

Differences in triglyceride levels before and after whey protein intervention in field workers exposed and unexposed to arsenic

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ABSTRACT

Background: A work environment that is exposed to heavy metals, such as a coal mining environment, can change fat metabolism in the body. Changes in fat metabolism will lead to cardiovascular disease. Consumption of dairy products, e.g. whey protein, can reduce the risk of metabolic disorders and cardiovascular disease.

Objectives: To test and analyze the differences in triglyceride levels before and after whey protein intervention to field workers with different working conditions.

Materials and Methods: This experimental research with pretest and posttest was conducted on field workers at PT Bukit Asam Tbk. Tarahan Port Unit, Lampung as an Arsenic exposed group and PTPN VII Way Berulu Business Unit as an Arsenic unexposed group. Both groups received 24 grams of whey protein daily for 28 days. The triglyceride levels before and after the intervention were measured by laboratory analysis using the enzymatic calorimetry method. The data were analyzed using independent t-test, Mann Whitney test, and Wilcoxon test.

Results: The average triglyceride levels increased by 50.48 ± 98.09 mg/dL in the exposed group and 16.78 ± 67.67 mg/dL in the unexposed group. There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups.

Conclusions: The whey protein intervention increased the triglyceride level in the exposed group and decreased it in the unexposed group.

Keywords: Triglycerides; Whey Protein; Arsenic (As).

BACKGROUND

With a good nutritional status and health status, the workforce will provide maximum results for the company. The work environment greatly influences their nutritional needs and health status. The work environment with exposure to chemical substances will affect the health status of the workers if it is not properly attended to by the manager.¹ Mining work environment, such as coal mining, is susceptible to the exposure of hazardous chemicals. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.^{2,3} Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. It can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.⁴

Some epidemiological studies have shown that the consumption of dairy products can reduce the

risk of metabolic disorders and cardiovascular disease.⁵ Protein in cow's milk consists of 80% casein and 20% whey protein, both of which are rich in amino acids.⁶ Whey protein is a food source that has high protein and essential amino acids, as well as branched-chain amino acids (BCAA) that play an important role in tissue growth and repair.⁵ The results of the previous studies indicate that after the intervention of whey protein on 20 male subjects aged 30 to 50 years with relatively high serum cholesterol levels (≥ 200 mg/dL), their serum triglyceride levels decreased and their serum HDL levels increased after 8 weeks of intervention.⁷

MATERIALS AND METHODS

This research is an experimental study with pretest and posttest. The sample was assigned into two groups, Arsenic (As) exposed group (field workers of PT Bukit Asam Tbk. Tarahan Port Unit, Lampung, a coal mining product management company) and Arsenic (As) unexposed group (field workers of PT Perkebunan Nusantara VII Way Berulu Business Unit, a company engaged in the

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management of rubber plantations). Both groups received intervention in the form of taking whey protein for 28 days⁸ with a dose of 24 grams per day dissolved in 300 ml of mineral water.⁹ The variable measured was the subjects' triglyceride levels before and after the whey protein intervention.

This research was conducted after obtaining permission from the Health Research Ethics Committee of the Faculty of Medicine, Diponegoro University, Semarang, from January to March 2020. Whey protein intervention was carried out at the research site, PT Bukit Asam Tbk. Tarahan Port Unit in Lampung and PT Perkebunan Nusantara VII Way Berulu Business Unit. Subject characteristic data including name, age, and place of birth were collected through interviews using a questionnaire. The anthropometric data of the subjects consisted of body weight, measured using digital scales, and height, measured using a microtoise. Body mass index (BMI) was calculated using the BMI calculation formula based on measurements of weight and height. The physical activity data of the subjects were collected through interviews using the Physical Activity Level (PAL) questionnaire and then calculated with the PAL calculation formula. Food intake data were collected through interviews using a 24-hour food

recall questionnaire 3 times each week during the study. The triglyceride levels before and after the intervention were measured by laboratory analysis using the enzymatic calorimetry method.

