**Effect of 8 Weeks of High-intensity Interval Training versus Traditional Endurance Training on the Blood Lipid Profile in Humans**

by

David James Elmer

A dissertation submitted to the Graduate Faculty of
Auburn University
in partial fulfillment of the
requirements for the Degree of
Doctor of Philosophy

Auburn, Alabama
August 3, 2013

Keywords: Dyslipidemia, HIIT, Inflammation, HDL function



Approved by

David Pascoe, Chair, Professor of Kinesiology
Kimberly Garza, Assistant Professor of Pharmacy Care Systems
Bruce Gladden, Professor of Kinesiology
John Quindry, Associate Professor of Kinesiology

ABSTRACT

INTRODUCTION: High-density lipoprotein (HDL) plays an important role in the prevention of atherosclerosis and coronary heart disease (CHD), which is the leading cause of death in the United States. While the importance of adequate concentrations of HDL is recognized, HDL must exhibit anti-inflammatory properties and participate in reverse cholesterol transport to be beneficial. Chronic C-reactive protein (CRP) levels may indicate whether HDL can function in this manner, with reductions in CRP being associated with greater HDL function and thus protection from CHD. Traditional endurance training (ET) is an effective way to reduce chronic CRP concentration, increase HDL function, and increase HDL concentration. However, High-intensity Interval Training (HIIT) is equal or superior to ET for improving measures of cardiovascular health across a wide range of populations, and may be more beneficial than ET for improving inflammatory status, HDL function, and HDL concentration. PURPOSE: To compare the effects of duration- and work-matched ET and HIIT on HDL function and concentration, as well as on CRP, the acute IL-6 response to exercise, and cardiovascular fitness. METHODS: Twelve young males (age 21.6 $\pm$ 1.6 years, HDL 34 $\pm$ 8 mg/dL, $VO_2$max 41.6 $\pm$ 5.4 ml/kg/min) divided into 2 matched groups (HIIT or ET) based on HDL concentration and $VO_2$max completed 8 weeks of duration- and work-matched exercise training 3 days per week. Each exercise session lasted 30 min, with a progression in average intensity from 70 to 80% of $VO_2$max. HDL function and concentration, resting CRP, acute IL-6 response to exercise, $VO_2$max, body composition, and blood pressure were determined before and after the 8 weeks of training. RESULTS: After 8 weeks

of training, there were no significant differences in HDL function, resting CRP, HDL concentration, or $VO_2$max within or between groups ($p>0.05$). HIIT lowered plasma triglyceride (TRG) concentrations (-31 ± 28 mg/dL, $p=0.04$) significantly more than ET ($p=0.009$). HIIT also significantly increased treadmill Vmax (0.6 ± 0.5 mph, $p=0.02$) and reduced HRmax (-5 ± 3 bpm, $p=0.01$) from baseline levels, but there was no significant difference between the groups. ET resulted in significant reductions in the percentage of android fat (-2.60 ± 2.41%, $p=0.045$) and TC:HDL ratio (-0.60 ± 0.41, $p=0.02$), but neither were significantly different from HIIT. CONCLUSION: When average intensity, workload, and duration are equal, these results indicate no difference between HIIT and ET for improving HDL function, HDL concentrations, or resting CRP. However, in this time period HIIT was significantly better than ET for reducing plasma TRG concentrations. Also, HIIT alone improved Vmax and reduced HRmax, while only ET was beneficial for reducing the percentage of android fat. A larger sample size, longer training period, or different exercise protocols may be necessary to alter HDL function and chronic inflammation.

ACKNOWLEDGMENTS

I would like to thank Dr. David Pascoe, for giving me the opportunity to do what I love, and for his mentorship and advice over the last four years. I would like to thank the members of my committee, Dr. Bruce Gladden, Dr. John Quindry, and Dr. Kimberly Garza, for their wisdom and critical eyes, and for their patience with a rushed, disorganized grad student. I would also like to thank the Department of Kinesiology for its support and for providing me with the experience needed for my future career. I want to thank Matt Barberio for showing me how to navigate every aspect of graduate school, Rich Laird for suffering with me through this process, Graham McGinnis for being a great workout partner and for being patient with me as I worked in his space, and Khalil Lee for his gentle spirit and willingness to help in any way he can. I would like to thank my parents, Ken and Julie Elmer, for their love, the fantastic example they set for me, and for the gentle pushes to always seek to do my best. I would like to thank my in-laws, Charlie and Leslie Barron, for adopting me into their family, loving me, and treating me as one of their own. I especially want to thank my wife, Laura, for her constant love, support, excitement, and encouragement. Without you I would have been a lonely, depressed wreck during this process. I love you now and always. And finally, I must give all praise and honor to God, my heavenly Father, for infinite love, blessings, and purpose in this life, the Lord Jesus Christ for salvation and a promise of the life to come, and his Holy Spirit, for encouragement, comfort, and peace.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF ABBREVIATIONS

ABCA-1..... ATP-binding cassette, sub-family A, member 1

ApoA-1.....Apolipoprotein A-1

a-v$O_2$d.....Difference between arterial and mixed venous oxygen content

CAD..... Coronary artery disease

CETP.....Cholesterol ester transfer protein

CHD.....Coronary heart disease

CRP.....C-reactive protein

CVD.....Cardiovascular disease

EDTA.....Ethylenediaminetetraacetic acid

EL..... Endothelial lipase

ESR.....Erythrocyte sedimentation rate

ET..... Endurance training

FU/min.....Fluorescence units per minute

Glu.....Glucose

GSH.....Glutathione

HDL.....High-density lipoprotein

HIIT..... High-intensity interval training

HRmax..... Maximum heart rate

Kcal..... Kilocalorie

LDL……Low-density lipoprotein

LCAT……Lecithin-cholesterol acyl-transferase

LPL……Lipoprotein lipase

LT……Lactate threshold

NADPH……Nicotinamide adenine dinucleotide phosphate

Ox-HDL……Oxidized high-density lipoprotein

Ox-LDL……Oxidized low-density lipoprotein

PON-1……Paraoxonase-1

RER……Respiratory exchange ratio

SAA……Serum amyloid A

TC……Total cholesterol

TRG……Triglyceride

Vmax…… Running velocity at maximum volume of oxygen consumption

$VO_2$max…… Maximum volume of oxygen consumed in one minute

# CHAPTER I

# INTRODUCTION

Dyslipidemia and its contribution to cardiovascular disease is a problem that plagues the Western world. According to the CDC, heart disease is still the leading cause of death in the United States, claiming nearly 600,000 people per year. Of all the factors that increase the risk of developing coronary heart disease (CHD), research has shown that low concentrations of high-density lipoprotein (HDL) are the best lipid predictor of future CHD events (e.g. myocardial infarction).[1, 2] Raising these HDL levels drastically reduces the risk of CHD, with research suggesting a 3% reduction in risk for every 1% increase in HDL.[1] Research also indicates that in healthy individuals, as well as people with a documented history of CHD, cardiovascular fitness ($VO_2$max) is a powerful predictor of mortality,[3-5] with an estimated 44% reduction in all-cause mortality risk in men who went from unfit to fit,[6] and 15% decrease in risk for every 1 ml/kg/min increase in $VO_2$max.[7]

*HDL function*

High-density lipoprotein is partially able to reduce the risk of CHD by exerting an anti-atherogenic and cardioprotective effect through a process called reverse cholesterol transport, which removes cholesterol stored within the arterial wall. High levels of circulating low-density lipoprotein (LDL) are directly related to the onset of CHD and atherosclerosis, with any increase above 100 mg/dl seeming to be highly atherogenic.[8, 9] LDL cholesterol molecules can be oxidized (Ox-LDL) by the major cells of the arterial wall, with oxidation by macrophages predominating in

the early stages of atherogenesis.[10] LDL oxidation depends on the oxidative susceptibility of LDL, which can be increased by decreasing the size of the LDL molecule and exposing the LDL to reactive oxygen species (ROS).[10, 11] Macrophages can also become more pro-oxidant in the presence of oxidative stress, resulting in greater NADPH oxidase activity, which increases ROS production and promotes reductions in the antioxidant glutathione (GSH), which is an indicator of inflammation.[10] Ox-LDL is bound by a scavenger receptor, and LDL and macrophages begin to accumulate in the extracellular space of cells in the arterial wall. This accumulation of cholesterol and macrophages leads to atherosclerosis, in which the LDL cholesterol and macrophages form foam cells, which can progress to a fatty streak, and potentially lead to thrombosis and a CHD event.

