JSSF (10) (2) (2024)

Journal of Sport Sciences and Fitness

http://journal.unnes.ac.id/sju/index.php/jssf

The Effect of Administration Ginger and Lemongrass Drink on *Interleukine 6* Football Player After Exercise

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Article's Info

Article's History:
Received October 2024
Accepted December 2024
Published December 2024

Keywords:
Physical exercise, ginger, lemongrass, interleukine 6, football player

Abstract

Physical exercise can increase the production of free radicals. Free radical can trigger oxidative stress and inflammation. Ginger and lemongrass has bioactive compound that function as antioxidant and antiinflammatory. This study aim to determine the effecs of giving ginger and lemongras drink on IL6 level in football player. Quasi experimental study with pre and post control group design. The subject were 24 football player divided 2 namely control and intervention group. The intervention group given a ginger and lemongrass drink. The control group was only given brown sugar drink. The intervention are given after exercise for 28 days. Blood samples were collected and tested after exercise. Interleukine 6 were measured using the ELISA Method. Our result showed there was differences in interleukine 6 after giving ginger and lemongrass drink (p=0.004). The delta of interleukine 6 level after intervention was 3.24 pg/ml. Conclusion this research are there was a decrease in interleukine 6 level after giving ginger and lemongrass drink for 28 days to football players after exercise.

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ISSN 2252-6528

INTRODUCTION

Physical exercise on football can cause a physical stressor that can disrupt the balance of the body's metabolism, cause a feedback respons from the body's organ that can increase in respiratory frequency and increase hearth rate (Gabriel & Zierath, 2017). Physical exercise on football cause incerase oxygen consusmption 10-20x in the body and increase 100x in the muscle which can trigger to produce free radical or reatcive oxygen species (ROS) (Sinaga, 2016). Physical exercise can increase ROS which exceed the body's antioxidant can cause oxidative stress (Yuji & NAITO, 2002). Oxidative stress can cause cell damage and as a base of pathogenesis for chronic disease such as cardiovasvular, autoimmune, pulmonary, metabolic disorders and aging (Nurdyansyah, 2017). Free radical will cause cell damage and has a role to the causes of chronic disease, muscle damage and immune function reduced, so that it can be affected exercise performance. (Jamuna Rani & Mythili, 2014). Heavy physical exercise can cause tissue damage muscles and blood vessel endothelium in athlete, suppressing the body immune system so that it is susceptible to infection and injury on athletes (Petersen & Pedersen, 2005). Physical exercise can trigger inflammatory process in endothelial cell. Cell membrane damage due to lipid peroxidation reactions stimulates macrophages to release inflammatory mediators such as cytokin (Yuniarti, 2014). Interleukine 6 (IL6) is one of pro inflammatory cytokines, so this cytokine has the opportunity to be used indicator to assess the level of inflammation due to microtrauma that occurs in muscles during physical exercise. Interleukine 6 is one of the cytokines produced during the early stages of the inflammatory process, which is also triggered by physical exercise (Nielsen et al., 2016). The research reported that a significant increase in IL6 levels before and after the race in 15 male marathon runners (Bernecker et al., 2013). The research on 22 half marathon runners and 18 full marathon runners showed there was a significant increase in IL6 levels immediately after training (Reihmane et al., 2013). The reseacrh showed there was an increase in IL6 level ($p<0,001$) in west sumatra students sport education and training center football athlete after submaximal training (Yuniarti, 2014).

Prevention of oxidative stress and inflammation after axercise aims to prevent infection, injury and decreased performance on athlete. This prevention can consume high antioxidant food or drink. Ginger and lemongrass contain bioactive compound that function as antioxidant and antiinflammatory (Ballester et al., 2022). Ginger contains secondary metabolite including fenolic compound such as gingerol, shogaol and paradol which function as antioxidant (Mao et al., 2019).The Potential mechanism of antioxidant in ginger is 6 shogaol activates the nuclear factor erythroid 2–related factor 2 (NrF2) signaling pathway and increase the expression of the NrF2 gene target (endogenous antioxidant gene) (Nile & Park, 2015). Gingerol and shogaol on ginger can inhibit cyclooxygenase (COX2) and Lipoxygenase (LOX) enzyme and prevent the metabolism of arachidonic acid so can inhibiting the biosynthesis of prostaglandin E2 (PGE2) and leukotrienes which are inflammatory mediators (Mao et al., 2019). Lemongrass has secondary metabolite compounds such as saponins, tannins, alkaloids, steroids, phenols and

