https://doi.org/10.48048/tis.2024.7428

Anti-Diabetic Effect of Okara Noodles on Streptozotocin-Nicotinamide Induced Diabetic Rats

Kaisun Nesa Lesa[1,2,3], Nazir Ahmad[4], Yunika Mayangsari[1,*], Muhammad Nur Cahyanto[1] and Wahyu Dwi Saputra[1]

[1]Department of Food and Agricultural Product Technology, Faculty of Agricultural Technology, Universitas Gadjah Mada, Yogyakarta 55281, Indonesia
[2]Department of Pediatrics, Nihon University Hospital, Tokyo 101-8309, Japan
[3]Department of Nutrition and Food Technology, Jessore University of Science and Technology, Jashore 7408, Bangladesh
[4]Department of Pharmacology and Clinical Pharmacy, Faculty of Pharmacy, Universitas Gadjah Mada, Yogyakarta 55281, Indonesia

([*]Corresponding author's e-mail: yunika.mayangsari@ugm.ac.id)

Received: 26 August 2023, Revised: 9 October 2023, Accepted: 16 October 2023, Published: 25 February 2024

Abstract

Okara is a by-product of soybean curd residue resulting from the processing of soy milk and tofu. Okara noodles were made from wheat flour (WF) and okara flour (OF) with a few modifications that consist of high dietary fiber. Therefore, okara can be considered as an effective functional component with health-promoting benefits, especially for diabetes mellitus. Albeit, the anti-diabetic effect has not been conscientiously investigated. The goal of the current research was to evaluate the anti-diabetic effect of okara noodles on Streptozotocin-Nicotinamide induced diabetes mellitus rats. In the experiment, the different ratios of OF was used to prepared 3 formulated noodles (F_0, F_1 and F_2) where F_0 (0 % OF; 100 % WF), F_1 (25 % OF; 75 % WF) and F_2 (45 % OF; 55 % WF). Twenty-four male wistar rats were randomly divided into 4 groups (6 rats/groups), normal group of rats fed with standard feed and 3 diabetics groups, respectively fed with standard feed and formulated noodles variety (F_0, F_1 and F_2). In *in vivo* analysis, it indicated, diabetes mellitus rats fed okara noodles had lower blood glucose levels and HOMA-IR and higher serum insulin levels and HOMA-β index than those fed control noodles (F_0) as well as body weight managing activity. These outcomes demonstrated okara noodles possess significant anti-diabetic activity, prompting the applicability of okara noodles as potential food for the diabetes mellitus food products.

Keywords: Okara noodle, Anti-diabetic effect, STZ-NA, Diabetes mellitus, Rats

Introduction

Diabetes mellitus (DM), commonly known as diabetes [1], is a serious metabolic disease in which the human body does not produce enough insulin (a hormone that regulates blood sugar or glucose) or respond normally to it [2,3]. There are major 2 types of diabetes: Type-1 DM and Type-2 DM [4]. Type-2 DM is more common and well-known diabetes. This type of diabetes accounts for 90 - 95 % of all diabetic patients [5,6]. The prevalence of DM is gradually increasing [1,7] and affects many people worldwide [8]. Individuals with uncontrolled diabetes and chronic hyperglycemia may develop complications including neuropathy, macro-vascular disease, nephropathy and retinopathy, in addition, patient is being more vulnerable to infections [9-11]. International Diabetes Federation demonstrated the total number of people (adults, 20 - 79 years) who are diagnosed with DM has significantly increased at an alarming rate from 108 million in 1980 to 537 million in 2021. They also predicted that by the year 2045 this rate will be gradually increased to 643 million by 2030 and 783 million by 2045 [12,13]. The adoption of an obesogenic lifestyle, growing westernization, and unhealthy dietary pattern has contributed to increasing the prevalence of DM [14]. For the people with DM, continuous pharmaceutical drugs over time causes worrying issue of unwanted side effect, expensive cost and starchent boredome. Thus, for the management of DM, healthy diet management including dietary fibers (DFs) enriched, high protein, and low-calorie diet is an important keystone to maintaining their DM condition [15]. Therefore, several scientific studies on the designing and consumption of natural by-products for anti-diabetic activity without harmful effects have been reported.

