

**ANTI-GLICATION ACTIVITY TEST OF EFFERVESCENT GRANULES
COMBINATION OF GREEN OKRA (ABELMOSCHUS ESCULENTUS L) AND
BETTER GOURD (MOMORDICA CHARANTIAL L) FRUIT EXTRACT USING UV-
VIS SPECTROPHOTOMETRY METHOD****Suharyanto¹, Tri Harningsih^{1*}, Sri Harini²**¹Sekolah Tinggi Ilmu Kesehatan Nasional, Jl. Raya Solo - Baki, Bangorwo, Kwarasan, Grogol, Sukoharjo, Central Java 57552, Indonesia²Universitas Islam Negeri Maulana Malik Ibrahim Malang, Jl. Gajayana No. 50, Dinoyo, District. Lowokwaru, Malang City, East Java 65144, Indonesia*tri.harningsih@stikesnas.ac.id**ABSTRACT**

Glycation is a non-enzymatic chemical reaction between reducing sugars and amino groups from proteins, nucleic acids or phospholipids to form advanced glycation end products (AGEs). The formation of AGEs is related to blood glucose control. Inhibition of AGEs formation in diabetes can limit tissue damage. The bad impact of increasing blood glucose levels is hyperglycemia. Bitter melon fruit empirically lowers blood glucose in test animals by reducing blood glucose levels. through the process of inhibiting gluconeogenesis in the liver, protecting pancreatic beta cells, increasing insulin sensitivity and reducing oxidative stress. Meanwhile, okra fruit contains metabolite compounds for the treatment of diabetes mellitus, through inhibiting the activity of the α -glucosidase enzyme, improving pancreatic β cells and antioxidant properties. Effervescent is something that contains gas bubbles, like water-soluble tablets with a foaming effect. The aim of this research is to determine the Anti-Glycation Combination of Effervescent Granules from Green Okra and Bitter Gourd fruit extracts. The research was carried out by extracting Bitter Gourd and Green Okra. The extraction results are mixed in the same ratio to make three Effervescent Granule formulations. The results of the Effervescent granule formulation were tested for anti-glycation activity using the spectrophotometric method. From the research results, it can be concluded that the combination formula of 200mg bitter melon extract and 200mg green okra extract has the best results with a % reduction in glycation of $52.650\% \pm 0.208$; IC50 108.5255 ppm with %KV 0.522

Keywords: ages; anti-glycation; bitter gourd; green okra

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INTRODUCTION

Diabetes is a metabolic disorder characterized by multilevel chronic hyperglycemia with derangement of carbohydrates, fats and proteins caused by impaired insulin secretion. One of the causes of diabetes is due to a lack of the hormone insulin, which functions to enter glucose into cells to be used as an energy source. Diabetes can result in increased insulin resistance. This insulin resistance forms a group that causes an increase in glucose levels in the blood so that insulin production will decrease (Kholifah, S., & Sari, R., 2021). A side effect of increasing plasma glucose levels is the occurrence of hyperglycemia known as glycation (Singh, V., 2014).

In 2019, DM cases in Indonesia amounted to 10.7 million people and are predicted to increase to 16.6 million in 2045 (IDF-International Diabetes Federation Atlas, 2009). Research shows that green okra fruit is effective in controlling diabetes mellitus (Raharjo, A.A., 2016). Okra fruit contains flavonoids which have antidiabetic effects (Astatl., & Kasmawati., 2017). Bitter melon fruit contains flavonoids, saponins and polyphenols (Yuda, I. K. A. and Dharmayudha, A. A. G. O. , 2013). Green okra and bitter melon fruit contain α -cellulose and hemicellulose which are included in the fiber group which has antidiabetic effects (Uraku, A.J, 2011). Glycation is a reaction between carbohydrates and proteins trigger hyperglycemia. This situation needs to be researched to prevent the occurrence of disease complications in patients. This research focuses on the analysis of effervescent granules from a combination of green okra and bitter melon fruit extracts, as a development of previous research.

METHOD

This type of research is experimental with a cross sectional design. The population of this research is Bitter Gourd Fruit and Hiju Okra which are sold at Pasar Gede Surakarta. The data collection technique was carried out randomly with sellers of Green Okra and Bitter Gourd. Sample preparation was carried out by chopping and heating under the hot sun. Simplicia each from green okra and dried bitter melon was weighed in the amount of 200 gr. Then extracted with maceration using 70% ethanol as a solvent with a ratio of 1:10 in 5 days. Maceration the first uses 1.5 liters of 70% ethanol for 3 days, then The macerate is filtered and separated between the dregs and the filtrate. Second maceration carried out by adding 70% ethanol using the remaining dregs from the maceration first 500 milliliters over 2 days. Then separate the maceration results with the dregs. The combination of the first and second Maserats has been processed together so that form a mixture. Then, the mixture was evaporated using a rotary evaporator and water bath (50oC) to produce a thick extract. The results are then calculated and the yield is calculated. The dry extract was then made into effervescent granule preparations with three formulations of a combination of Green Okra and Bitter Gourd.