flavonoids which have potential antioxidant and antiinflammatory (Sugiarti et al., 2023). The citral in lemongrass can inhibit activation of pro inflammatory mediators such as cyclooxygenase, lipooxygenase and inducible nitric oxyde synthase (iNOS). The flavonoid in lemongrass can shorten the inflammatory phase with eliminating ROS, detoxifying hydrogen peroxide so reducing lipid peroxide level (Katsukawa et al., 2010). A study showed that giving powdered ginger in capsule 3x500mg (1.5g/day) for 6 weeks on endurance athletes could reduce IL6 level after exercise (Zehsaz et al., 2014). The research reported giving ginger drink 330mlx5 at different administration times could reduce Delayed Onset Muscle soreness (DOMS) after anaerobic exercise for 2 weeks (Ricky Kurniawan et al., 2020). The research on male adolescent athletes showed that supplementation with ginger powder and kesumba a dose 2g/day for 4 week could increased muscle strenght and relieve delayed onset muscle soreness (DOMS) (Arikunto, 2019). This research aim to determine effect of giving ginger and lemongrass drink to reduce IL6 on football player after exercise.

METHOD

Research Design

This research is quasy experimental with a pre and post control group design. It was conducted from May to June 2024 at adolescent football athlete PS Undip Semarang. The subject were 24 adolescent football athletes divided 2 namely control and intervention group. The intervention group given a ginger and lemongrass drink (1g powder ginger, 2g powder lemongrass) with 20g brown sugar and brewed with 150ml warm water 60℃. The control group was only given brown sugar drink with formula 20g brown sugar and brewed with 150ml warm water 60℃. The intervention are given after exercise for 28 days (Arikunto, 2019). Physical exercise include running arround the football 5x, shuttle run with a distance 10m for 2 minutes, sit up 30x, back up 30x, push up 30x, squat thrust jump 30x. Blood samples were taken twice namely before intervention and after intervention ginger and lemongrass drink. Blood samples were taken immediately after exercise by professional laboratory. Interleukine 6 were measured using the Enzyme Linked Immunosorbent Assay (ELISA) Method. Ethics approval has been obtained from the Health Research Ethics Commission of the Nursing and Health Faculty, University of Muhammadiyah Semarang number 168/KE/03/2024.

Reseach Subject

The technique sampling was purposive sampling. Determining total sample used slovin formula with an error 10% and obtained total samples 24 athletes (Arikunto, 2019). Determination of samples group was carried out using the simple random sampling method. The inclusion criteria in this study were samples is registered as a football athlete at PS Undip, age 15-18 years, willing to sign informed consent, Body Mass Index (BMI) 18.5-25.0 bw/h2, no consume supplement during the research, not injured, not smooking, not consuming alcohol, do not take anti-inflammatory drugs. Exclusion criteria were samples withdraw from research, illness or injury, subjects consume supplement, took anti-inflammatory drugs, smoked and drank alcohol.

Athlete's Measurement

Athlete data collection include name, age, history of illness, history of smooking, history of

alcohol consumption, history of drug consumption.

Athlete's Antropometric

Anthropometric measurement include body weight, height, assessment nutritional status using Body Mass Index (BMI). Body weight and Body fat percentage are collected were using an Omron HBF 375 Bioelectrical Impedance analysis (BIA) with an accuracy 0.1 kg. Height data was measured using stadiometer SECA brand with an accuracy 0.1 cm.

Athlete's sleep quality

Athlete sleep quality was taken using the Pittsburg Sleep Quality Index (PSQI) questionnaire. The PSQI questionnaire is a screening to see sleep quality over the past month. There are 7 components score for assessing sleep quality. Total component scores < 5 is good, and if > 5 is bad (Khasanah & Hidayati, 2012).

Athlete's physical activity

Athlete's physical activity was taken using the international physical activity questionnairy (IPAQ). IPAQ determines Physical activity scores with the following formula = Mets-min/week = mets level (type of activity) x number of minutes of activity x number of day/week (Kyu et al., 2016).