HDL is able to reverse this process by removing cholesterol from the arterial wall and depositing it in the liver, where it will be secreted as bile. As shown in figure 1, apoA-1 (the primary subunit of HDL) and pre-β-HDL act by receiving cellular cholesterol from ATP-binding cassette, sub-family A, member 1 (ABCA-1), which translocates cellular cholesterol from within the arterial wall. ABCA-1 activity is important for the lipidation and subsequent maturation of newly formed apoA-1 and pre-β-HDL into mature HDL, which prevents their degradation by the liver, kidney, or steroidogenic tissue.[12] Otherwise, lipid-poor apoA-1 and pre-β-HDL would be rapidly catabolized, reducing HDL concentrations.[13] ApoA-1 and pre-β-HDL mature into HDL as they continue to gather and esterize cholesterol from the arterial cell membrane. Lecithin-cholesterol acyl-transferase (LCAT) aids in the esterification of the cholesterol molecules and moves the cholesterol esters to the core of the HDL molecule, allowing it to accept more cholesterol from the same or other locations.[12, 14] Cholesterol ester transfer protein (CETP) removes the cholesterol esters from HDL in the plasma, making it susceptible to clearance by the

liver, which disposes of the cholesterol through the secretion of bile in the intestines.[14] After CETP-mediated uptake, the resulting smaller HDL particles bind to scavenger receptors on the liver, stimulating the uptake of any remaining cholesterol esters on the HDL particle by the liver.[14] This process of removing cholesterol from the arterial walls and disposing of it in the liver is why HDL levels are associated with a reduced risk of developing cardiovascular disease or having a future CHD event.

Since HDL must be present in order to undertake the process of reverse cholesterol transport, the concentration of HDL in the blood is often used for CHD risk assessment. However, it is not just the number of HDL particles that is of importance for protecting against future CHD events; the functionality of the HDL is vital for exerting an anti-atherogenic effect in the body. Research suggests that the overall functioning of the HDL is potentially even more important than the total amount of HDL present.[15] This can be demonstrated in the fact that CHD events can occur even when the concentration of HDL particles is equal to or greater than the recommended level ($\geq$ 40 mg/dl). In the Framingham study, 44% of the men and 43% of the women who had a CHD event had appropriate HDL levels.[16] This finding may be due to the fact that HDL can function in a pro- or anti-oxidant manner, and there is no relationship between HDL concentration and whether it is pro- or anti-inflammatory.[15] Not only is pro-inflammatory HDL incapable of reverse cholesterol transport, but it actually aids LDL in binding to macrophages.[17] This effect comes from a secondary mechanism by which HDL normally protects against CHD. Anti-inflammatory HDL protects against atherosclerosis not only through reverse cholesterol transport, but also by preventing the oxidation of LDL by ROS. Functional HDL inhibits this first step in foam cell formation, reducing the amount of Ox-LDL present and foam cell formation. However, when HDL is behaving in a pro-oxidant manner, it increases the amount of inflammation, increasing Ox-LDL and further contributing to the risk of CHD.[18]

*Aerobic fitness*

One of the many benefits of regular exercise is a reduction in chronic inflammation.[19-21] This is potentially the mechanism by which exercise may improve HDL function along with HDL concentrations.[22, 23] The result of this improved HDL function due to exercise is an increase in resistance to LDL oxidation in the presence of oxidative factors.[24] However, increases in HDL function are not dependent on increases in concentration, as HDL function can increase with simultaneous decreases in concentration.[25] The relationship between physical activity, inflammation, and HDL function at least partially explains why physical activity is negatively associated with the risk of CHD, with greater increases in physical activity dramatically reducing the incidence of CHD events.[22] Exercise is so beneficial for reducing chronic inflammation that at least one study found that the reduction in inflammation seen was great enough to place the participants into different CHD risk categories at the conclusion of the study.[26] These protective effects of exercise result in a well-documented and strong associations between physical fitness, as determined by maximal oxygen consumption ($VO_2$max) and all-cause mortality.[3, 4, 7]

The potential mechanism by which chronic inflammation is reduced is repeated exposure to acute, exercise-induced inflammation, which results in increased anti-oxidant function.[27-29] The acute inflammatory response is dependent on the intensity of the exercise, with increased intensity dramatically increasing the response.[30, 31] Thus, exercise strategies that focus on high intensity may be more beneficial for improving HDL function since they potentially stimulate a greater reduction in chronic inflammation through greater acute inflammatory responses.[28, 29] High-intensity interval training (HIIT) is a safe and effective way to incorporate high intensity exercise, regardless of age, sex, or current health status.[32-43] HIIT may be more effective for improving aerobic fitness, and therefore health, than traditional endurance training (ET), with equal or greater improvements seen in $VO_2$max,[32] cardiac and endothelial function,[34, 41, 44] reduced chronic inflammation,[36] as well

as reductions in other risk factors associated with CHD such as ROS and carotid-femoral pulse wave velocity.[45]

*Conclusion and Purpose*

While research is emerging about how enhancing HDL function could reduce the risk of a future CHD event, to my knowledge there is no research examining the effect of different exercise training styles (e.g. traditional endurance training vs. high-intensity interval training) on HDL function, which could shed light on the best exercise strategy for reducing CHD risk. Therefore, the purpose of this research endeavor is: 1) to determine which exercise strategy is the most effective to improve HDL anti-oxidant function, 2) to determine which exercise strategy results in the greatest improvement in the concentration of HDL, 3) to evaluate the changes in chronic inflammation and analyze a potential relationship with the acute inflammatory response and the lipid profile, and 4) to examine the potential relationship of $VO_2$max and HDL function. This research could serve to demonstrate which type of aerobic exercise is the most effective way to improve the protective effects of HDL and prevent future CHD events.

# CHAPTER II

# REVIEW OF LITERATURE

*Inflammation, HDL concentration, and HDL function*

In 2012, Honda *et al.* published a study comparing the impact of oxidized HDL (Ox-HDL) and interleukin-6 (IL-6) on mortality and cardiovascular disease (CVD) in patients on hemodialysis.[46] While IL-6, which is a marker of inflammation, is a strong indicator of CVD risk,[47] Honda *et al.* found that greater concentrations of Ox-HDL, especially when paired with elevated IL-6, were associated with an increased risk of CVD.[46] It is also worth noting that in this study, patients with greater Ox-HDL and increased risk of CVD showed increased concentrations of HDL, which is normally associated with a reduced risk of CVD. The mechanism explaining this change in patients at risk for CVD could be related to the immune response to acute endotoxemic challenges. In the acute phase response to endotoxin release within the body, there is a change in HDL function from anti-inflammatory to pro-inflammatory.[48] This was illustrated by Van Lenten *et al.* in a research study examining the acute phase response in rats.[48] They reported that HDL no longer protects LDL from oxidation during the acute phase response to an endotoxemic challenge, but in fact enhances the oxidation of LDL. The change in function is the result of a remodeling of HDL, replacing the anti-inflammatory subunit apolipoprotein A-1 (apoA-1) with serum amyloid A (SAA), and removing enzymes, e.g. paraoxonase (PON1), which protect LDL from oxidative stress.[48, 49] Without this protection, LDL is easily oxidized and accumulates within endothelial cells. Not only do endotoxins reduce the activity of PON1, but the cytokines that mediate the acute phase response to the presence of endotoxins can directly reduce PON1 activity as well, making the

LDL more susceptible to oxidation.[49] Lindhorst *et al.* also induced inflammation in mice to study the acute phase response to an inflammatory challenge.[50] The inflammation caused HDL to lose apoA-1 subunits, which were replaced by SAA particles. The level of addition of SAA mirrored the inflammatory response, with a peak 24 hours after an inflammatory challenge. However, after 48 hours the apoA-1 subunits began to be added back to the HDL particle, restoring its anti-inflammatory and anti-oxidant qualities.

This response to an inflammatory challenge is likely beneficial in the short term, as cholesterol is involved in the heat shock protein response to a stressor, serving as a mediator in the signaling transduction pathway.[51] However, the altered role of HDL is likely atherogenic if maintained over a period of time. In fact, these pro-inflammatory HDL molecules, with reduced apoA-1 levels and PON1 activity, are found chronically in people with a history of coronary heart disease, even when no detectable endotoxins are present and their HDL concentrations are the same as healthy controls.[16] Ansell *et al.* gathered participants with a history of CHD and examined their HDL concentrations and determined the HDL anti- or pro-inflammatory qualities.[16] They discovered that in these patients with a history of CHD, the HDL functioned in a pro-inflammatory manner, even in a group of participants with HDL concentrations well above the NCEP recommendation of 40 mg/dl.[9] In fact, there was an entire group of participants with HDL concentrations greater than 84 mg/dl but with pro-inflammatory HDL function that suggested they were at increased risk of CHD.[16] The researchers found the relationship between the anti- or pro-inflammatory qualities of HDL and CHD to be stronger than the relationship between HDL concentration and CHD, as listed the accepted NCEP guidelines for healthy HDL levels and CHD risk.[9, 16]

Patients with CHD have been described as in a persistent "acute phase response," with continuously elevated markers of inflammation, as often indicated by increased C-reactive protein

