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1 Profile of cholesterol and glucose on the various age of volunteers with composite flour diet

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Abstract. The preliminary research showed that the glycemic index (GI) group of the composite flour made of Growol cassava and mung bean flour as raw material of artificial rice was low, so the composite flour might be potential for reducing blood glucose. This research is aimed to determine the profile of cholesterol and glucose level on the various age of volunteers with composite flour diet. The treatment of the research was the age group of volunteer and the diet treatment. There were three group volunteers based on the age, that were 15 - 26; 27 - 38; and 39 - 50 years old. The volunteers were adapted for 4 days with steamed rice diet as a normal diet, then were treated with composite flour as diet treatment for 16 days. The composite flour was formed like rice and steamed before used as the treatment. The blood trygliseride, total cholesterol, LDL and HDL cholesterol, and glucose level of the volunteers were analysed on 4th, 8th, 12 th, and 16th days for the treatment and before the treatment as control (0th). The result of this research showed that the potency of hypocholesterolemic of the diet treatment were shown by decreasing of blood LDL cholesterol at the age group of the 39 - 50 years old. The potency of hypoglycemic of the treatment were shown by decreasing of blood glucose level of volunteers at the all of age group. That was indicated that the composite flour might be potential to normalize cholesterol and glucose.

1. Introduction

staple food as the main source of energy that could not be substituted with the others, and must be eaten three times a day that has been the habit of Indonesian people. However the Indonesia rice production did not make adequate the need of rice consumtion, so Indonesia still imported rice. The researchers have developed composite flour from many agriculture product as raw material of artificial rice [1]. A lot of composite flour was made of original cassava that was only dried and milled for making the flour, so the GI of this composite flour was still hight. The GI group of rice and cassava were hight (>70), these were 80 and 73 respectively [2]. This was the problem for society with high risk of diabetes that continually increased because the change of diet and obesity of modern society. There were many traditional food product in Indonesia that had been used as staple food, for example *Growol*. Growol was traditional food from Kulonprogo Yogyakarta Indonesia that provided energy

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especially in dried climate condition at the high price of rice. Growol cassava flour was produced by spontaneous fermentation of cassava in water for 3 - 5 days, and then the fermented cassava was pressed to remove water, dried and milled [3]. The same product as Growolin the other country is gari or rale that is fermented cassava using lactic acid bacteria [4]. The protein of Growol cassava flour was lower than rice, so the addition of legumes flour is important to increase protein of *Growol*. In the preliminary research was known that the composite flour from Growol flour with addition of mung bean flour 30% could increased protein, was the same as rice. Based on this research also known that the GI of composite flour as the raw material of artificial rice was 53,83 that was lower than natural steamed rice [3]. The legumes protein had hypoglycemic property [5–7] and contained high of arginine [6] that was known potential for stimulation of insulin secretion [8]. The specifics amino acids especially Arg and Leu can activate mitochondrial metabolism in pancreatic β-cell via the tricarboxylic acid (TCA) cycle, resulting in the formation of ATP. The rise in ATP levels leads to closure of ATP-dependent K+ channels, which in turn depolarizes the cell membrane, thus opening of voltage-dependent Ca2+ channels and increasing intracellular Ca2+ concentration, which triggers insulin exocytosis and hence facilitating insulin secretion from pancreatic β-cell [8,9]. Some amino acids were hypercholesterolemic property especially lysine, and some amino acids were hypocholesterolemic property especially arginine. The ratio of arginine/lysine was important to controle the increasing of cholesterol level that had been indicated the in vivo bioassay showed the cholesterol level of soy protein was lower than casein, because the ratio arginine/lysine of soy protein was higher than casein these were 1,57 and 1,22 respectively [10]. This research was aimed to determine the profile cholesterol and glucose of the various age volunteers with the composite diet made of Growol cassava and mung bean flour.