Athlete's food Intake

Athlete's food intake was taken using 2x24 hour recall method. The nutrition values analyzed are vitamin A, vitamin C and vitamin E. Intake data was collected using recall 24 hour.

questionnaire with food model and porsimetri. The result dietary recall are processed using the nutrisurvey program and compared with nutritional adequay rate.

Interleukine 6 test

Interleukine 6 test wa taken using the Enzyme Linked Immunosorbent Assay (ELISA) method used elisa reader with wavelength 450nm. The reagent used are elikine human IL6 elisa kit. Interleukine 6 test was carried out in the Chemistry laboratory, Nutrition Department, Diponegoro University by professional laboratory. The normal value for interleukine 6 levels is less than 4 pg/ml.

Data Analysis

The statistical analysis using SPSS version 23.0 for windows and the normality of the data was analyzed using Shapiro wilk. Univariate Analysis was used to describe the characteristics respondent such as weigh, heigh, age, body mass index (BMI), body fat percentage, physical activity, sleep quality, food intake and interlukine 6 level. Paired t test or Wilcoxon used to determine the difference of IL6 level before and after intervention. Independent t test or Mann whitney used to determine difference of IL6 in each control and intervention group. The linear regression test used a linear regression to determine the influences confounding variables on IL6 level.

RESULT AND DISCUSSION

Table 1 presents the characteristics of aged, body weight, height, body mass index and body fat percentage respondent.

Table 1. Subject Characteristic

Characteristics	Control group (n = 12)		Intervention group (n=12)		*P value*
	Mean +SD	min – max	Mean±SD	min-max	
Age	16.42 ± 0.515	16 – 17	16.58 ± 0.515	16 - 17	0.514**
Body weight	61.97 ± 3.64	57.20 – 68.0	60.64 ± 7.02	51.20 – 76.0	0.569*
Height	169.27 ± 4.54	161.50 – 176	168.36 ± 5.45	161 – 177.20	0.662*
BMI	21.67 ± 1.71	19.49 – 24.73	21.35 ± 1.68	19.05 – 24.20	0.651*
Body Fat Percentage (%)	12.35 ± 3.29	7.30 – 17.40	13.20 ± 4.03	8.90 – 19.50	0.799**

**independent t test* ** *Mann Whitney test*

Based on table 1, the result Body Mass Index (BMI) and body fat percentage of respondents are normal category (21.67 kg/m2 and 21.35 kg/m2) and (12.35% and 13.20%), this is accordance with the inclusion criteria. Independent t test and mann whitney showed that age, weight, height, BMI and body fat percentages have p value > 0.05, it means no difference between control and intervention group so the data is homogen.

Table 2 presents the characteristics of physical activity, sleep quality and food intake of Vitamin A, Vitamin C and Vitamin E respondents.

Table 2 Characteristic of physical activity, sleep quality and food intake respondents.

Characteristic	**Control group**			**Intervention group**			***p value***
	n	**%**	**Average ± SD**	**n**	**%**	**Average ± SD**	
Physical activity							
Before Intervention							
Low active	1	8.3	5139.96 ±	1	8.3	4580.63 ±	0.915*
Moderate active	11	91.7	1349.21	10	83.3	446.14	
High active	0	0		1	8.3		
After Intervention							
Low active	1	8.3	5691.42 ±	0	0	4643.13 ±	0.033**
Moderate active	10	83.3	1776.48	10	83.3	1523.94	
High active	1	8.3		2	16.7		
Sleep Quality							
Before Intervention							
Good	7	58.3	5.0 ± 1.54	7	58.3	4.67 ± 1.72	0.630**
Bad	5	41.7		5	41.7		
After Intervention							
Good	8	66.7	5.0 ± 2.0	6	50.0	5.33 ± 2.54	0.724*
Bad	4	33.3		6	50.0		
Food Intake							
Before Intervention							
Vitamin A (RE)							
Adequate	6	50	881.95 ± 338.8	3	25	709.63 ±	0.160**
Inadequate	6	50		9	75	276.74	
Vitamin C (mg)							
Adequate	0	0	16.33 ± 12,9	0	0	26.83 ± 9,84	0.114**
Inadequate	12	100		12	100		
Vitamin E (mg)							
Adequate	0	0	3.5 ± 1.8	0	0	3.3 ± 0.84	0.731*
Inadequate	12	100		12	100		
After intervention							
Vitamin A (RE)							
Adequate	5	41.7	693.94 ±	5	41.7	683.26 ±344.90	0.945*
Inadequate	7	58.3	402.36	7	58.3		
Vitamin C (mg)							
Adequate	0	0	13.00 ± 20.75	0	0	6.01 ± 5.18	0.551**
Inadequate	12	100		12	100		
Vitamin E (mg)							
Adequate	0	0	2.32 ± 2.05	0	0	2.06 ± 1.1	0.704*
Inadequate	12	100		12	100		