(CRP) in the absence of any infection or acute stress.[16, 52] Ridker *et al.* found that in a relatively homogenous population, patients with higher baseline levels of CRP, SAA, and IL-6 at rest were significantly more likely to have a cardiovascular event compared to others in the sample group, with CRP concentrations being the single best predictor of such an event.[2] Elevations of SAA result in a remodeling of HDL and a change in function, and the elevation of SAA and CRP is found during the acute phase response to an endotoxin, resulting in the removal and elimination of apoA-1 subunits.[53] Ridker *et al.* also found the concentrations of CRP to be highly associated with SAA, further suggesting a link between inflammation and the functional ability of HDL molecules.[2] With this evidence, it is no surprise that in patients with chronic inflammatory diseases, such as chronic kidney disease, the risk of death due to coronary artery disease (CAD) is very high.[54] The potential cause of this increased risk is increased inflammation leading to a reduction in HDL function, as well as greater concentrations of Ox-HDL that are incapable of functioning as anti-oxidants.[54, 55]

Chronically increased levels of inflammation not only help form pro-inflammatory HDL, which is ineffective for reverse cholesterol transport and for protecting LDL from oxidative stress, but also increase the metabolism of HDL, potentially resulting in a reduction in HDL levels. This is an extension of what is seen during the acute phase response to an endotoxemic challenge, during which HDL metabolism is significantly increased and HDL concentrations fall.[53] Cabana *et al.* witnessed large increases in CRP and a 90% reduction in HDL concentrations when they injected rats with croton oil.[53] This was accompanied by SAA becoming the major component in the remaining HDL molecules, inhibiting the function of whatever HDL molecules were left. In 2005, Lewis and Rader found that increases in inflammation inhibit lipoprotein lipase (LPL) activity, which transfers apoA-1 from triglyceride-rich lipoproteins to HDL.[12] LPL is responsible for HDL maturation, and the activity of this enzyme is directly associated with HDL concentration.[56, 57] LPL is primarily secreted from adipose and muscle tissue that is metabolically active, and also plays a

role in the breakdown of triglycerides. Blades *et al.* discovered in an observational study that men with lower HDL concentrations, regardless of triglyceride level, had lower LPL activity than normolipidemic controls.[57] This study compared the activity of LPL and hepatic lipase (HL) in men with low HDL and normal TG, low HDL and high TG, and normal HDL and TG. Hepatic lipase plays a significant role in the metabolism of HDL by remodeling the HDL, removing apoA-1, and generating HDL remnants that are more easily cleared by the kidneys.[12] The results showed that in men with low HDL, LPL activity was reduced, but the increase in HL activity was significantly more pronounced, leading to reductions in HDL independent from other factors such as smoking.[57]

Broedl *et al.* reported in 2004 that inflammation also increases the activity of endothelial lipase (EL), an enzyme that speeds the breakdown of HDL in the plasma.[56] This enzyme was discovered by Jaye *et al.* in 1999, receiving its name because it is synthesized by endothelial cells and acts at the endothelial surface.[12, 58] They found that when they injected mice with EL, HDL and apoA-1 were nearly eliminated. This effect was seen in both mice bred for elevated HDL and in mice bred for elevated LDL and suppressed HDL. Jin *et al.* also used a mouse model to examine the effect of EL inhibition on HDL concentrations.[59] They found that when EL was inhibited, HDL concentrations rose up to 60%, with a peak increase 48 hours after inhibition. Judging by this research, it seems that when EL is inhibited HDL levels rise,[59] but increases in EL activity reduce HDL present in the blood through increased HDL metabolism.[58] Therefore, reductions in chronic inflammation could act to increase HDL concentration by limiting the action of EL, which would attenuate the breakdown of HDL. In light of all this evidence, the reduction in anti-atherogenic capabilities of HDL and lowering of total HDL caused by inflammation contribute to the classification of atherosclerosis as an inflammatory disease.[60]

*Measuring HDL function*

In the past, measuring the function of HDL relied on a cell-based, monocyte chemotaxis assay involving human aortic endothelial cells, as described by Navab *et al.* in 2000.[61] HDL and LDL were added to cocultures of these aortic endothelial cells, followed by oxidative agents. The resulting lipid hydroperoxide was then added to a Neuroprobe chamber with isolated human monocytes. The chemotactic activity of the monocytes was measured by removing non-migrated monocytes and microscopically counting the migrated monocytes.[61] However, a cell-free assay was developed that measures HDL function by examining its ability to limit the oxidation of dichlorodihydroflourescein diacetate (DCF-DA) to fluorescent DCF, with greater reductions in standard DCF-DA oxidation rate indicating greater HDL anti-oxidant function.[17] The accuracy of this assay was confirmed by a comparison to the cell-based monocyte chemotaxis assay.[17] The ability to quench the oxidation of DCF-DA is HDL-dose dependent, so a standard quantity of HDL must be added to truly measure HDL function.[17] This cell-free assay has since been updated, allowing the use of the more stable dihydrorhodamine 123 (DHR) instead of DCF-DA to improve the inter-assay consistency.[62] Since the oxidation rate of DHR is linear when exposed to air between 10-50 min, this is the time frame during which the effects of HDL on DHR oxidation are measured.[62] The rate of oxidation is still HDL dose-dependent with this assay, with a linear effect between 2.5-15 μg of added HDL per well.[62] The intra-assay variability is low, around 6%, which lead the researchers to conclude that this assay can reliably measure HDL oxidative function.[62] However, one must be careful analyzing the results between studies because, although the assay can be run with HDL isolated by a number of different means, oxidation rates are significantly higher when the HDL is isolated by ultracentrifugation compared to fast-performance liquid chromatography and precipitation.[62]

*Physical activity and inflammation*

Along with medications and changes in diet, increasing weekly physical activity is recommended in the treatment of hypercholesterolemia.[9] While no definitive mechanism has been established for how exercise affects HDL structure and concentration, research suggests that it could be due to the effect of exercise on inflammation,[25] which as mentioned above can severely affect HDL function and metabolism. Roberts *et al.* conducted a study examining inflammatory markers, endothelial function, lipid concentrations, and rate of potential atherosclerotic progression in men with metabolic syndrome factors.[25] After 3 weeks of exercise and a high-fiber, low-fat dietary intervention, participants showed significant reductions in inflammatory markers, as well as an improved lipid profile and reduced rates of potential atherosclerotic progression. In fact, at the end of the study 9 of the 15 men who began with diagnosed metabolic syndrome no longer could be classified as having the disease. Interestingly, the participants showed significantly reduced HDL concentrations, but greater HDL function after the exercise and diet intervention, as determined by monocyte chemotaxis activity. In the discussion of the findings, Roberts *et al.* suggested that the reduction in inflammation could be responsible for the overall improvements seen in their participants.[25] This theory of physical activity decreasing chronic inflammation is consistent with what is seen elsewhere in the literature. In 2000, Mattusch *et al.* found significant reductions in resting levels of CRP after 9 months of endurance training.[19] Other research indicates that athletes exhibit both lower resting CRP levels [20] and higher HDL concentrations.[23] However, the increase in HDL concentrations is not a universal result of exercise; Roberts *et al.* found decreased HDL concentrations after their relatively short intervention.[25]

When determining the projected outlook for the risk of CVD, research by Mora *et al.* indicates that physical activity and inflammation are the two best predictors of future CVD events.[22] They employed a long term (average follow-up = 10.9 years), observational study with a cohort of

over 27,000 women with no personal history of CVD in an effort to determine the primary risk factors for CVD events. During this time, they reported that 979 CVD events occurred among their cohort. Their primary finding was a linear decrease in CVD risk with increased physical activity. The results indicate that 200-599 kcal/week, 600-1499 kcal/week, and >1500 kcal/week resulted in a 27%, 32%, and 41% decrease in CVD risk, respectively.[22] When examining the potential factors for how physical activity may reduce risk, they found that reductions in inflammation contributed to the greatest degree, with 32.6% of the risk reduction accounted for by the decreased chronic inflammation. Reductions in blood pressure explained 27.1% of the reduction in risk, with 19.1% of the reduction due to an improved lipid profile (HDL, LDL, and total cholesterol) and 15.5% attributable to increased concentrations of apoA-1.[22] Interestingly, the body mass index (BMI) of their participants did not play a primary role in the reduction in CVD risk, with the contribution only around 10% of the total reduction. In their discussion, the researchers state their belief that the increase in physical activity results in reduced inflammation, which in turn accounts for the greatest reduction in CVD risk.[22] Other large-scale studies have found similar results in regards to the negative association between physical activity and CRP. Wannamethee *et al.* completed a 20 year longitudinal study, examining the relationship between physical activity and hemostatic and inflammatory variables in 3810 elderly men.[21] The results from this study indicate that the strongest relationship for all of the variables measured was the negative association between physical activity and CRP, even when taking confounding variables into consideration. They also found that CRP levels were initially higher in men with a history of CVD, but the response to physical activity was no different regardless of previous CVD incidents.[21]