2. Material and Methods

Mung bean and cassava were obtained from Beringharjo and Karangkajen market in Yogyakarta. Chemical agents for analysis of blood kholesterol and glucose level were purchased from Yogyakarta Health Laboratory, Jalan Ngadinegaran, MJ III No.62, Mantrijeron, Kota Yogyakarta, Daerah Istimewa Yogyakarta 55143 The sequences of experimental activities were performed as follows:

2.1. Preparation of composite flour and sample of treatment diet

The composite flour were prepared by mixing mung bean flour and cassava *Growol* flour. Growol was made of cassava through many step process [3]. The first step, cassava was pelled, washed, and size reduced. After that cassava was fermented by soaking in water for 5 days. Then the fermented cassava are pressed to remove water and dried. The dried cassava was milled and mixed with 30% mung bean flour. The composite flour was formed like rice and steamed for preparing artificial steamed rice sample as the treatment diet. The treatment diet was consumed by volunteers in the following experimental activity. The proximate composition of the composite flour diet that were water, ash, fat, protein and carbohydrate (by difference), were 9.77; 1.23; 0,54; 6.53; and 81.92 respectively [3].

2.2. Blood cholesterol and glucose level bioassay of volunteers

This experiment was done with 39 volunteers that were divided in 3 group based on their age, these were 15-26, 27-38 and 39-50 years old. The breakfast diet of the volunteers were arranged for 16 days that were divided in 2 step, these are adaptation step and then treatment step. The volunteers consumed natural steamed rice as normal diet for 4 days in the adaptation step, and then the volunteers consumed the steamed composite flour as the treatment diet for 12 days, so the total duration of the experiment was 16 days. The weight of normal or treatment diet as a breakfast was 100g per portion. The volunteers were injected at 2 hours after breakfast for taking their blood on 4th, 8th, 12th, 16th days and before treatment diet as control (0th). The blood of the volunteers were analysed the triglyceride, cholesterol total, High Density Lipoprotein (HDL) cholesterol, Low Density Lipoprotein (LDL) cholesterol and glucose level. The experimental design of this research was randomized complete design with 2 factors. The first factors were the various age of volunteers, these were 15-26, 27-38 and 39-50 years old. The second factors were the duration of normal and

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treatment diet, these were 0th, 4th, 8th, 12th, and 16th days. The data of this experiments was statistical analysed by Anova (analysis of varian) and DMRT (Duncan Multiple Range Test).

3. Result and Discussion

3.1. Tryglicerid

Table 1 showed that the trygliceride of the various age of volunteers were no significant differences. The normal trygliceride of human according to US National Cholesterol EducationProgram (NCEP) was < 150 mg/dl [1]. Based on this data indicated that composite diet could inhibit increasing trygliceride although there were not significant.

Table 1. The effect of composite flour diet on the blood tryglyceride of volunteers (mg/dl)*

The age of volunteers	0 th day	4th days	8 th days	12th days	16th days
15-26 years old	102.00 ^a	106.86a	71.00 ^a	70.33 ^a	75.33 ^a
27-38 years old	126.00^{a}	132.00 ^a	112.00 ^a	100.67^{a}	107.33^{a}
39-50 years old	143.33a	116.67ª	80.67ª	90.67ª	72.33a

^{*)} The same notation of statistic in the table showed not significantly differences at the different column and row

3.2. Total cholesterol

The diet treatment affected on the total cholesterol of the various age of volunteers, that was seen at Table 2. The decreasing total cholesterol during the treatments were not significant, except between the diet treatment 8th and 12th days on 15 – 26 years old with before the diet treatment (0th days). The data indicated that composite flour was potential to control cholesterol level of volunteers. The cholesterol level of all volunteers were still normal because there were lower than the total cholesterol standar of human according to US National Cholesterol Education Program (NCEP), that was < 200mg/dl.