**independent t test **mann whitney test*

Based on table 2, the physical activity respondents are mostly *moderate active* between control and intervention group. *Independent t test* showed there was no difference in the physical activity score before intervention (p=0.915). *Mann whitney* test showed there was difference the physical activity score between control and intervention group after giving ginger and lemongrass drink (p=0.003).

The sleep quality of respondents are mostly good category in control and intervention group. The independent t test and Mann whitney test showed there was no difference in sleep quality score between the control and intervention group either before intervention (p=0.630) and after intervention (p=0.724).

Dietary intake of vitamin A, vitamin C and Vitamin E respondens are mostly inadequate either control and intervention groups. The average intake of vitamin A in the control and intervention groups was <900 RE/day. The average intake of vitamin C and E in the category is less <90mg/day for vitamin C and <15mg/day for vitamin E. The result of recall intake 2 x 24 hours showed consumption fruits and vegetables respondents is inadequate so the adequacy of vitamin A, C and E is not met.

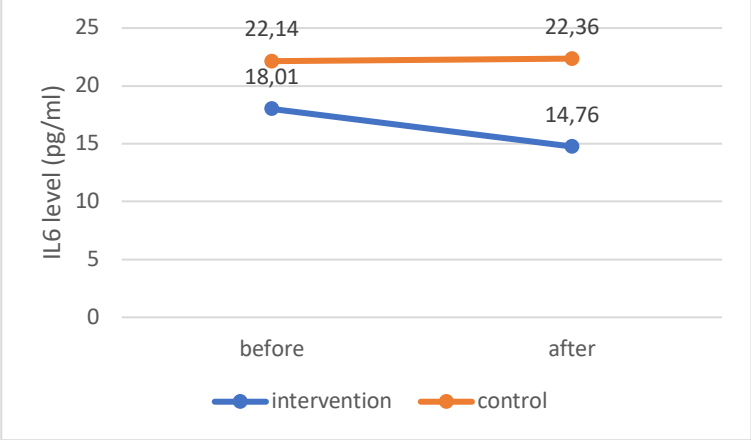
Table 3 present Interleukine 6 level respondents before and after intervention ginger and lemongrass drink :

Table 3. *Interleukine 6* (*IL6*) level before and after intervention

***IL6* Level (pg/ml)**	**Control group**		**Intervention group**		***p value***
	Average ± SD	**Min – Max**	**Average ± SD**	**Min – Max**	
Before	22.14 ± 7.93	14.96 – 43.87	18.01 ± 5.09	11.33 – 29.87	0.89[a]**
After	22.36 ± 8.38	11.14 – 44.42	14.76 ± 3.30	9.69 – 21.60	0.02[a]**
Δ IL6	0.22 ± 1.68	-3.82 – 2.27	-3.24 ± 3.05	-9.82 – 1.91	0.02[a]**
P value	0.347[b]**		0.004*		

**paired t test* ** [a]*Mann whitney* dan [b]*Wilcoxon*

Based on table 3 showed there was difference in IL6 level after giving ginger and lemongrass drink between control with intervention groups (p=0.02). There was difference in IL6 level on intervention group after giving ginger and lemongrass drink (p=0.004). There was a decrease in IL6 level in the intervention groups from 18.01 pg/ml to 14.76 pg/ml with the delta 3.24 pg/ml.