Data analysis was carried out through the SPSS program with the Shapiro-Wilk test to determine the normality of data distribution for each group, independent t-test, and Mann Whitney test to determine the average difference between the subject's characteristics and nutritional intake variables. Wilcoxon test was also used to determine differences in triglyceride levels before and after the intervention.

RESULTS

The data in this study are primary data taken from the research sample, field workers exposed to different work environments. The total sample consisted of 64 people (31 from the exposed group and 33 from the unexposed group).

Subject Characteristics

Subject characteristics data consisted of age, body weight (BW), body height (BH), body mass index (BMI), and physical activity level (PAL). They are shown in Table 1.

Table 1. Subject Characteristics in Both Groups

	Exposed (n = 31)		Unexposed (n = 33)		p
	Mean±SD	minimum-maximum	Mean±SD	minimum-maximum	
Age (year)	29.84±8.84	20-56	38.94±4.50	28-54	0.000 ²
Body weight (kg)	70.87±14.11	45-109	56.55±6.96	44.08-75.00	0.000 ¹
Body height (cm)	168.03±5.89	154-179	162.01±4.84	153-171	0.000 ¹
BMI (kg/m²)	25.31±4.21	18.09-36.84	21.55±2.26	17.56-26.23	0.000 ¹
PAL (unit)	1.57±0.13	1.41-1.87	1.99±0.24	1.44-2.78	0.000 ²

¹ difference test using Independent T-Test, ² difference test using Mann Whitney

Table 1 shows significant differences in all variable subject characteristics ($p < 0.05$). The average weight, height, and BMI were higher in the exposed group compared to the unexposed group, whereas the average age and PAL were higher in the unexposed group.

Nutritional Intake

Nutritional intake data consists of total intake per day and intake adequacy. Nutritional intake data are presented in Table 2 and the data on the intake adequacy levels in both groups in Table 3.

Table 2. Overview of Total Nutritional Intake Per Day in Both Groups

	Exposed (n = 31)		Unexposed (n = 33)		p
	Mean±SD	minimum-maximum	Mean±SD	minimum-maximum	
Energy (kcal)	2018.72±452.88	1313.50-3314.90	2149.83±515.61	1213.20-3890.00	0.285 ¹
Protein (g)	96.21±29.04	56.10-180.90	86.62±26.66	56.30-167.30	0.116 ²
Fat (g)	83.89±40.50	35.80-153.30	67.01±30.06	22.30-170.50	0.134 ²
Carbohydrate (g)	252.30±68.47	148.7-452.80	298.47±66.39	191.80-485.90	0.004 ²

¹ difference test using Independent T-Test, ² difference test using Mann Whitney

Table 2 shows that the average energy and carbohydrate intake in the unexposed group was higher than those of the exposed group. The average protein and fat intake show a higher value in the

exposed group. There was no significant difference in the average energy, protein, and fat intake, but there was a significant difference in the average carbohydrate intake ($p = 0.004$).

Table 3. Nutritional Intake Adequacy Level in Both Groups

Adequacy Level	Category	Category	
		Exposed (n = 31)	Unexposed (n = 33)
Energy	Deficient	23 (74.2%)	26 (78.8%)
	Sufficient	8 (25.8%)	7 (21.2%)
Protein	Deficient	14 (45.2%)	21 (63.6%)
	Sufficient	17 (54.8%)	12 (36.4%)
Fat	Deficient	13 (41.9%)	22 (66.7%)
	Sufficient	15 (48.4%)	9 (27.3%)
	Excessive	3 (9.7%)	2 (6.1%)
Carbohydrate	Deficient	28 (90.3%)	26 (78.8%)
	Sufficient	3 (9.7%)	7 (21.2%)

Table 3 shows that almost all of the adequacy levels of energy, protein, fat, and carbohydrate intakes in both groups were insufficient. Meanwhile, the fat adequacy of 9.7% of the total subjects in the exposed group and 6.1% in the unexposed group fell into the excessive category.