Okara is a by-product of soybean curd residue resulting from the processing of soy milk and tofu [16-18]. In dry matter basis, okara contains high DFs (50 %), high protein (25 %) [19-21], hemicellulose,

phenolic content, lipid (10 %) [22], antioxidants, Isoflavones (12 to 40 %) [22,23] and vitamins [24-26] that are needed by the human body, particularly for the DM patients. Moreover, there is much evidence for the health benefits of DFs including hypo-glycemic activities, reducing body weight, hypo-lipidemic and anti-oxidant activities [27-30]. For this reason, okara application in the food industry is increasing day by day [22]. In the past decades, okara has been used in the food market to make bread [31,32], pan-cake, roti, paratha [33], puffing food [22], biscuits, cakes [34,35], candy, vegetable slices [35], cake [22], healthy beverage, sausage [35] and nutritional flour [22]. However, in the food industry and/or market, there is a lack of suitable food products (noodles) that are enriched in functional food, related to Type-2 DM patients. The consumption of special diabetic food products supplemented with functional food ingredients, such as okara can play an essential role in achieving health benefits for DM patients.

Noodles are the conventional main food items and consume in high rate in many Asian countries including Indonesia, Japan, China and Korea [15,36]. In the food industry, noodles are more popular because they are ready to eat, low in cost, convenient, varieties of tastes, availability, longer shelf life [37], as well as their ideal target for nutritional improvement [38]. The presence of DFs content ingredients (OF) in noodles improves their acceptability, nutritional quality, and improvements are achieved by changing the ratio of raw ingredients whole grains other than wheat, or by replacing or changing the ratio of DFs content food sources in basic recipes [39].

There has no *in vivo* study for the anti-diabetic effects of the okara noodles to evaluate by blood glucose levels (BGLs) and serum insulin levels (SILs) using in *in vivo* study. In this regard, considering the proposed newly developed okara noodles made from OF that achieved the protective effect against DM, which acted as a functional food. These outcomes may provide potential functions of okara for the treatment and prevention of anti-diabetic effects by STZ-NA-induced diabetic wistar white rats (*Rattus Norvegicus*).

Materials and methods

Reagents and raw materials

Rats (aged between 3 - 3.5 months old, weight 200 - 250 g) were collected from Nutrition Laboratory of PSPG-UGM (House of Experimental Rats CFNS-UGM), Indonesia. Other objects for the analysis of BGLs were white rat blood serum, Glucose standard FS (Diasys Germany), ELISA kit for insulin (Elabscience Biotechnology Co., Ltd), Glucose GOD-PAP biochemistry kit reagent (Diasys Germany), and sterile distilled water. The ingredients used in the preparation of experimental animal hyperglycemia were ketamine (Ket-A-100), STZ (Cayman) and NA (Sigma Aldrich) and Standardized Rat Feed (AIN93 M) consist of casein (The Tatua Cooperative Diary Company Ltd), mineral mix (AIN-93G-MX, MP Biomedicals, LLC), vitamin mix (AIN-93-VX, MP Biomedicals, LLC), L-cysteine (Merck KGaA, Germany), Choline bitartrate (Sigma Aldrich), sucrose (Merck KGaA, Germany), tert-butylhydroquinone (TBHQ), and other ingredients i.e. corn starch (maizenaku) and soybean oil (Happy soya oil) were collected from local supermarkets at Yogyakarta, Indonesia. All chemicals and reagents used in *in vivo* experiment were of analytical grade.

Animal experimental protocol

To determine the anti-diabetic activity an in *in vivo* test was carried out referring to Subamia *et al.* [2] with little modifications. All rats were put into individual caged (stainless steel, 15×15×30 in cm^3) in a closed with controlled light conditions 12:12 h day and night, adequate ventilation in the cage, air temperature at room temperature (22 - 25 °C), 60 - 65 % RH (humidity), and cleaning of the cage was carried out every day. The cages were individual cages and rats were separated from the feces and urine therefore there was no need for bedding. Drinking water and diet were available *ad libitum* during the experiments. In the experiment, a total of 24 animals were randomly divided into 4 groups, with each group consisting of 6 (4×6 = 24). Feeding treatment was for 21 days (3 weeks) according to the treatment group, which consisted of the control/normal G-I (N) group, rats fed with AIN-93M standard feed and drinking water *ad libitum* [40]. G-II, III and IV were DM induced rat, respectively fed with standard feed, drinking water *ad libitum* [2] and noodles variety of F_0, F_1 and F_2, where G-II was given control formulated noodles, F_0 (1.26/200 g BW/day/head), G-III was given formulated okara noodles, F_1 (1.26/200 g BW/day/head), and G-IV was given formulated okara noodles, F_2 (1.26/200 g BW/day/head). The induction of STZ as a diabetogenic agent was carried out intraperitoneally at a dose of 45 mg/kg BW (body weight) which had been dissolved in citrate buffer as well as 2 mL/200 g of the rat's BW and 15 min previously had been injected with NA at a dose of 110 mg/kg BW intraperitoneally which has a protective effect from the toxicity of NA [12,41]. Blood samples were taken twice from the retro-orbital vein by using the microcapillary method, the 3rd day after the induction of STZ-NA and after the treatment ends to analyze