Church *et al.* also completed a longitudinal study examining the relationship between CRP concentrations and body mass and fitness in men, recruiting 722 participants from a range of ages.[26] They included body mass and waist circumference in their measures because previous findings

indicated that CRP was positively associated with body mass and waist circumference,[63] and that weight loss resulted in reductions in CRP.[64] However, these previous studies failed to take fitness into consideration. Church *et al.* found that CRP was not only negatively associated with fitness, but that the association was independent of body mass and waist circumference.[26] They also found a negative association between fitness and CRP within the overweight and obese group and within groups based on waist girth, indicating that systemic inflammation can be reduced without a reduction in body mass or waist girth.[26] Although it was not their primary aim, it is interesting to note that this was also one of the first studies to find that the changes in CRP were significant enough to place their participants into different risk categories for developing CVD based on physical fitness, and that the reduction in CRP was accompanied by a significant increase in HDL concentrations.[26] These findings led the researchers to state, "Thus, if the goal is to minimize the risk of having a high CRP value, avoiding the low fitness category should be a priority."[26]

The mechanism for reduced resting inflammation potentially lies in the adaptation to repeated acute inflammatory challenges. Taylor *et al.* recorded a 300% elevation in CRP after a strenuous, prolonged exercise bout,[65] and Siegel *et al.* reported an average CRP increase of 100% in participants immediately after completing the Boston marathon.[66] The inflammatory response to exercise appears to be dependent on the duration of the activity. Strachan *et al.* measured the CRP response in trained runners after completing events of various distances, ranging from 15km to 88km runs.[67] The CRP response was minimal in races shorter than 21km and more pronounced in the 56km and 88 km races.[67] However, CRP may not be the best indicator of acute inflammation resulting from exercise, since its release is moderated by other cytokines, such as interleukin-6 (IL-6).

IL-6 is not only produced by immune cells and the liver, it is also activated in and released from working skeletal muscle.[68] One would consider this to mean that by increasing the mass of

active skeletal muscle, the concentration of IL-6 would increase. This relationship was suggested by Febbraio and Pedersen in a review paper published in 2002.[30] However, this is not a definitive conclusion. One of the studies cited by Febbraio and Pedersen to validate this position actually conflicts with this statement. The study in question by Starkie *et al.* compared the IL-6 response to a 60 min cycling exercise or running exercise at the same metabolic workload in moderately trained individuals.[69] The results of this study showed that even though a greater muscle mass is used during running there was no difference in the magnitude of increase in IL-6.[69] The other study cited by Febbraio and Pedersen, conducted by Nieman *et al.*, does in fact agree with their statement.[70] This study also compared how running and cycling affected IL-6 levels post-exercise, but instead used experienced triathletes completing 2.5 hours of exercise. Results from this study showed that running resulted in a greater increase in IL-6 concentrations than cycling when both exercise sessions were completed at the same exercise intensity.[70] While it seems reasonable, and may very well be the case, that increased muscle mass would increase the IL-6 response to exercise, results are inconclusive.

While the influence of the size of the active muscle mass is debatable, the effect of different exercise intensities and durations is clearer. In response to an exercise bout, the anti-inflammatory cytokine IL-6 may be elevated up to 100x its resting concentration, with increases in intensity corresponding with an increased IL-6 response.[30, 31] Nielsen *et al.* completed a study in rowers finding significantly increased IL-6 concentrations and a significantly elevated immune response after only 6 min of maximal effort rowing.[71] They also reported that there was no suppression of natural killer cells, and thus immune function, which normally accompanies long duration exercise when the exercise bouts are short and highly intense in nature.[71] Ostrowski *et al.* measured the IL-6 concentrations in 53 runners before and after completing the Copenhagen marathon, finding that there was a negative correlation between finishing time and IL-6 response ($r = -0.30$, $p<0.05$).[31]

Since faster finishes correlated with an increase in IL-6 response, this finding could also be phrased to say that exercise intensity was positively correlated with IL-6 accumulation immediately post-exercise.

Other research has shown that IL-6 concentrations may not increase in prolonged activity until later in the exercise, which indicates a potential relationship between exercise duration and IL-6 response as well as exercise intensity and IL-6 response.[72] Pedersen and Febbraio suggest an exponential relationship between exercise time and increasing IL-6 concentrations, since a ten-fold increase in IL-6 requires about 2 hours of exercise and a 100-fold increase only requires about 6 hours of exercise.[73] Some researchers have suggested that this IL-6 response to increasing exercise duration may be more pronounced than the response to increasing exercise intensity since the response may be enhanced by the glycogen depletion that occurs during prolonged exercise.[68, 72] While this may be true, the IL-6 response does not seem to be limited by glycogen depletion alone, but also by limited carbohydrate utilization on the whole. Evidence supporting this theory includes a significant suppression in the IL-6 response when carbohydrates are ingested during exercise.[69] Starkie *et al.* had participants complete 60 min bouts of running or cycling either with or without carbohydrate supplementation.[69] In the carbohydrate supplementation trials, the plasma glucose concentration was significantly higher during exercise than during the trials without carbohydrate supplementation, and the increase in IL-6 was also significantly attenuated over the 60 min exercise period with carbohydrate supplementation.[69] However, IL-6 was not attenuated by increasing the glycogen stores, as carbohydrate supplementation does not spare muscle glycogen stores.[74] Carbohydrate supplementation simply allows carbohydrate oxidation to continue due to an increase in available blood glucose.[74]

The increase in IL-6 resulting from exercise is likely a response to oxidative stress and inflammation, as evidenced by delayed, yet potentially massive spikes in CRP due to exercise.[75] The

influx of IL-6, along with other anti-inflammatory cytokines, creates an anti-inflammatory environment, which can suppress the production of pro-inflammatory cytokines such as TNF-α.[76] In 2003, Starkie *et al.* demonstrated that exercise and the resulting increase in IL-6 may play a role in eliminating the increase in TNF-α in response to an endotoxin.[77] This indicates that a prior bout of exercise can be beneficial for reducing the inflammatory response to an endotoxemic challenge. The response to exercise mimics the acute phase response to an endotoxemic challenge, with large increases in inflammation and increases in Ox-LDL after both exercise and during the acute phase response.[11, 48] Sanchez-Quesada *et al.* demonstrated this relationship by measuring Ox-LDL in runners before and after a continuous 4-hour run.[11] The Ox-LDL in these highly-trained runners increased from near 10% to around 50% at the end of the run. This response could be partially explained by the modification of HDL during prolonged exercise. Margeli *et al.* studied the inflammatory and cholesterol response in participants completing a long-duration endurance event.[75] They found that while HDL concentrations were increased at the end of the event, the apoA-1 concentration steadily declined while the SAA levels skyrocketed from 3.2 mg/L to 340.8 mg/L at the end of the race, continuing to rise to 444.6 mg/L 48 hours later. This change would greatly reduce the ability of HDL to prevent LDL oxidation and participate in reverse cholesterol transport. The time courses of the inflammatory response and lipid response to extreme exercise and a traditional endotoxemic challenge are similar as well. While Margeli *et al.* showed that CRP and SAA could be elevated and apoA-1 could be depressed beyond 48 hours after an intense exercise bout,[75] Van Lenten *et al.* demonstrated that HDL may not recover its anti-inflammatory properties (recovery of apoA-1 subunits and increase in PON1 activity) until 3 days after a severe endotoxemic challenge.[48]

Through the repeated exposure to the inflammatory stress of exercise, physically active individuals also exhibit a reduced acute inflammatory response to a challenge.[27] This may be due to decreased cytokine production and a reduction in receptors on monocytes,[78] which also play a role in atherosclerosis by binding to Ox-LDL to produce foam cells. McFarlin *et al.* completed a study examining the differences in inflammatory markers and toll-like receptors in active versus inactive individuals, both young and old.[79] They found that physical activity not only reduced the chronic inflammation, but also reduced the expression of toll-like receptors, which mediate the inflammatory response. Fischer *et al.* examined how exercise training affects the muscular contraction-induced release of IL-6.[27] They found that 10 weeks of exercise training was sufficient to reduce the IL-6 response to exercise by 950%, despite the fact that the exercise workload was 44% higher at the end of the exercise training period. This evidence suggests that the reduction in chronic inflammation is most likely the result of increased antioxidant enzyme function resulting from physical activity.[19, 27-29, 80] Tyldum *et al.* illustrated this increase in anti-oxidant enzyme function with different exercise strategies by examining endothelial dysfunction following a high-fat meal.[29] The acute ingestion of a high-fat meal results in acute increases in systemic inflammation that promote endothelial dysfunction which subsides after around 4 hours.[81] This acute inflammation and endothelial dysfunction can be eliminated by simultaneously ingesting anti-oxidant vitamins along with the meal.[82] Tyldum *et al.* hypothesized that exercise may have a similar effect to the ingestion of anti-oxidant vitamins, so they examined whether a prior exercise bout would reduce the impairment in endothelial function and markers of inflammation.[29] Eight healthy men consumed a high-fat meal 3 days after either completing a continuous exercise bout, a high-intensity interval training (HIIT) bout, or with no prior exercise session. This time frame would allow the pro-inflammatory environment after exercise to return to baseline levels. The results showed that a prior exercise bout eliminated the postprandial inflammation and endothelial

dysfunction.[29] Interestingly, while the continuous exercise bout only protected against these detrimental effects, the HIIT bout actually improved the anti-oxidant status and endothelial function above resting control values, even when in the postprandial state.[29] This comparison between traditional, continuous exercise and HIIT will be explored later.