Triglyceride Levels Before and After Whey Protein Intervention

The triglyceride levels before and after whey protein intervention were tested for normality using the Shapiro Wilk test, resulting in the abnormal distribution of triglyceride levels with a p-value of < 0.05 ; thus, the Wilcoxon test was used to see the differences in triglyceride levels before and after the whey protein intervention.

Table 4. Triglyceride Levels Before and After Whey Protein Intervention in Both Groups

Triglyceride Levels (mg/dL)	Exposed (n = 31)		Unexposed (n = 33)		p
	Mean±SD	minimum-maximum	Mean±SD	min-max	
Before	125.32±68.97	35.00-310.00	157.18±104.82	38.00-562.00	0.202 ¹
After	175.80±127.99	48.00-590.00	140.39±80.78	43.00-425.00	0.344 ¹
Difference/delta (Δ)	50.48±98.09	414.00(-68.00)	-16.78±67.67	184.00(-137.00)	0.000 ¹
p	0.003 ²		0.038 ²		

¹ difference test using Mann Whitney, ² difference test using Wilcoxon

Table 4 shows that the average triglyceride level before the intervention in the unexposed group was higher than in the exposed group, and the average triglyceride level after the intervention in the exposed group was higher. The average

triglyceride level in the exposed group increased by 50.48 ± 98.09 mg/dL and in the unexposed group decreased by 16.78 ± 67.67 mg/dL. The Wilcoxon test results showed differences in triglyceride levels in both groups ($p < 0.05$).

DISCUSSION

This study explores the differences in triglyceride levels in field workers with different work environment exposures before and after the whey protein intervention. The groups in this study consisted of field workers in a coal mining environment with exposure to Arsenic, an obesogenic substance, and field workers in a rubber plantation environment. Whey protein intervention was carried out for 28 days with a dose of 24 grams per person per day.

The results showed that the intervention could reduce triglyceride levels in the obesogen unexposed group. Meanwhile, in the exposed group, there was an increase in triglyceride levels after the intervention. The decrease in triglyceride levels in the unexposed group was due to several factors, such as lower BMI value, higher PAL value, and lower fat intake of the unexposed group compared to those of the exposed group.

On the other hand, the triglyceride level in the exposed group was found to increase after the whey protein intervention. This is due to several factors, including higher BMI value, lower PAL, and higher

fat intake, as well as Arsenic exposure that occurs every day in the work environment of the exposed group.

The BMI value of the unexposed group was lower than that of the exposed group. Previous research has suggested that there is a positive correlation between BMI and triglyceride levels. High BMI is directly proportional to triglyceride levels in the body.¹⁰ The linear relationship between BMI and triglyceride levels is seen in individuals with higher BMI between 18 to 35 kg/m², posing a 5.1% greater risk of having high triglyceride levels.¹¹ The PAL value of the exposed group was lower than that of the unexposed group. Previous research found that moderate to heavy PAL could increase the average HDL level between 0.89 to 1.71 mg/dL and reduce the average triglyceride level between 0.93 to 0.98 mg/dL.¹² Besides, fat intake also greatly affects triglyceride levels in the body. Based on the results of the 3×24 hour recall in the study, fat intake in the exposed group was higher than that in the non-exposed group. The results of statistical tests also indicate the same, where the average fat intake of the exposed group is higher than that of the unexposed group. Previous research has shown that an increase in fat by 10% of daily needs will lead to increased triglyceride levels by 0.06 mmol/L.¹³

Arsenic exposure that occurs every day in the work environment of the exposed group also influences changes in lipid metabolism in the body. Based on a study conducted on ApoE mice exposed to moderate Arsenic, it was found that mice with a normal diet and received 200 ppb-Arsenic exposure for 8 weeks had a significant increase in plasma triglyceride levels and triglyceride levels. The average triglyceride levels of mice exposed to 0 ppb, 200 ppb, and 1,000 ppb of Arsenic for 8 weeks were 138 ± 18 mg/dL, 233 ± 29 mg/dL, and 178 ± 26 mg/dL, respectively. This implies that chronic Arsenic exposure will increase triglyceride levels in the body.¹⁴