BGLs & SILs and then went through a centrifugation process for further analysis including measurements of HOMA-IR and HOMA-β. Rats were declared as DM at minimum fasting BGLs of 200 mg/dL [42]. The provision of the noodles diet was carried out by force-feeding. The dietary intervention of the formulated noodles was carried out for 3 weeks. AIN-93M was given every day and the rest of the feed was weighed the next day. Furthermore, every 3 days later BW, BGLs and SILs were analyzed twice, after induction and after the treatment ended (**Figure 1**).

Parameters evaluated

Body weight measurement: Body weight was weighed every 3 days later.

Estimation of BGLs: Analysis of fasting BGLs of samples was carried out repeatedly, during pre-treatment (3rd day after DM induction) and post-treatment (21st day of treatment) quantitatively by using the enzymatic method GOD-PAP (Glucose Oxidase Phenol Aminophenazone) [43]. On day 3rd and 21st day of treatment, the rats fasted overnight for 8 - 10 h and then blood was taken from the retro-orbital vein by using the microcapillary method. Identification of glucose levels of serum was conducted by using an analysis kit from DiaSys consisting of a solution of standards and reagents. The procedure for calculating Fasting BGLs was referred to the research of [44]:

$$\text{BGLs} = \left(\frac{\text{Sample Absorbance (nm)}}{\text{Standard Absorbance (nm)}}\right) \times \text{Standard Glucose Levels } \left(\frac{\text{mg}}{\text{dL}}\right) \quad (1)$$

Estimation of SILs: The 2nd analysis of SILs was carried out at the end of the observation using an ELISA kit (Rat Insulin ELISA Kit) DRG brand with No. EIA 2048 catalog and microplate reader. The procedure for calculating SILs referred to research by Adriawan *et al.* [45].

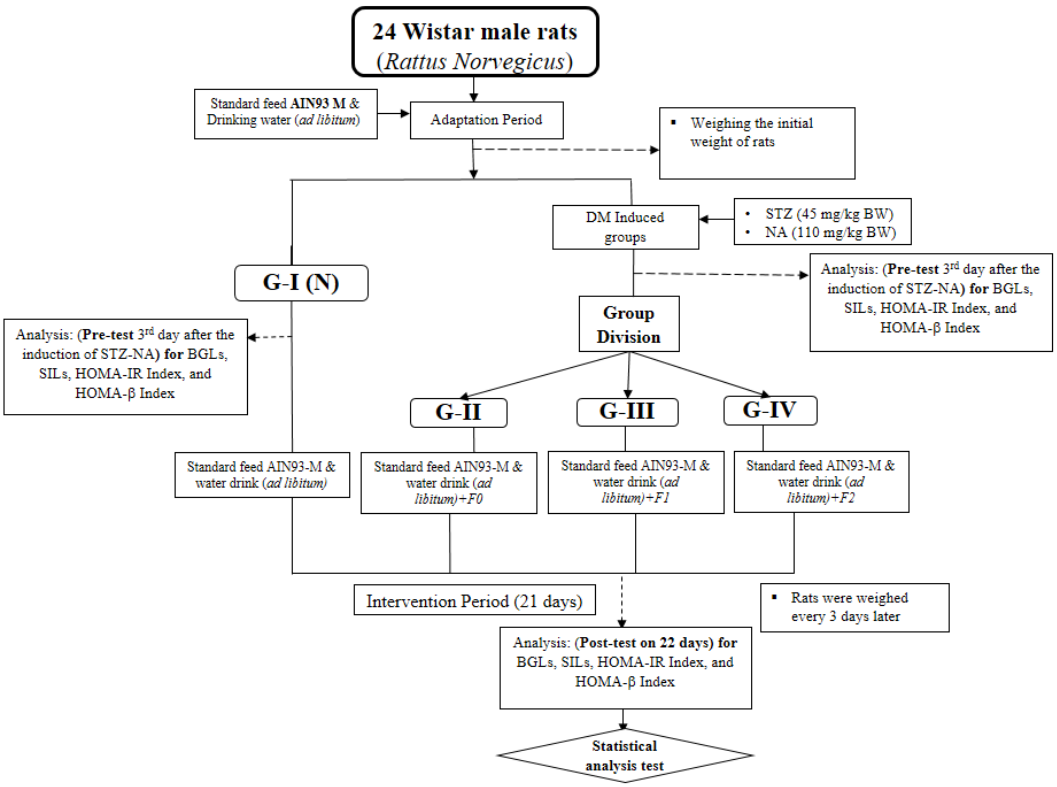

Figure 1 In *in vivo* testing scheme.

