIV. Y. S. SOEKA, "The ability *Bacillus licheniformis* to produce $\hat{\pm}$ -amylase enzyme," in *Prosiding Seminar Nasional Masyarakat Biodiversitas Indonesia*, 2015, vol. 1, no. 5, pp. 1162–1166.