Table 2. The effect of composite flour diet on the blood total cholesterol of volunteers (mg/dl)*

The age of volunteers	0 th day	4th days	8th days	12th days	16 th days
15-26 years old	183,00 ^{ab}	183,67 ^{ab}	157,00ª	158,33ª	163,33ab
27-38 years old	174,33ab	177,67ab	167,33ab	164,67ab	170,00 ^{ab}
39-50 years old	195,67 ^b	190,67ab	171,67 ^{ab}	182,33ab	168,33ab

^{*)} The same notation of statistic in the table showed not significantly differences at the different column and row

3.3. HDL cholesterol

Decreasing HDL of the volunteers could be avoided by treatment diet, that was seen in Table 3. The HDL of all volunteers were not significant. The normal HDL cholesterol of human according to US National Cholesterol Education Program (NCEP) was > 60mg/dl. The data indicated the HDL choletsreol of all volunteers were abnormal. The trend of HDL choleterol level of the treatments increshed, compared to before the treatment (0th days). Legumes in the composite flour might be role in this phenomena. The result conformed with [11] who showed that protein isolate of soybean feed treatment could increase 50% HDL and decrease 30-40% total cholesterol. According to [7] showed that protein isolate of cowpeas sprout could increase HDL through in vivo bioassay.

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Table 3. The effect of composite flour diet on the blood HDL cholesterol of volunteers (mg/dl)*

The age of volunteers	0 th day	4th days	8th days	12 th days	16 th days
15-26 years old	47,33ª	50,00a	48,67ª	50,00°	46,00°
27-38 years old	36,33a	36,33ª	41,67 ^a	45,00°	42,33 ^a
39-50 years old	38,33a	40,67 ^a	44,00 ^a	46,00°a	43,33 ^a

^{*)} The same notation of statistic in the table showed not significantly differences at the different column and row

3.4. LDL cholesterol

The LDL of all volunteers were abnormal that was seen at Table 4, The normal LDL cholesterol of human according to US National Cholesterol Education Program (NCEP) was < 100mg/dl. The data showed that the treatment diet affected on the LDL cholesterol level that decreased significantly at 39 – 50 years old. The result indicated that composite flour, that was made of *Growol cassava* and mung bean flour, could inhibit the increase of LDL cholesterol. The phenomena might be the role of arginine that was hightly contained in protein of legumes. Arginine had been known hypocholesterolemic property [10].

Table 4. The effect of composite flour diet on the blood LDL cholesterol of volunteers (mg/dl)*

The age of volunteers	0 th day	4th days	8th days	12th days	16 th days
15-26 years old	110,00a	111,33ª	99,00ª	$99,00^{a}$	102,33ª
27-38 years old	111,67ª	109,67a	103,33ª	$101,00^{a}$	110,33ª
39-50 years old	150,33 ^b	129,00ab	106,33a	116,33 ^a	110,67 ^a

^{*)} The same notation of statistic in the table showed not significantly differences at the different column and row

3.5. Glucose

Table 5 showed that blood glucose level of all volunteers were 63,67 – 98,33 mg/dL. This indicated that all volunteers were normal or not diabetic. The glucose level of human diabetic condition was higher than 180mg/dL [12]. The glucose level of treatment diet was lower than normal diet. The data showed that composite flour decreased glucose level of voluneteers significantly, especially at 27 – 38 years old after 8th days and 39 – 50 years old after 12th days. The preliminary research had been known that the GI group of this composite flour as raw material of artificial rice was low [3].

Table 5. The effect of composite flour diet on the blood glucose of volunteers (mg/dl)*

The age of volunteers	0 th day	4th days	8th days	12th days	16 th days
15-26 years old	88,00 ^{cde}	91,33°	63,67ª	68,33 ^{ab}	72,00 ^{abcd}
27-38 years old	89,00 ^{de}	91,67°	83,67 ^{bcde}	73,33abcd	70,67abc
39-50 years old	94,33°	98,33°	81,00 ^{abcde}	74,00 ^{abcd}	80,67abcde

^{*)} The same notation of statistic in the table showed not significantly differences at the different column and row

4. Conclusion

The potency of hypocholesterolemic of composite flour were shown by decreasing of LDL cholesterol at 39 - 50 years old volunteers, and the potency of hypoglycemic of composite flour were shown by decreasing of blood glucose level for all volunteers especially at 27 - 38 years old after 8th days and 39

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- 50 years old after 12th days. This result indicated that composite flour, that was made of *Growol cassava* and mung bean flour might be used to prevent diabetic and its complication that was caused by cholesterol.

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