HOMA-IR and HOMA-β index calculation:

At the end of the experimental period, terminal blood collected was done after overnight fasting for the determination of Homeostatic Model Assessment (HOMA) score for insulin resistance and β-cell function (HOMA-IR and HOMA-β) which was calculated by using the following formula (2) and (3) [46]. Higher HOMA-IR index mentioned high insulin resistance. The higher value of HOMA-β index indicated the better the cell strength.

HOMA-IR = [(Fasting serum insulin in μU/L×Fasting blood glucose in mg/dL)/405] (2)

HOMA-β = (Fasting serum insulin in μU/L×360/Fasting blood glucose in mg/dL-63) (3)

The procedure for calculating the HOMA-IR and HOMA-β index referred to [47]. HOMA-IR < 4.0 showed normal value and value > 4.0 indicated insulin resistance, while the HOMA-β value which is in the range of 70 - 150 % means normal and if < 70 % means that pancreatic cell dysfunction has occurred [48].

Statistical analysis

Data were generated as mean ± SD. The means of all parameters were examined for significance by using one-way analysis of variance (ANOVA) (IBM SPSS 23.0 Statistical Software Program for Windows) with the Tukey test. A *p*-value < 0.05 was considered as statistically significant [49].

Results and discussion

In *in vivo* analysis on *Rattus norvegicus* rats:

In *in vivo* analysis aim was to determine the anti-diabetic effect of okara noodles in DM rats induced by STZ-NA. In *in vivo* analysis on rats was carried out with the measurement of BW, fasting BGLs, SILs, HOMA-IR and HOMA-β. The dietary intervention of the okara noodles diet was performed for 21 days.

Changes in body weight

The changes in BW of rats were documented to observe the growth condition of DM rats when fed with several formulated noodles in all groups during the intervention period. **Figure 2** showed the changes in BW of rats from the beginning to the end of the intervention period.

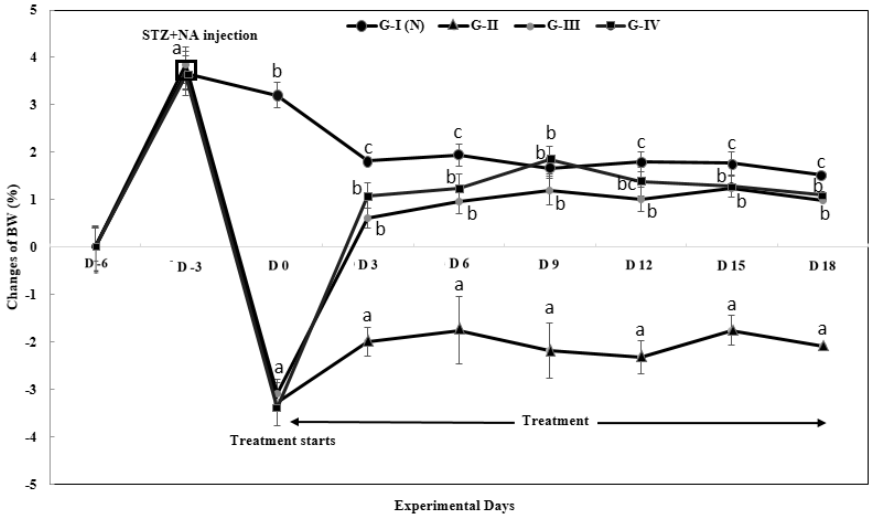

Figure 2 Body weight changes during the intervention period. Note: Values are mean ± SD (n = 24); means with different scripts notated significant differences ($p < 0.05$) between groups in the same time period with Tukey's test. Here, DM: Diabetes Mellitus; OF: Okara Flour; WF: Wheat Flour; F_0: Control noodles (O % OF, 100 % WF); F_1: Okara noodles (25 % OF, 75 % WF); F_2: Okara noodles (45 % OF, 55 % WF); N = Normal or healthy group. Group II (DM + F_0), Group III (DM + F_1), and Group IV (DM + F_2) were provided 1.26 g of noodles per rat's BW (200 g).