*Physical activity and HDL function*

An increase in anti-oxidant capacity results in fewer ROS, less oxidative stress, and reduced inflammation. As discussed earlier, an inflammatory challenge, which is a possible result of exercise, results in a remodeling of HDL particles.[48-51] This HDL remodeling, removing and replacing apoA-1 subunits, could be responsible for improved HDL anti-oxidant function with aerobic exercise.[25] A similar breakdown/rebuild mechanism is seen elsewhere in the body, as protein in skeletal muscle is broken down and resynthesized in order to adapt to the recurring mechanical stress of resistance exercise, a concept known as muscle plasticity.[83]

As mentioned previously, research has demonstrated that regular exercise can result in a transition from pro-inflammatory HDL to anti-inflammatory HDL, even if the HDL concentration decreases slightly.[25] This may be due to increased LPL enzyme activity resulting from exercise, as discovered by Zhang *et al.*[84] They found significantly elevated LPL activity 24 hours after an acute exercise bout in both normo- and hypo-lipidemic men.[84] In 1993, Gupta *et al.* demonstrated that athletes had significantly greater reverse cholesterol transport ability, even though the concentration of HDL was similar between the athletes and sedentary controls.[85] High-functioning anti-inflammatory HDL could be the reason why the athletes exhibited this trait, especially since the HDL concentration was similar between the athletes and sedentary individuals.[85] Evidence supporting this idea can be found in a study by Olchawa *et al.* from 2004.[86] They examined the differences in HDL function and lipid profile between trained athletes and age-matched, moderately active individuals. The results indicate that the athletes exhibited greater HDL function

and LCAT activity, along with increased HDL concentration and apoA-1 content, which were positively associated with increases in $VO_2$max.[86] The association between HDL function and $VO_2$max was also considered by Ibbora *et al.* when they examined how 18 weeks of continuous speed exercise training near 70% of $VO_2$max affects HDL concentrations and function in participants with and without type II diabetes mellitus.[87] While neither of the groups showed changes in HDL concentrations, the ability of HDL to inhibit LDL oxidation was improved in the group with type II diabetes.[87] The amount of improvement in HDL function correlated with the degree of increase in $VO_2$max at the end of the study ($r = -0.68$, $p<0.01$).[87] However, to my knowledge there has been no research completed examining the changes in HDL function resulting from different exercise intensities or different types of exercise, and how these factors might influence potential adaptations.

*Physical activity and the lipid profile*

With physical activity, not only can HDL function potentially improve, but HDL concentrations can improve as well. In a review, Leon and Sanchez found overall inconsistent cholesterol responses to aerobic endurance training, but increases in HDL were common.[88] In fact, HDL has been repeatedly shown to increase with increasing exercise volume in a dose-dependent manner.[89, 90] Both Kokkinos *et al.* in 1995 and Williams in 1997 examined the total distance run per week and lipid concentrations in runners, finding that increased mileage accumulated during a week was associated with increased HDL concentrations.[89, 90] A study by Drygas *et al.* found increased HDL concentration with increasing physical activity.[91] In this observational study, the researchers divided their 126 participants into four categories: sedentary, low activity (1-999 kcal/week), moderate activity (1000-1999 kcal/week), and high activity (>2000 kcal/week). The researchers found that a general reduction in CVD risk could be accomplished with moderate activity, but favorable changes in HDL concentration were only seen in the high activity group.[91]

This lead them to suggest that the exercise volume must be sufficient to elicit this response, potentially by generating enough oxidative stress for HDL adaptations.[91] The idea of reaching a training threshold in order to see HDL concentration improvements has been put forth by several researchers. Drygas *et al.* and others have theorized that a training volume of >2000 kcal/week may be necessary for HDL adaptations,[23, 91] while Kodama *et al.* suggested that only 900 kcal/week would suffice for moderate increases.[92] Others have used different methods to define the threshold of training required to elicit an HDL concentration increase. Farrell and Barboriak found that HDL levels increased linearly 1 mg/dl/week in participants exercising at 70% of $VO_2$max for 30 min/day, 3-4 days/week.[93] Stein *et al.* demonstrated that reaching an exercise intensity threshold near 75% of HRmax may be required to improve HDL levels.[94] In this study, participants exercised for 12 weeks at either 65%, 75%, or 85% of HRmax, but no increases in HDL were seen in participants exercising at 65% of HRmax.[94] In 2007, Slentz *et al.* published a study in agreement with previous work on exercise volume and intensity, which concluded that increasing the exercise intensity can further raise HDL levels and profoundly reduce other CHD risk factors as well.[94-96] There were 3 groups in the study, a low amount/moderate intensity group (14 kcal/kg/week, 40-55% $VO_2$max), a low amount/high intensity group (14 kcal/kg/week, 65-80% $VO_2$max), and a high amount/high intensity group (23 kcal/kg/week, 65-80% $VO_2$max). The results showed that both high intensity exercise groups saw increased HDL concentrations over baseline, but only the increase in the high amount/high intensity group was sustained over a two-week inactive period at the end of the study.[96]

These findings provide evidence that not only is the volume of exercise important, but the intensity of the exercise may be important too.[89, 94, 96] The results of studies that manipulated the exercise intensity are highly varied, with studies either failing to alter any blood lipids regardless of training intensity,[97, 98] only finding increased HDL with increases in intensity,[94] only finding

increases in HDL with lower intensity,[99] or finding that increasing the volume of exercise was better for increasing HDL levels than increasing the intensity.[95] Thomas *et al.* completed a study in 1985 that examined the effect of isocaloric exercise (500-550 kcal per session) at different intensities, but did not find any significant change in the lipid profile from baseline for any of the three groups after 11 weeks.[97] In 1984, Gaesser and Rich compared exercise at 85% of $VO_2$max and 45% of $VO_2$max over 18 weeks, but also failed to find a significant difference in any facet of the blood lipid profile despite decreases in body fat and increases in aerobic fitness in both groups.[98] However, the findings of Stein *et al.* in 1990 mentioned previously do not support these prior studies.[94] They compared three groups exercising at different intensities, either 65%, 75%, or 85% of maximum heart rate (HRmax). They found that HDL concentrations were significantly increased in the groups exercising at 75% and 85% of HRmax, but not in the group exercising at 65% of HRmax. This brought the researchers to the conclusion that an exercise intensity of at least 75% of HRmax is likely necessary in order to improve the lipid profile.[94] The results of a recent study by Nybo *et al.* do not agree with Thomas *et al.*, Gaesser and Rich, or Stein *et al*.[99] They compared HIIT to traditional endurance training and its effects on the lipid profile, and found that the lower intensity exercise was more beneficial for increasing HDL than the higher intensity exercise.[99] These results do not seem to agree at first glance with the findings of Stein *et al.,* but in the Nybo study, the "lower" intensity group exercised at 80% of HRmax, while the "high intensity" group from the study by Stein *et al*. exercised at 75% of $VO_2$max.[94, 99] Finally, Williams completed an observational study on runners, finding that training volume has a greater effect on plasma HDL concentration than training intensity, despite the finding that higher intensity was more beneficial for improving systolic and diastolic blood pressure and waist circumference.[95] None of these studies appear to be in agreement with the study by Tyldum *et al.* discussed earlier, which showed relative

improvements to be more profound after high-intensity interval training than traditional endurance training.[29]

However, each of the studies that found significant changes in HDL concentrations when examining the effect of intensity had the same methodological problem, specifically; the volume of exercise completed, as defined by total work and time, between participant groups was not matched. This is problematic due to the potential effect of exercise volume on HDL levels, since the high-intensity groups completed either a greater volume or smaller volume of exercise than their lower-intensity counterparts.[94, 95, 99] This was especially evident in the study by Nybo *et al.*, in which the high-intensity interval training group only exercised for less than 20 min per day at 95% of HRmax an average of 2 days per week, while the traditional endurance training group completed an hour of training at 80% of HRmax an average of 2.5 days per week.[99] In this light, it is no surprise that the traditional endurance training group had significant results while the high-intensity group did not. Therefore, when comparing the effect of different intensities in these studies, the effect of differing training volumes cannot be eliminated. To my knowledge, there has not been a research study that found a significant increase after training that effectively manipulated the exercise intensity while maintaining an equal exercise volume.

