

THESIS

**THE EFFECT OF EXERCISE INTENSITY ON SERUM TNF- α LEVELS
IN FRUCTOSE INDUCED MICE (*Mus musculus*)**



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To fulfill the requirements for the undergraduate stage of the Medical Program,
Faculty of Medicine, Universitas Airlangga

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THE EFFECT OF EXERCISE INTENSITY ON SERUM TNF- α LEVELS IN FRUCTOSE INDUCED MICE (*Mus musculus*)

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SUMMARY

Fructose is a type of carbohydrate which is a monosaccharide. Fructose can be obtained from everyday foods and beverages, such as fruits, honey, and sugary drinks that contain corn syrup. Unlike glucose, fructose does not require adenosine triphosphate hydrolysis and it doesn't depend on sodium absorption leading to increased fructose uptake by the liver. Fructose passes through glycolysis more rapidly than glucose. If there are excess calories, they will be stored in fat tissue. The increase in fat tissue mass causes an increase in pro-inflammatory cytokines, one of which is Tumor Necrosis Factor Alpha (TNF- α). High levels of TNF- α in the body is a characteristic of malignant cancers, such as breast cancer, and can lead to insulin resistance. One of the strategies used to reduce pro-inflammatory cytokines is exercise. However, the effect of exercise intensity on TNF- α levels on exposure to a high-carbohydrate diet has not been revealed. This study was conducted to explore the effect of exercise intensity on serum TNF- α levels in mice exposed to high carbohydrates in the form of fructose.

This study was a laboratory study with a randomized posttest-only control group design using 24 male mice (*Mus musculus*). Firstly, mice were measured their body weight and blood glucose levels to ensure that the mice did not suffer from diabetes mellitus. Mice were randomly divided into 4 groups, then acclimatized for 1 week. On the 9th day, mice were introduced to water but still did not swim. On the 10th and 11th days, the mice started to swim but were still not given the load. On day 12, the maximum swimming duration of the mice will be measured. The release of large bubbles indicates the maximum duration. The swimming duration given at the time of the intervention was 80% of the maximum duration of the mice. The water temperature is kept at 30-36°C. Exercise interventions were given in the form of low-intensity swimming (load of 3% body weight), moderate-intensity swimming (load of 6% body weight), and high-intensity swimming (load of 9% body weight). Each group was given 30% fructose ad libitum for 8 weeks. G1 was not given additional intervention, G2 was given low intensity swimming, G3 was given moderate-intensity swimming, and G4 was given high intensity swimming.

Swimming was done every evening at the same time with a frequency 3x/week for 8 weeks. Body weight was measured at the end of the 8th week using a digital kitchen scale. Blood was taken from the left ventricle of experimental animals as much as 1 ml. Blood sampling was performed 24 hours after the last intervention. Measurement of TNF- α levels using the ELISA kit method. Statistical analysis using SPSS software. Normality test using Shapiro-Wilk test, homogeneity test using Levene and Brown-Forsythe test, comparative test using ANOVA test and Paired Sample T-Test, and correlation test using Pearson test. All data are shown with mean $\pm$ SD. All statistical analyzes used a significant level ($P<0.05$).

The results obtained were pre-intervention body weight had a significance value of $p>0.05$, which means there was no significant difference between all groups. The blood glucose data were normally distributed, but the Levene test was not homogeneous, so the Brown-Forsythe homogeneity test was carried out with a significance value of $p>0.05$, which means there was no significant difference between groups. Then the pre- and post-intervention weights were compared, and a significance value of $p<0.05$ was obtained, which means there is a significant difference between pre and post-intervention weights. The pre and post-intervention weight gain data had a significance value of $p>0.05$, which means there was no significant difference between groups. TNF- α levels were tested statistically and obtained a significance value

of $p > 0.05$, which means there was no significant difference between groups. Pearson correlation test was carried out on data of weight gain during intervention and serum TNF- α levels; the correlation value was $0.51 < r < 0.75$ with a significance value of $p < 0.05$, which means there was a strong correlation between changes in body weight (Δ) during intervention with serum TNF- α levels.