Figure 2 demonstrated the BW of G-I (N), G-III (DM + F_1) and G-IV (DM + F_2) groups were not significantly different ($p > 0.05$) at both the initial and final stages. However, the BW of the 3 DM groups was significantly decreased ($p < 0.05$), compared to those of the G-I (N) group due to impaired glucose metabolism in DM rats and several formulated noodles dietary interventions. Our results were similar to the results of [50] that Type-2 DM led to decrease in BW. According to Luo *et al.* and Burhans *et al.* [51,52], in DM patients with abnormal glucose metabolism, energy is accomplished by breaking down protein and AD tissue (adipose tissue) in the body. As a result, there is weight loss in DM patients. In same condition of DM rats, although BGLs increase, glucose cannot be used as an energy source. To get the

energy it needs, the body breaks down protein or AD tissue in the body. This process refers to Gluconeogenesis [53-55].

Changes in BW of rats during the 3 days of the adaptation period indicated BW gain with the same percentage of 4 % in all treatment groups that indicated that the standard feeding was sufficient and nutritional and energy requirements for rats to carry out their activities. Rats decreased their BW at day 0 as much as 4 % experienced by rats in all DM groups, compared to the G-I (N) who experienced weight gain as much as 3 % of BW before STZ-NA induction. STZ causes DNA damage in pancreatic β-cells which reduced insulin production, whereas NA inhibits STZ in damaging DNA, therefore, STZ can only decrease the ability of β-cells to pancreas through decreased insulin sensitivity (IS) [56].

According to Aboonabi *et al.* [57] report, rats in G-I (N) experienced a developing BW during the 3 weeks intervention compared to other groups of rats induced by STZ-NA. Reduced Insulin sensitivity (IS) and Insulin resistance (IR) means glucose in blood cannot enter the cells to be converted into energy. As a result, most cells in the body will produce fatty acids and fatty acids muscle protein for energy. Due to reason, DM loss of BW [52,58]. In the DM rats groups G-III (DM + F_1) and G-IV (DM + F_2) there was an increase in BW of the treatment periods (D 3 to D 18). The weight gain of DM rats in group G-II (DM + F_0) was lower than the weight gain of rats in DM groups G-III (DM + F_1) and G-IV (DM + F_2). While the change in weight gain was the highest experienced by G-IV (DM + F_2) as well as 1.08 % at D 3, 1.25 % at D 6, 1.84 % at D 9, 1.38 % at D 12 and 1.09 % at D 18 at the same dose of feeding (1.26/200 g BW/day/head).

In dry matter basis, okara contains high dietary fibers, DFs (50 %) [52,59,60]. The weight gain of rats after force-feeding treatment in the form of control and okara noodles samples (F_0, F_1 and F_2) based on every 3 days later of BW measurement during the intervention period proved that the consumption of existing DFs in the treatment feed had a positive effect on maintaining BW. DFs can increase IS, resulting, IR decreased and the process of gluconeogenesis can be inhibited. Rats did not lose BW owing to decreasing speed gluconeogenesis which helps to decrease the breakdown of protein and fat in the body. Consumption of DFs-containing food (okara noodles) was able to suppress postprandial blood sugar rises and increase IS, compared with the G-I (N) during the intervention period always experiences weight gain due to inadequate intake of nutrients and energy fulfilled from the standard feed, the DM group experienced a decrease in BW during the intervention period. However, no significant difference was observed between G-III (DM + F_1) and G-IV (DM + F_2) treatment groups at the same dosages of feeding whereas G-II (DM + F_0) was a significant difference. These results showed the treatment of okara noodles at (1.26/200 g BW/day/head) managed to maintain the body weight of DM rats.

Fasting blood glucose levels

The fasting BGLs test is the most important test for the DM. This test aimed to determine the effect of feed containing noodle samples on changes in BGLs of Type-2 DM rats during the treatment period. Measurement of BGLs was performed for 2 times, before the intervention and after the intervention period. Changes in BGLs in G-I (N) rats and DM treatment groups (G-II, G-III and G-IV) were mentioned in **Figure 3**(**a**).