*High-intensity interval training*

High-intensity interval training (HIIT) is an effective way to manipulate the intensity of exercise without altering the training volume, with many studies using work- and time-matched or isocaloric HIIT and ET protocols.[32, 35, 38, 40-42, 44, 45] HIIT can be broadly defined as exercise composed of repeated bouts of relatively high-intensity exercise interspersed with active or passive recovery.[100, 101] This method of training originally gained popularity in the 1950s after Emil Zatopek, who employed HIIT, won gold medals in the 5k, 10k, and marathon events, all in Olympic record times, in the same week at the 1952 Helsinki Olympic Games. In recent years,

researchers discovered the potential health benefits of HIIT when performed by only recreationally-active or sedentary individuals. This training strategy has even been applied to elderly and diseased populations, with significant adaptations being realized, often in short amounts of time, that greatly improve health status and reduce the risk of mortality in these patients.[33-35, 37, 41, 102]

Increases in $VO_2$max are associated with increased aerobic performance. However, in highly-trained individuals, it can be difficult or nearly impossible to stimulate further increases in $VO_2$max by increasing training volume.[103, 104] However, with the inclusion of HIIT, further increases in $VO_2$max are possible in these individuals. In a study by Laursen *et al.* in 2002, the researchers added different HIIT sessions to the training program of trained cyclists and triathletes over a 4 week period.[103] At the end of the 4 weeks, the athletes who incorporated HIIT had significantly improved in $VO_2$max, as well as peak and average power, above participants who continued with their normal training.[103] In 2006, Esfarjani *et al.* added HIIT to traditional endurance training (ET) in highly-trained runners over a 10 week period, and found significant improvements in $VO_2$max, the velocity that elicits $VO_2$max (Vmax), the time that Vmax can be maintained (Tmax), as well as improved lactate threshold (LT) and 3000m time trial performance.[105] However, improving $VO_2$max is of interest outside of the realm of sport performance due to its implications for the health benefits of exercise. At least one researcher has suggested that for each 1 ml/kg/min increase in $VO_2$max there is a 15% decrease in the risk of death.[7]

Comparisons between ET and HIIT for improving $VO_2$max, along with a host of other performance- and health-related adaptations, have been common in the literature.[43, 100, 106] In a series of experiments led by Gibala, low volume HIIT, comprised of repeated 30 second Wingate tests interspersed with 4 min of passive rest, was shown to be equally or more effective for improving $VO_2$max along with other adaptations related to performance and health in sedentary or recreationally active individuals.[107-111] These adaptations include increased muscle oxidative and

buffering capacity,[107] increased lipid oxidation,[111] and increased endothelial function and artery distensibility.[110] In 2010, Hazell *et al.* used a 10 second maximum power protocol with either 2 or 4 min of passive rest in between to stimulate improvements in $VO_2$max, peak power output, and 5km time trial equal to the improvements seen using the Gibala protocol, with even greater improvements in peak training power.[112] Daussin *et al.* used a slightly different protocol to elicit adaptations in a sedentary population, and also was one of the only researchers to use a true cross-over design.[44] The HIIT protocol employed consisted of 1 minute work intervals at 90% Vmax and 4 min rest intervals at 56% of Vmax. Eleven participants (7 males, 4 females) were randomly assigned to complete either HIIT or ET. After 8 weeks of training, participants detrained for 12 weeks before completing 8 weeks of the other training protocol, which was matched for total work performed. The training sessions were initially 20 min in length, but progressed to 35 min by the end of the 8 week period. The results of this study employing repeated measures to compare the two training styles indicated that HIIT and ET were equally effective for improving $VO_2$max, but they accomplished this by different means. ET increased $VO_2$max primarily through peripheral mechanisms, increasing the arterio-venous oxygen difference (a-$vO_2$d) by increasing capillarization.[44] However, HIIT increased $VO_2$max by increasing the a-$vO_2$d through increased capillarization and mitochondrial activity, but also via a central mechanism: increasing cardiac output through increases in heart rate and stroke volume.[44] HIIT was also more effective than ET for increasing exercise time to exhaustion and $VO_2$ on-kinetics, despite the matched workloads of the two exercise training strategies.[44]

In 1996, Hellsten *et al.* completed a study examining the effect of HIIT, consisting of 10 second work intervals at peak power output with 50 sec of rest in between, on the antioxidant status in skeletal muscle.[28] They found that with frequent training sessions, HIIT was able to increase antioxidant enzyme levels in the muscle. They proposed that alternating between very

high-intensity work and low intensity work may generate a greater degree of oxidative stress, which would result in elevated anti-oxidants as a result of training.[28]

In the interest of comparing different HIIT protocols, Helgerud *et al.* compared the fitness adaptations to a short duration interval (15 sec) and a long duration interval (4 min).[32] Adaptations to these two protocols were also compared to traditional long, slow distance training and training at the lactate threshold over an 8 week period, with exercise conducted on 3 days per week. Participants were recreationally active, with an average starting $VO_2$max of 57.9 ml/kg/min. All exercise strategies were matched for total oxygen consumption per week. The short interval protocol consisted of 15 sec intervals at 90-95% of $VO_2$max interspersed with 15 sec intervals at 70% of $VO_2$max, with the total exercise session lasting 47 min. The long interval protocol involved 4 x 4 min intervals at 90-95% of $VO_2$max with 3 min rest intervals at 70% of $VO_2$max. Long, slow distance training was conducted at 70% of $VO_2$max for 45 min with lactate threshold training at 85% of $VO_2$max for 24 min. Only the HIIT protocols resulted in improvements in $VO_2$max, stroke volume and cardiac output, with no significant difference between the two.[32] Improvements were also made in running economy and velocity at which lactate threshold occurs in all groups.[32] Based on these results, both long and short high-intensity interval periods appear to be equally effective for improving $VO_2$max, cardiac output, running economy, and the velocity at which the lactate threshold occurs.

The search for an ideal HIIT protocol was also investigated by Guiraud *et al.* in 2009.[113] They compared the acute effects of 4 different HIIT protocols in participants with CHD: 1) 15 sec at 100% of $VO_2$max with 15 sec passive recovery, 2) 15 sec at 100% of $VO_2$max with 15 sec recovery at 50% of $VO_2$max, 3) 60 sec at 100% of $VO_2$max with 60 sec of passive recovery, 4) 60 sec at 100% of $Vo_2$max with 60 sec of recovery at 50% of $VO_2$max. The benefits of HIIT do not seem to be altered in people with CHD, which will be discussed later. The variables of interest in

this study were time spent above 85, 90, and 95% of maximum heart rate (HRmax), rating of perceived exertion (RPE), and time to exhaustion, with the duration of exercise capped at 35 min. They found that while participants were able to continue the 60 sec intervals with passive rest for an extended period of time, this protocol resulted in significantly less time spent with the HR above 85, 90, and 95% of HRmax, which could potentially result in fewer adaptations if this protocol were used over a period of weeks or months.[113] The 15 sec protocol with passive rest resulted in the same time spent at elevated HRs, but the protocol was able to be maintained for a significantly greater amount of time compared to the protocols with active rest (1724 sec compared to 733 and 836 sec).[113] The researchers felt that the sustained time made this protocol superior to the others,[113] but if the qualification for an effective protocol is time spent above 85, 90, and 95% of HRmax, then this protocol is no better than the protocols with active recovery at 50% of $VO_2$max but takes twice as long to complete. In this regard, the protocols with active rest, whether the intervals last 15 sec or 60 sec, appear to be equally effective for elevating the HR for a substantial period of time while only requiring a fraction of the time commitment.[113]

Training adaptations to HIIT are not limited to the young and healthy. In 1990, Makrides *et al.* demonstrated that the gains from HIIT are equal or potentially greater in old men when compared to young men.[33] In this study, 20-30 year old men and 60-70 year old men participated in 12 weeks of HIIT. This HIIT protocol consisted of 5 min at high intensity and 5 min at low intensity, with the high/low intensities increasing from 65/45% of $VO_2$max at the beginning of the study to 85/65% at the end. At the end of the 12 weeks of training, both young and old men had significantly improved $VO_2$max, cardiac output, stroke volume, a-$vO_2$d, fatigue index, and LT.[33] HIIT has also been shown to be both safe and effective in diseased populations as well, with studies including patients with coronary artery disease (CAD),[34-37, 102, 114] heart failure,[41] diabetes,[115, 116] metabolic syndrome,[40] and obesity.[39, 117]