Picture 1. *IL6* level before and intervention ginger and lemongrass drink

Physical exercise on football can increase the production of reactive oxygen species (ROS) or free radical and cause oxidative stress. Oxidative stress can cause cell damage, trigerring an inflammatory response that activates inflammation cells (Sinaga, 2016). Physical exercise can increase inflammation within muscle cells. Intramuscular inflammation is a coordinated process that leads to cellular adaptation process such as skeletal muscle hypertrophy. Microtrauma of the muscle is one of the triggers for an inflammatory response. This damage can stimulate the release of proinflammatory mediators such as interleukine 6 (Peake et al., 2017). Ginger contains phenolic compounds mainly gingerol, shogaol and paradol which are responsible for various bioactives of ginger (SARI & NASUHA, 2021). 6-gingerol of ginger can inhibit the release of cyclooxygenase (COX2) enzyme. Cyclooxygenase is the enzyme responsible for the formation of prostaglandins. Prostaglandins are the main mediator of inflammation. Inhibition of COX2 can suppresse the occurrence of increase inflammation (Riduan, 2015). Cyclooxygenase is also known as prostaglandine 2 (PG) synthase which has a role in the process of synthesizing the enzyme prostaglandine E2 (PGE2) from arachidonic acid. Cyclooxygenase is activated during inflammation and is easily activated by several activators namely proinflammatory cytokines, growth factor, tumor promotors and oxidative stress (Bare et al., 2019). 6 gingerol in ginger can also inhibit the nuclear factor kappa betha (NFkβ) pathway, which is one of the component of inflammation. Nuclear factor kappa betha is a transcription factor in the cytoplasm of every cell and translocated to the nucleus when activated. Nuclear factor kappa betha controls the release of a number important gene s in immune and inflammatory process including Granulocyte–Macrophage Colony Stimulating Factor (GM-CSF), IL6, IL2 dan TNF α. Oxidative stress activates NFkB which in turn encodes genes transcription such as genes encoding cytokins, chemokins and COX2 enzyme stimulating the production of proinflammatory melecules that trigger inflammation and oxidative stress (Riduan, 2015). The reseach showed ginger powder supplementation at a dose 1.5g/day for 6 weeks can reduce inflammatory cytokines (TNF α, IL 1β dan IL-6) in endurance running athletes (Zehsaz et al., 2014). The another research showed there was a decrease in IL6 level in female taekwondo athletes after being given pure ginger with dose 2g/day for 24 days (Karizak et al,2024). Lemongrass has secondary metabolite compounds such as saponins, tannins, alkaloids, steroids, phenol and flavonoid that have potential as antioxidant and anti-inflammatory (Sugiarti et al., 2023). Lemongrass also has content active compounds citronellal and geraniol which are antioxidant. The citronellal of lemongrass content 32-45% and geraniol 12-18% (Febrina, 2019). A study showed antioxidant activity test of lemongrass using DPPH method has IC50 value 50.68 ppm, which means that lemongrass plants are strong antioxidant (Febrina, 2019). An another research, antioxidant activity test on lemongrass leaf extract showed an IC50 value of 64.17 ppm which means it has strong antioxidant activity (Fitria et al., 2022). The phenolic content in lemongrass can inhibit the nuclear factor kappa betha (NFkβ) pathway and cytokine expression. Lemongrass extract can reduce the

concentration of ROS, lipid peroxidation, DPPH and support the endogenus antioxidant defense system in alveolar macrophage cells through increase superoxidase dismutase (SOD) activity and gluthatione peroxidase (GPX) formation (Mukarram et al., 2022). Measurement with DPPH radicals, lemongrass compound can clean superoxide anion, inhibit xantin oxidase enzyme and lipid peroxidation in human erythrocytes. Citral on lemongrass can inhibit inflammatory mediator, suppress TNF α, inhibit inducible nitric oxide synthase (iNOS), Nitric Oxide (NO) production, NFkβ pathway and suppresses the COX2 enzyme (Said et al., 2019). The anti-inflammatory effect of lemongrass is the content of phenolic acid, flavonoids and tannis. An invitro study, lipopolysaccharide (LPS) induced RAW 264.7 macrophage cells with skin derived dendritic cell line (FSDC), lemongrass extract can suppress the expression of the NO, PGE2 and iNOS. Flavonoid and tannin fraction in lemongrass showed efficacy as anti-inflammatory. Phenolic acid effectively reduces prostaglandine E2 production in LPS induced 264.7 macrophage (Figueirinha et al., 2008).

CONCLUSSION

The result of this study showed there was a decrease on interleukine 6 level in football player after being given ginger and lemongrass drink (p=0.004) for 28 day with the delta 3.24 pg/ml.

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