Past findings suggested, okara and related food products had glucose metabolic property [61-63]. mentioned that fasting BGLs differences of rats during the 3 weeks. Comparing to G-I (N) rats, all of other DM groups of rats had significantly ($p < 0.05$) higher Fasting BGLs (> 250 mg/dL) in the range of 266.43 ± 2.91 to 270.97 ± 4.67 mg/dL before the intervention, which was much higher that indicated all DM rats were successfully STZ-NA induced [64]. After 1 week of intervention, G-III and G-IV groups showed significantly ($p < 0.05$) decrease in Fasting BGLs in comparison to the G-II group and G-I, but there were no significant difference between these okara noodles groups themselves. Our results indicated the similar trends that all okara noodles groups had significantly ($p < 0.05$) lower Fasting BGLs than control noodle samples rats, but with no significant ($p > 0.05$) difference among the 2 groups for every 3 days later. This result revealed, there is an influence positive result of feeding treatment in the form of okara noodles during the intervention period. However, after 1 week of intervention, all of the control noodles feeding groups of rats (G-II) still had significantly ($p < 0.05$) higher fasting BGLs (271.31 ± 1.92). The average BGLs of G-I (N) in the pre-treatment period was 66.73 mg/dL. This implied that the rats used in this study have normal fasting BGLs. The analytical outcome for each group of rats during the 3 weeks of the intervention period illustrated the decrease in BGLs for the treatment group of G-III and G-IV is different significantly that proved the treatment of giving okara noodles with the same doses has the ability to lower BGLs. BGLs in the administration group by the same doses of control and okara noodles for 21 days of treatment were 271.31 ± 5.77, 131.23 ± 3.38 and 119.21 ± 2.23, respectively. This result showed that G-IV has the highest capability to decrease BGLs compared to other groups due to higher content of DFs (okara noodles, where

OF was 45 %). Gowd *et al.* [65] and Capuano [66] stated, there is a correlation between the hypoglycemic properties and insoluble DFs that increase in viscosity in the digestive tract which is considered to be the main factor affecting the rate of glucose absorption. DFs contenting foods have an effect on the viscosity and absorption of sugar that affects starch digestibility and blood sugar-lowering activity. DFs can be fermented as well and gave high viscosity in the large intestine that has the potential role to reduce the glycemic response and increase IS [67,68].

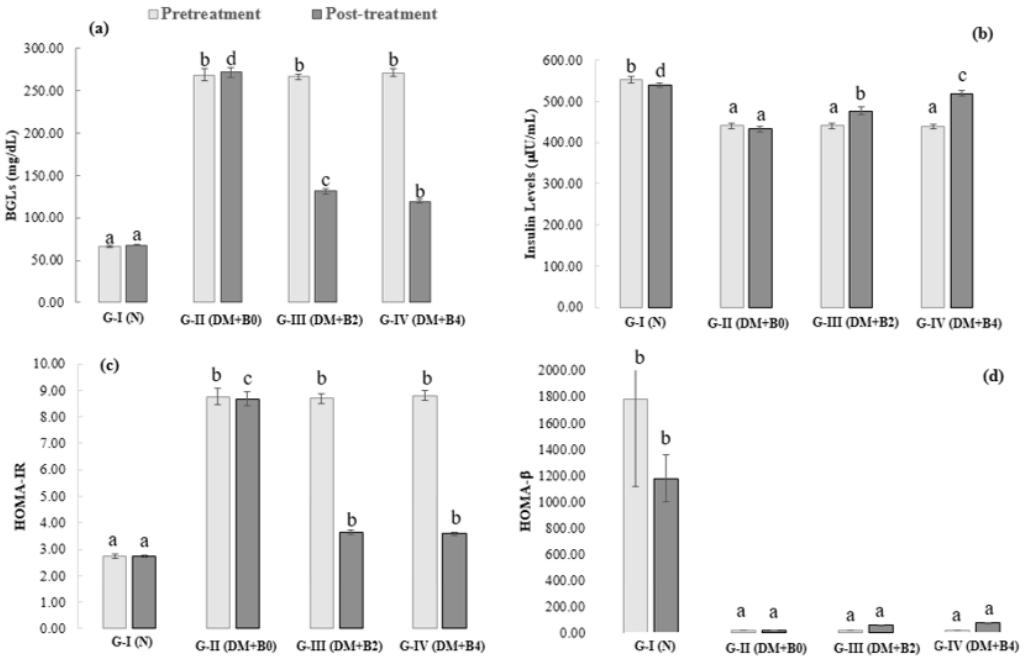

Figure 3 Effects of okara noodles on blood glucose levels, serum insulin levels, HOMA-IR and HOMA-β. Note: Different letter notation on the bar with the same color shows a real difference. A p-value < 0.05 with Tukey test was considered as statistically significant.

Serum insulin levels

Fasting SILs is also an important analysis related to DM. The aim of this testing was to determine the effect of okara noodles at the same doses (1.26/200 g BW/day/head) on SILs in DM rats during the intervention period. From **Figure 3**(**b**), changes in SILs in individual groups during the treatment period can be seen that with the consumption of okara noodles, for 21 days can increase insulin levels in DM rats. The highest level of insulin was increased in the G-IV group (DM + F_2, OF: 45 % and WF: 55 %) due to high DF-containing formulated okara noodles, which was higher than the other DM groups, where this value was much lower compared to the normal/control G-I (N) group.