Triglycerides or triacylglycerols are formed from three fatty acids and mono glycerol. Triglycerides function as energy substances. As cells need energy, the lipase enzyme in fat cells breaks down triglycerides into glycerol and fatty acids to be released into the blood vessels.¹⁵ Triglyceride synthesis is influenced by Growth Hormone (GH). GH increases the activity of Hormone Sensitive Lipase (HSL) which can break down triglycerides into free fatty acids in fat tissue.¹⁵ Triglyceride synthesis occurs when the energy source from carbohydrates is sufficient, and then the fatty acids will experience esterification, forming esters with glycerol and triglycerides as long-term energy

reserves. Once there is no energy reserve from carbohydrates, the fatty acids and triglyceride reserves in the tissues will be broken down through the lipolysis process. Triglyceride levels in the body are influenced by several factors including genes, age, gender, food intake, obesity, physical activity, and smoking habits. The accumulation of excess triglyceride levels in the body will increase the risk of dyslipidemia. Dyslipidemia is a condition of an abnormal lipid profile in the body that increases levels of total cholesterol, triglycerides, and LDL and decreases HDL levels in the blood.¹⁶

Milk consists of two protein contents, 80% casein and 20% whey. When casein is separated from whole milk, the whey will remain and contain 68% protein.⁵ Whey protein is a liquid byproduct of cheese making. Whey protein consists of several biological components, including BCAA which are easily absorbed and used by the body, lactoferrin as a non-enzymatic antioxidant, immunoglobulin as an antibody, β-Lactoglobulin as BCAA that plays a role in retinol-binding and can modulate the lymphatic response in humans, α-Lactalbumin which can increase antibody response to systematic antigen stimulation and suppress T cell response in lymphocytes, lactoperoxidase which can remove harmful bacteria for the human body, glycomacropptide, and serum albumin.¹⁷

The high protein and amino acid content in whey protein make it an alternative food source of protein and amino acids with many health benefits. Increased protein intake can lead to changes in circulating lipids and lipoprotein levels, such as decreased LDL, decreased serum triglycerides, and increased HDL concentrations.¹⁸ Research on female Wistar rats showed that whey protein concentrate supplementation could reduce cholesterol levels and serum triglyceride concentrations after consumption for 8 weeks compared to casein supplementation.¹⁹ The decreased lipid profile levels with whey protein consumption occur through the role of BCAA in whey which accelerates digestion and absorption rates in intestines. β-Lactoglobulin content helps the absorption of cholesterol in the intestine, inhibits the expression of genes in charge of the absorption and synthesis of intestinal fatty acids and cholesterol, and increases the excretion of fecal steroids.²⁰

Global competition demands a productive workforce for companies. A productive workforce will be created through good nutritional and health status. Some epidemiological studies have shown that nutrient intake is positively associated with labor productivity.¹ The work environment condition is one factor that influences the health and nutritional status of the workforce. A work environment with exposure

to hazardous chemicals such as a mining environment can affect the health status of workers. Coal, one of the largest natural resources in Indonesia, creates many job opportunities in the coal mining environment. Coal contains fine particles of Arsenic (As) heavy metal which is a potential source of obesogens. Obesogens are environmental xenobiotic compounds that can interfere with developmental control and normal homeostasis of adipose tissue, alter lipid storage in the body, and disrupt energy balance.^{2,3} Several studies have proven that Arsenic is a potential obesogen through mechanisms affecting white adipocyte tissue. Arsenic can influence adipocyte tissue growth, adipokine secretion, lipid metabolism, and glucose metabolism.⁴

LIMITATION

This study did not analyze the levels of Arsenic (As) in the body of the research subjects, so the exact amount of exposure that affects triglyceride levels in the body remains unknown. Further research on this topic still needs to be conducted.

CONCLUSIONS

There was a significant difference in triglyceride levels before and after the whey protein intervention in the two groups. Whey protein intervention led to an increase in triglyceride levels in the exposed group and a decrease in triglyceride levels in the unexposed group although only a slight decrease was produced with the whey protein intervention for 28 days.

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