*HIIT versus ET for health improvements*

Rognmo *et al.* published a paper in 2004 in which they compared the effect of HIIT and ET for increasing $VO_2$max in patients with a documented history of CAD.[37] Both protocols were matched for amount of work completed, with the HIIT protocol consisting of 4 min intervals at 90% of $VO_2$max and 3 min rest periods at 60% of $VO_2$max and the ET protocol consisting of 41 min of continuous-speed exercise at 50-60% of $VO_2$max. The results of this study indicated that HIIT was superior to ET for improving $VO_2$max (HIIT = 18%, ET = 8%), with a $VO_2$max improvement of 0.63% per HIIT session completed compared to just 0.29% per ET session completed.[37] This is especially impressive considering that, although these were patients with a history of CAD, the average initial $VO_2$max was 32 ml/kg/min, indicating that the participants were starting from a somewhat elevated fitness level compared to other studies using patients with CAD.[37] In 2009, Moholdt *et al.* also compared HIIT to ET for the effects on cardiovascular health and quality of life in 59 patients with CAD 4-16 weeks after undergoing bypass surgery.[35] The HIIT protocol was similar to that of Rognmo *et al.*: 4 min at 90% of $VO_2$max followed by 3 min at 70% of $VO_2$max. The ET protocol was matched for work, with participants exercising for 47 min at 70% of $VO_2$max. Exercise was completed 5 days per week for the first 4 weeks under researcher supervision, followed by 6 months of exercise completed 3-4 days per week at home. While both groups significantly increased $VO_2$max over the first 4 weeks, only the HIIT continued to improve over the following 6 month period.[35] Both groups in this study also significantly improved other metrics for quality of life.[35] In a follow-up study in 2012, Moholdt *et al.* investigated the differences in HIIT and ET in patients who had a myocardial infarction between 2-12 weeks prior to the study.[114] Participants exercised 3 times per week for 12 weeks, with the HIIT and ET protocols the same as in the previous study. They found that HIIT was twice as effective as ET for improving $VO_2$max, and both were effective for increasing endothelial function. However, only HIIT resulted

in an increase in HDL concentrations in these patients with a very recent prior history of myocardial infarction, with no significant change seen in the participants who completed ET.[114] Munk *et al.* completed a study using participants with CAD who had recently undergone a percutaneous coronary intervention, or stent implantation, to examine the effects of HIIT versus a control group on late luminal loss post-surgery and endothelial function.[36] The HIIT protocol was identical to the one used by Moholdt *et al.* in 2009, with participants beginning the exercise program an average of 11 days after surgery. After 6 months of HIIT, participants completing HIIT increased their $VO_2$max by 17% on average, which was accompanied by increases in flow-mediated dilation and reductions in resting concentrations of CRP and body mass index.[36]

In 2007, Wisloff *et al.* performed a study in patients with stable post-infarction heart failure, comparing 12 weeks of HIIT versus ET as a means to again examine which method was most effective for improving cardiovascular function and overall health outlook.[41] This study is especially interesting due to the patients' age and severely inhibited cardiac function, with an average age of 75.5 years old, average left-ventricular ejection fraction of 29%, and initial $VO_2$max of 13 ml/kg/min. The HIIT protocol again followed a similar structure as Moholdt *et al.* and Munk *et al.*, with 4 min intervals at 95% of $VO_2$max divided by 3 min intervals at 50-70% of $VO_2$max, while the isocaloric ET protocol consisted of 47 min of exercising at 70-75% of $VO_2$max. The results showed that HIIT was superior to ET for improving $VO_2$max (HIIT = 46%, ET = 14%) and work economy, as indicated by a 15% reduction in oxygen cost at a given intensity. These adaptations were accompanied by left-ventricular remodeling, increased stroke volume and cardiac output, flow-mediated dilation, and mitochondrial function, none of which were seen in the ET group.[41] These results indicate that HIIT may be vastly superior to prolonged, moderate intensity exercise for improving multiple components of general cardiovascular functioning and thus the health of the individual.

In patients with the metabolic syndrome, HIIT is an effective means for improving health and fitness. Tjonna *et al.* examined the effects of 16 weeks of HIIT or ET on patients with the metabolic syndrome, measuring changes in $VO_2$max and a host of other metabolic health markers. HIIT and ET protocols were identical to those used by Moholdt *et al.* in 2009. They found that HIIT was more effective than ET for improving $VO_2$max (HIIT = 35%, ET = 16%), as well as for improving endothelial function (HIIT = 9%, ET = 5%), improving insulin sensitivity (HIIT = 24%, ET = -22%), pancreas beta cell function (HIIT = 26%, ET = 3%), and increasing the concentration of HDL (HIIT = 22%, ET = 8%).[40] Tjonna *et al.* also completed a study comparing HIIT versus ET in obese adolescent individuals over the course of 12 months. After only 3 months of training, the HIIT group already exhibited significant increases in $VO_2$max and stroke volume over the other group, which were still significantly elevated after 12 months as well.[39] Significant improvements in HDL concentration, along with improvements in blood pressure and body composition, were only seen in the HIIT group as well, with the improvement evident after 3 months of training.[39]

While improvements are often seen in HDL concentrations in people who are initially dyslipidemic, Musa *et al.* were able to significantly improve HDL concentrations and the total cholesterol (TC):HDL ratio in normolipidemic young males using HIIT.[118] Young, normolipidemic (TC = 147 mg/dL), but untrained males were recruited to complete 8 weeks of HIIT 3 days per week. The HIIT protocol consisted of four 800 meter intervals at approximately 90% of HRmax, with a 1:1 work to rest ratio. This protocol resulted in an average workload of 423 kcal per session and 1270 kcal per week. The results show that HDL concentrations were significantly increased (18%, $p < 0.001$) and the TC:HDL ratio was significantly reduced (-18%, p , 0.0001) after 8 weeks of training. It is important to note that in each of the studies discussed above that used HIIT in either highly-trained,[103, 105] sedentary,[44, 107-112] or diseased populations,[34-37, 39-41, 102, 114-117] there

were no reports of any adverse events resulting from HIIT despite the intense work intervals, indicating that this is a safe method of training for participants from any population.

One study is especially effective for tying together the concepts of HIIT versus ET and the effect of exercise on the lipid profile, endothelial function, and systemic inflammation. Tyldum *et al.* explored the response to a high-fat meal after completing either a HIIT session or ET session.[29] When a high-fat meal is consumed, there is an acute disturbance in the blood lipid profile and endothelial function which promotes atherogenesis, including increased susceptibility of LDL to oxidation and reduced HDL concentrations.[29, 119] There is also evidence that consuming a high-fat meal increases inflammation in the body, as evidenced by increased tumor necrosis factor alpha (TNF-α) and interleukin-6 (IL-6).[120] However, when exercise is performed within 3 days prior to consuming a high-fat meal, there is a reduction in postprandial endothelial dysfunction.[121] When comparing the effect of a prior HIIT session versus an ET session on the postprandial response, Tyldum *et al.* discovered that HIIT not only attenuated postprandial endothelial dysfunction (as ET did), but improved flow-mediated dilation.[29] Along with this finding, they discovered that HIIT also improved the antioxidant status of the participants. In the discussion section of their article, the researchers suggested a potential link between the two phenomena, since they observed that following HIIT there was the greatest increase in anti-oxidant status and the greatest basal arterial diameter.[29]

*Conclusion and purpose*

This body of research indicates that there is potentially a strong and causal link between physical activity, inflammation, and HDL function and concentration. Research also suggests that HIIT is more beneficial than ET for improving anti-oxidant status as well as other primary health outcomes. Due to the potential benefits of increased exercise intensity through HIIT and the lack of research on HDL cholesterol function and concentration that adequately controlled for the effects

of different volumes, I am proposing a research study in which the relative volume (kcal/week) and duration (time/week) of exercise training are matched for each subject, but the time spent at high intensity (≥90% $VO_2$max) is manipulated. Altering the time spent at high intensity will be accomplished using HIIT in order to eliminate the effects of different volumes and durations of exercise and determine the effect of exercise intensity alone on the blood lipid profile. The effect of time spent at high intensity on chronic levels of inflammation and the acute inflammatory response to an exercise bout will also be considered.

The primary research question is this: Is high-intensity interval training (HIIT) more effective than traditional endurance training (ET) for improving HDL function? Other queries include: Does the time spent at higher intensities have an effect on the blood lipid profile when exercise relative volume and duration are matched? Does HIIT result in a greater reduction in resting inflammation than ET? How much acute inflammation is generated by HIIT and ET? Also, comparisons will be made on the effects of HIIT versus ET on blood pressure, improvements in $VO_2$max, and alterations in body composition.