The correlation between DFs and Type-2 DM was conducted in many meta-analyses, in past decades [65,66]. A higher DFs diet has positive effects in the treatment of Type-2 DM [66,67,69], as DFs diminishes postprandial hyperglycemia by delaying the digestion and absorption of carbohydrates with maintaining BW. In IR individuals, DFs can possibly enhance peripheral IS through short-chain fatty acids (SCFAs), which are produced by the fermentation of DFs in the gut [70]. During DFs mechanism, it cannot be digested by digestive enzymes because DFs enter to the large intestine (LI) intact. DFs intact in the LI where DFs fermented by bacteria in the LI to form SCFAs including propionate, acetate and butyrate [9,71]. A decrease in hepatic glucose production happen due to increase in SCFAs. The formation of SCFAs is thought to trigger the generation of the hormones Glucagon such as Gastric Inhibitory Polypeptide (GIP), Peptide-1 (GLP-1) and Peptide YY (PYY) that increases IS and finally conduce to decreased IS. GLP-1 is an incretin hormone that can stimulate the release of insulin by pancreatic β-cells, whereas PYY is an intestinal hormone that acts as a role in creating a feeling of fullness [9,72,73].

In general, damage to the islets of Langerhans cells (a type of immune cell) in the pancreas, which is responsible for producing insulin, resulting insulin insufficiency, which is the major cause of DM. The low SILs in DM rats in recent study after STZ-NA induction could be caused by a decrease in the ability of insulin in peripheral tissues (insulin resistance) and β-cell dysfunction which resulted in the pancreas not

being able to produce enough insulin to replace insulin resistance, causing insulin deficiency, inhibiting insulin secretion, insulin resistance and synthesis [74,75]. Insulin has the ability to lower blood sugar in several mechanisms including stimulating the uptake of glucose from the blood by tissue cells for storage or glycolysis by the liver as the form of glycogen [76,77]. Insulin inhibits the hormone glucagon to inhibit glucose production in the kidney and liver [72,77,78]. This is linked to fasting glucose levels. If fasting blood insulin levels are high, then fasting BGLs will be low. Because insulin is a hormone that plays an important role in regulating blood sugar balance. Under normal/controlled conditions, blood glucose promotes the synthesis and secretion of insulin by pancreatic cells.

HOMA-IR and HOMA-β index

HOMA, is a statistical method for assessing insulin resistance (HOMA-IR) and pancreatic β-cell function (HOMA-β) of basal glucose and insulin or c-peptide [79,80]. The correlation between insulin and glucose in basal conditions reflect the balance between hepatic glucose output and secretion insulin that maintained by a feedback loop between the liver and β-cells pancreas. HOMA linked between blood glucose and insulin secretion predicting long-term fasting glucose and insulin concentrations of possible combination of insulin resistance and pancreatic β-cell function [46,81]. SILs are influenced by the effect of pancreatic β-cell on glucose concentration whereas glucose concentration is regulated by glucose production which is mediated by insulin through the liver. Therefore, decreased β-cell function will give less response to β-cell for insulin secretion stimulated by glucose. Insulin resistance (IR) is a condition characterized by insulin intake decreased into cells occurred in patients with Type-2 DM due to reducing the number of receptors that must bind to insulin or the receptors are not functioning appropriately, there is a disturbance in the insulin transport system in the cell to the burning site. Intake of excessive glucose may result in an increase in the number of fat cells, resulting in BW gain and adiposity.

HOMA can be used to estimate insulin sensitivity with the formula of fasting BGLs and serum insulin concentrations. Insulin resistance (HOMA-IR) and pancreatic-βcell (HOMA-β) functions were calculated by using fasting BGLs and fasting SILs (using different calculations). The HOMA method is a reliable and useful parameter to measure insulin sensitivity and function of pancreatic β-cells in DM patients treated with insulin or without insulin due to its simplicity, reproducibility, and good correspondence with other techniques [82].