Based on these results, it can be concluded that there is a strong correlation between changes (Δ) in body weight and serum TNF- α levels; although in this study the three exercise intensities were not sufficiently proven to reduce serum TNF- α levels. Further research taking into account the study's limitations needs to be done to see how the effect of exercise intensity on serum TNF- α levels in fructose-induced mice.

ABSTRACT

Introduction: High consumption of fructose can induce obesity easier than other glucose. Excess calories will lead to an increase in adipose tissue. In line with the increase in adipose tissue, TNF- α will also increase. TNF- α can cause insulin resistance, developing into type 2 diabetes mellitus. Exercise is a strategy for lowering TNF- α levels in the blood. This study aimed to know the effect of exercise intensity on serum TNF- α levels in fructose-induced mice. **Methods:** This study was a true experimental with randomized posttest-only control group design using 24 male mice (*Mus musculus*) that were divided into 4 groups. All groups given 30% fructose *ad libitum*. Exercise given in the form of swimming. G1 was not given swimming intervention, G2 was given low intensity swimming (loads 3% body weight), G3 was given moderate intensity swimming (loads 6% body weight), G4 was given high intensity swimming (loads 9% body weight). The dependent variable in this study was the levels of serum TNF- α . The statistical analysis used the SPSS software. **Results:** There was no significant difference in serum TNF- α levels between 4 groups ($p=0,258$). However, there was a strong correlation between changes (Δ) in body weight and serum TNF- α levels ($r=0,608$; $p=0,002$) **Conclusion:** Changes in body weight has strong correlation with serum TNF- α levels positively. Low, moderate, and high-intensity exercise do not have differences in reducing serum TNF- α levels.

Keywords: High-fructose, exercise intensity, swimming, TNF- α , mice

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TABLE OF ABBREVIATIONS

ACC	= Acetyl-CoA Carboxylase
ADP	= Adenosine Diphosphate
ApoB	= Apolipoprotein B
ATP	= Adenosine Triphosphate
CA	= Citric Acid
CPT-1	= Carnitine Palmitoyl Transferase 1
CVD	= Cardiovascular Disease
DAG	= Diacylglycerol
DHAP	= Dihydroxyacetone Phosphate
DNA	= Deoxyribonucleic Acid
DNL	= De Novo Lipogenesis
F1,6BP	= Fructose-1,6-bisphosphate
F6P	= Fructose-6-phosphate
FAS	= Fatty Acyl-CoA Synthase
FBG	= Fasting Blood Glucose
FoxO1	= Forkhead box protein 01
G1P	= Glucose-1-phosphate
G6P	= Glucose-6-phosphate
G6Pase	= Glucose-6-phosphatase
GA3P	= Glyceraldehyde-3-phosphate
GA3PDH	= Glyceraldehyde-3-phosphate Dehydrogenase
Gal1P	= Galactose-1-phosphate
GALT	= Galactose-1-phosphate uridylyltransferase
GK	= Glucokinase
GLUT	= Glucose Transporter
HCC	= Hepatocellular Carcinoma
HK	= Hexokinase
HRmax	= Heart Rate Maximum
IL	= Inter Leukin
JNK	= c-Jun N-terminal protein kinase
LCFA	= Long Chain Fatty Acids
LCN2	= Lipocalin 2
METs	= Metabolic Equivalent of Task
NAFLD	= Non Alcoholic Fatty Liver Disease
NASH	= Non Alcoholic Steatohepatitis
NF-κB	= Nuclear Factor Kappa B
NK	= Natural Killer
OAA	= Oxaloacetic Acid
PFK	= Phosphofructokinase
PGK	= Phosphoglycerate Kinase
PK	= Pyruvate Kinase
PKA	= Protein kinase A
PKCε	= Protein Kinase C Epsilon
PTP-1B	= Protein-tyrosine Phosphatase 1B
ROS	= Reactive Oxygen Species
TCA	= Tricarboxylic acid
TNF	= Tumor Necrosis Factor
TNFR1	= Tumor Necrosis Factor Alpha Receptor

TPI = Triosephosphate Isomerase
UA = Uric Acid
UDP = Uridine Diphosphate
UDPG = Uridine Diphosphate-glucose
UDPGal = Uridine Diphosphate Galactose
WAT = White Adipose Tissue
 α KG = Alpha-ketoglutarate
 α KGDH = Alpha-ketoglutarate Dehydrogenase