RESULTS

Quantitative analysis the quantitative test results for the reduction in % Gication of FI (Formulation I), FII (Formulation II) and FIII (Formulation III) are presented in the following table:

Table 1.
Glycation % Reduction Test Results for Formulas I, II, III

Concentration (ppm)	% Glycation Reduction					
	F I / IC ₅₀	%KV	F II / IC ₅₀	%KV	F III / IC ₅₀	% KV
25	34,220±0,416	1,216	38,060±0,453	1,190	32,250±0,208	0,645
50	37,820±0,208	0,005	41,470±0,360	0,868	35,900±0,360	1,003
75	41,650±0,312	0,749	45,240±0,360	0,796	39,790±0,453	1,138
100	45,180±0,330	0,730	40,180±0,375	0,933	43,440±0,360	0,829
125	48,650±0,312	0,641	52,650±0,275	0,522	46,730±0,208	0,445
	133,623 ppm		108,523 ppm		146,082 ppm	

The graph of each formula is presented in the following image:

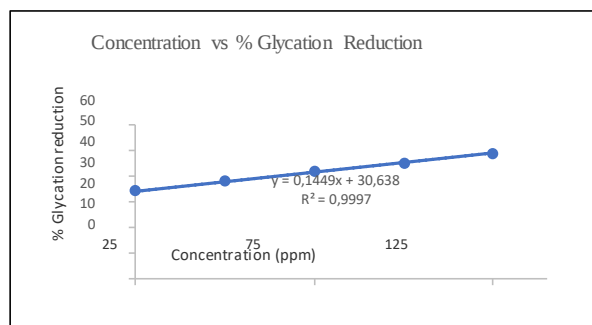


Figure 1: % Glycation Reduction Graph for Formula I Samples

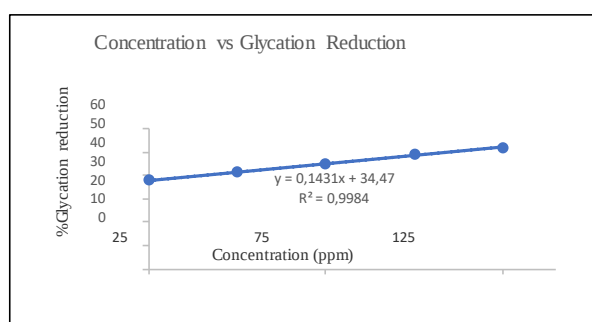


Figure 2: % Glycation Reduction Graph for Formula II Samples

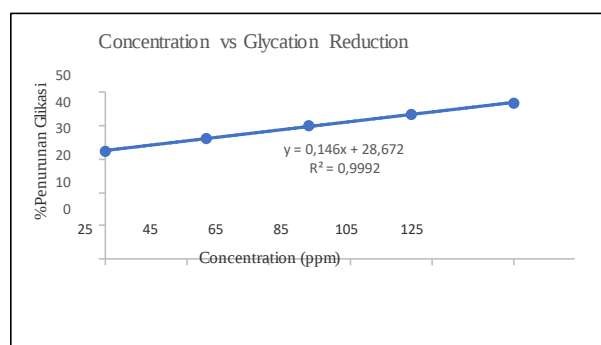


Figure 3: % Glycation Reduction Graph for Formula III Samples

DISCUSSION

Qualitative tests on samples with Formula I, Formula II and Formula III showed that they were positive for containing phenolics and flavonoids. Quantitative tests were carried out in this research. Table 1 shows that at a concentration of 125 ppm, Formula II has the highest % reduction in glycation which results in a lower IC₅₀ compared to Formula I and Formula III. This means that mixing 200 mg of bitter melon and 200 mg of green okra is optimal. The mixing process is greater, the % decrease in glycation is smaller, followed by a greater IC₅₀. Previous research showed that the % reduction in green okra in its single form was 1500 ppm with an average % reduction in glycation of 45,99 (Berliana, 2023). Meanwhile, bitter melon in previous research in juice form had an IC₅₀ of 69,24% or 69,24gr/100 ml in its single form (Widyanto, 2015). Previous research can state that the combination of bitter melon and green okra in effervescent form is better. Coefficient of variation is the comparison value between deviations standard with the calculated average value of comparison between standard deviation and the average value of a data distribution. The large coefficient of variation value

means the data less evenly distributed (heterogeneous), if it gets smaller coefficient of variation means the data is evenly distributed (homogeneous). The criteria for a good %KV value is 2% or less. Based on research data, the %KV value of each formula has a %KV of less than 2%. This shows that the level of accuracy in research is very good. The effervescent granule form has a better IC₅₀ than the extract form because the effervescent granule form has a faster dissolution speed resulting in higher inhibition (Harahap, 2017).

The granular effluent form has a better flow rate than the thick extract form. This will result in a better % reduction in glycation and IC₅₀ compared to the extract form (Lailiyah, 2023). The percentage reduction in glycation, the higher the IC₅₀ value, becomes smaller, so that the mixture of bitter melon and green okra effervescent granules has an inhibitory effect on the formation of AGEs in line with the presence of flavonoids and phenolics which have a structure containing hydroxyl groups, thus inhibiting the formation of glycation (Sovia, E. et al., 2011). The linear regression equation for Formula I, Formula II and Formula III using Microsoft Excel has a positive r value, meaning that in the linear equation this means that an increase in concentration will be followed by an increase in inhibitory potential so that it is % (Widyanto, 2015). Based on the IC₅₀ value, it shows that Formula I, Formula II and Formula III have the potential to reduce the %Glycation, although not too high. Bitter Gourd and Green Okra have an inhibitory effect on the formation of AGEs in line with the presence of flavonoids and phenolics which have a structure containing hydroxyl groups, thus inhibiting the formation of glycation (Sovia, E. et al., 2011).

CONCLUSION

Effervescent Granules Combination of Green Okra (*Abelmoschus esculentus* L) and Bitter Gourd Fruit Extracts (*Momordica charantia* L) have anti-glycation activity with % reduction in anti-glycation of $52,650 \pm 0.208$ with IC₅₀ 108,523 at a concentration of 125 ppm

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