HOMA-IR index

All changes for the HOMA-IR index before and after treatment for each group of rats can be seen in **Figure 3**(**c**) that showed the consumption of okara noodles at the same doses for 3 weeks can decrease the value of HOMA-IR in the group of DM rats. The mean value of HOMA-IR in the DM rat groups for G-III and G-IV was lower compared to the DM group G-II without okara noodles. This figure also indicated there were significant differences ($p < 0.05$) between before and after the intervention period with the provision of okara noodles. In all DM treatment groups, the HOMA-IR value > 4 indicated that insulin resistance has occurred in all groups at the time before the intervention period. After the intervention period, administering the same dose of okara noodles (G-III and G-IV) had HOMA-IR values less than 4.0 (3.63 and 3.58, respectively) which proved the normal grade, whereas the same dose of control noodles was given DM group G-II, HOMA-IR value was greater than 4.0 (8.68) which indicated insulin resistance in DM groups with control noodles was still higher. Increased DFs diet affects the reduction of insulin resistance and improve insulin sensitivity as indicated by a significant decrease in HOMA-IR [82,83]. **Figure 3**(**c**) reveled that there is a strong linkage between HOMA-IR and Insulin levels.

Insulin resistance interfere the use of blood glucose by the body's cells, resulting glucose accumulated in blood. Under conditions of insulin deficiency can cause decreased translocation of GLUT-4, failure of IRS (Insulin Receptor Substrate) complex phosphorylation, and glucose oxidation which causes glucose cannot enter into the cells (only stays in the blood) (hyperglycemia) [84]. Simultaneously, the onset of Type-2 DM is a surge in blood sugar concentration compared to normal, which is still accompanied by high insulin secretion (hyperinsulinemia). That indicated the defects in both insulin receptors and post-receptors. In insulin resistance, there is an increase in glucose production and a decrease in glucose use resulting in an increase in blood sugar levels (hyperglycemic) [85].

In Type-2 DM, insulin resistance, dysfunction occurs in pancreatic β-cells [86,87]. Cells undergo rapid apoptosis due to metabolic alterations of mechanisms including lipotoxicity, glucotoxicity, mitochondrial dysfunction, glucolipotoxicity and stress oxidative stress [87,88]. The advanced stages of Type-2 DM are the damage to pancreatic β-cells and their function is replaced by amyloid tissue that results in absolute insulin deficiency [89,90].

HOMA- β index

HOMA-β is one of the parameters to measure the level of the strength of the pancreatic β-cells that produce insulin. HOMA-β index indicated the strength of pancreatic β-cells, higher HOMA-β level means cells are better strength. In our research we used the HOMA-β model to monitor the successful effect of the consumption of okara noodles with an increase in pancreatic β-cell function. Changes of HOMA-β index before and after treatment in each group of rats can be seen in **Figure 3**(**d**).

This figure explained there is a significant difference ($p < 0.05$) between before and after the intervention period by giving different noodles samples. In all DM groups (G-II, G-III and G-IV) without noodles samples indicated the HOMA-β value is below 70 %, which proved the pancreatic β-cell dysfunction had occurred in all DM groups before the intervention period. After the intervention period, the administration of okara noodles (G-III and G-IV) at the same doses had a HOMA-β value greater than 70 %, while the DM group (G-II) with control noodles at same doses had a lowest HOMA-β value (< 70 %). Consumption of okara noodles with same doses for 3 weeks can increase the HOMA-β value in DM rats. During the intervention period, increased the value of HOMA-β reported that the DM groups G-III and G-IV separately successively while compared to the DM group G-II. Increased the value of HOMA-β in the treatment groups (G-III and G-IV) with same doses were 75.51 ± 3.44 and 99.88 ± 4.077, respectively. This report supported by Mohammed *et al.* [50] that increase the value of HOMA-β index (β-cell function) for the intervention group (DM rats) showed progressively result. This result proved that the treatment feed (okara noodles) contains DFs which plays an important role in increasing the strength of pancreatic β-cells, approaching normal group.

Conclusions

Okara contains high protein and dietary fiber, low in calories, and also considerable quantities of other nutrients which is particularly effective for DM patients. Dietary-based therapy may be an effective alternative for controlling and preventing DM and the complications that are associated with it. The outcome of the current study suggested, okara noodles significantly benefited reduced body weight, decreased BGLs, and enhanced SILs. Therefore, okara noodles may be applied as a dietetic superior for the management of Type-2 DM. This research also demonstrated okara noodles have health-beneficial effects for anti-diabetic activity in pre-clinical (*in vivo*) experiments. Consequently, further research at the clinical level can provide input on nutritional therapy in the form of suitable snacks for patients with Type-2 DM as well as the perfect utilization can be recommended of local functional food ingredients from by-products (okara) in the food industry.

Ethical approval

This research was permitted by the Ethical Clearance Commission number 00021/04/LPPT/VII/2022 and issued by the Integrated Research and Testing Laboratory, Gadjah Mada University (UGM), Indonesia.

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