REFERENCES


1. Boden, W.E., *High-density lipoprotein cholesterol as an independent risk factor in cardiovascular disease: Assessing the data from Framingham to the Veterans Affairs high-density lipoprotein intervention trial.* American Journal of Cardiology, 2000. **86**(12A): p. 19l-22l.
2. Ridker, P.M., et al., *C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women.* New England Journal of Medicine, 2000. **342**(12): p. 836-843.
3. Blair, S.N., et al., *Physical fitness and all-cause mortality. A prospective study of healthy men and women.* The Journal of the American Medical Association, 1989. **262**(17): p. 2395-401.
4. Kavanagh, T., et al., *Prediction of long-term prognosis in 12 169 men referred for cardiac rehabilitation.* Circulation, 2002. **106**(6): p. 666-671.
5. Blair, S.N., et al., *Physical fitness and all-cause mortality in hypertensive men.* Annals of Internal Medicine, 1991. **23**(3): p. 307-12.
6. Blair, S.N., et al., *Changes in physical fitness and all-cause mortality. A prospective study of healthy and unhealthy men.* The Journal of the American Medical Association, 1995. **273**(14): p. 1093-8.
7. Keteyian, S.J., et al., *Peak aerobic capacity predicts prognosis in patients with coronary heart disease.* American Heart Journal, 2008. **156**(2): p. 292-300.
8. Wilson, P.W.F., et al., *Prediction of coronary heart disease using risk factor categories.* Circulation, 1998. **97**(18): p. 1837-1847.
9. Cleeman, J.I., et al., *Executive summary of the Third Report of the National Cholesterol Education Program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III).* The Journal of the American Medical Association, 2001. **285**(19): p. 2486-2497.
10. Aviram, M. and B. Fuhrman, *LDL oxidation by arterial wall macrophages depends on the oxidative status in the lipoprotein and in the cells: Role of prooxidants vs. antioxidants.* Molecular and Cellular Biochemistry, 1998. **188**(1-2): p. 149-159.
11. SanchezQuesada, J.L., et al., *Increase of LDL susceptibility to oxidation occurring after intense, long duration aerobic exercise.* Atherosclerosis, 1995. **118**(2): p. 297-305.
12. Lewis, G.F. and D.J. Rader, *New insights into the regulation of HDL metabolism and reverse cholesterol transport.* Circulation, 2005. **96**(12): p. 1221-32.
13. Rader, D.J., et al., *Markedly Accelerated Catabolism of Apolipoprotein a-Ii (Apoa-Ii) and High-Density-Lipoproteins Containing Apoa-Ii in Classic Lecithin - Cholesterol Acyltransferase Deficiency and Fish-Eye Disease.* Journal of Clinical Investigation, 1994. **93**(1): p. 321-330.
14. Assmann, G. and A.M. Gotto, *HDL cholesterol and protective factors in atherosclerosis.* Circulation, 2004. **109**(23): p. 8-14.
15. Navab, M., et al., *High-density lipoprotein: antioxidant and anti-inflammatory properties.* Current Atherosclerosis Reports, 2007. **9**(3): p. 244-8.

16. Ansell, B.J., et al., *Inflammatory/antiinflammatory properties of high-density lipoprotein distinguish patients from control subjects better than high-density lipoprotein cholesterol levels and are favorably affected by simvastatin treatment.* Circulation, 2003. **108**(22): p. 2751-2756.
17. Navab, M., et al., *A cell-free assay for detecting HDL that is dysfunctional in preventing the formation of or inactivating oxidized phospholipids.* Journal of Lipid Research, 2001. **42**(8): p. 1308-1317.
18. Navab, M., et al., *The role of dysfunctional HDL in atherosclerosis.* Journal of Lipid Research, 2009. **50**: p. S145-S149.
19. Mattusch, F., et al., *Reduction of the plasma concentration of C-reactive protein following nine months of endurance training.* International Journal of Sports Medicine, 2000. **21**(1): p. 21-24.
20. Aronson, D., et al., *C-Reactive protein is inversely related to physical fitness in middle-aged subjects.* Atherosclerosis, 2004. **176**(1): p. 173-9.
21. Wannamethee, S.G., et al., *Physical activity and hemostatic and inflammatory variables in elderly men.* Circulation, 2002. **105**(15): p. 1785-1790.
22. Mora, S., et al., *Physical activity and reduced risk of cardiovascular events - Potential mediating mechanisms.* Circulation, 2007. **116**(19): p. 2110-2118.
23. Durstine, J.L., et al., *Blood lipid and lipoprotein adaptations to exercise - A quantitative analysis.* Sports Medicine, 2001. **31**(15): p. 1033-1062.
24. SanchezQuesada, J.L., et al., *LDL from aerobically-trained subjects shows higher resistance to oxidative modification than LDL from sedentary subjects.* Atherosclerosis, 1997. **132**(2): p. 207-213.
25. Roberts, C.K., et al., *Effect of a short-term diet and exercise intervention on oxidative stress, inflammation, MMP-9, and monocyte chemotactic activity in men with metabolic syndrome factors.* Journal of Applied Physiology, 2006. **100**(5): p. 1657-65.
26. Church, T.S., et al., *Associations between cardiorespiratory fitness and C-reactive protein in men.* Arteriosclerosis Thrombosis and Vascular Biology, 2002. **22**(11): p. 1869-1876.
27. Fischer, C.P., et al., *Endurance training reduces the contraction-induced interleukin-6 mRNA expression in human skeletal muscle.* American Journal of Physiology-Endocrinology and Metabolism, 2004. **287**(6): p. E1189-E1194.
28. Hellsten, Y., F.S. Apple, and B. Sjodin, *Effect of sprint cycle training on activities of antioxidant enzymes in human skeletal muscle.* Journal of Applied Physiology, 1996. **81**(4): p. 1484-1487.
29. Tyldum, G.A., et al., *Endothelial Dysfunction Induced by Post-Prandial Lipemia Complete Protection Afforded by High-Intensity Aerobic Interval Exercise.* Journal of the American College of Cardiology, 2009. **53**(2): p. 200-206.
30. Febbraio, M.A. and B.K. Pedersen, *Muscle-derived interleukin-6: mechanisms for activation and possible biological roles.* Faseb Journal, 2002. **16**(11): p. 1335-1347.
31. Ostrowski, K., P. Schjerling, and B.K. Pedersen, *Physical activity and plasma interleukin-6 in humans - effect of intensity of exercise.* European Journal of Applied Physiology, 2000. **83**(6): p. 512-515.
32. Helgerud, J., et al., *Aerobic high-intensity intervals improve VO2max more than moderate training.* Medicine and Science in Sports and Exercise, 2007. **39**(4): p. 665-671.
33. Makrides, L., G.J. Heigenhauser, and N.L. Jones, *High-intensity endurance training in 20- to 30- and 60- to 70-yr-old healthy men.* Journal of Applied Physiology, 1990. **69**(5): p. 1792-8.
34. Amundsen, B.H., et al., *High-intensity aerobic exercise improves diastolic function in coronary artery disease.* Scandinavian Cardiovascular Journal, 2008. **42**(2): p. 110-117.

35. Moholdt, T.T., et al., *Aerobic interval training versus continuous moderate exercise after coronary artery bypass surgery: A randomized study of cardiovascular effects and quality of life.* American Heart Journal, 2009. **158**(6): p. 1031-1037.
36. Munk, P.S., et al., *High-intensity interval training may reduce in-stent restenosis following percutaneous coronary intervention with stent implantation: A randomized controlled trial evaluating the relationship to endothelial function and inflammation.* American Heart Journal, 2009. **158**(5): p. 734-741.
37. Rognmo, O., et al., *High intensity aerobic interval exercise is superior to moderate intensity exercise for increasing aerobic capacity in patients with coronary artery disease.* European Journal of Cardiovascular Prevention & Rehabilitation, 2004. **11**(3): p. 216-222.
38. Schjerve, I.E., et al., *Both aerobic endurance and strength training programmes improve cardiovascular health in obese adults.* Clinical Science, 2008. **115**(9-10): p. 283-293.
39. Tjonna, A.E., et al., *Aerobic interval training reduces cardiovascular risk factors more than a multitreatment approach in overweight adolescents.* Clinical Science, 2009. **116**(3-4): p. 317-326.
40. Tjonna, A.E., et al., *Aerobic interval training versus continuous moderate exercise as a treatment for the metabolic syndrome - A pilot study.* Circulation, 2008. **118**(4): p. 346-354.
41. Wisloff, U., et al., *Superior cardiovascular effect of aerobic interval training versus moderate continuous training in heart failure patients - A randomized study.* Circulation, 2007. **115**(24): p. 3086-3094.
42. Gormley, S.E., et al., *Effect of Intensity of Aerobic Training on VO2max.* Medicine and Science in Sports and Exercise, 2008. **40**(5): p. S42-S42.
43. Laursen, P.B., *Training for intense exercise performance: high-intensity or high-volume training?* Scandinavian Journal of Medicine & Science in Sports, 2010. **20**: p. 1-10.
44. Daussin, F.N., et al., *Effect of interval versus continuous training on cardiorespiratory and mitochondrial functions: relationship to aerobic performance improvements in sedentary subjects.* American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 2008. **295**(1): p. R264-R272.
45. Ciolac, E.G., et al., *Effects of high-intensity aerobic interval training vs. moderate exercise on hemodynamic, metabolic and neuro-humoral abnormalities of young normotensive women at high familial risk for hypertension.* Hypertension Research, 2010. **33**(8): p. 836-843.
46. Honda, H., et al., *Oxidized high-density lipoprotein as a risk factor for cardiovascular events in prevalent hemodialysis patients.* Atherosclerosis, 2012. **220**(2): p. 493-501.
47. Honda, H., et al., *Serum albumin, C-reactive protein, interleukin 6, and fetuin A as predictors of malnutrition, cardiovascular disease, and mortality in patients with ESRD.* American Journal of Kidney Diseases, 2006. **47**(1): p. 139-148.
48. VanLenten, B.J., et al., *Anti-inflammatory HDL becomes pro-inflammatory during the acute phase response - Loss of protective effect of HDL against LDL oxidation in aortic wall cell cocultures.* Journal of Clinical Investigation, 1995. **96**(6): p. 2758-2767.
49. Feingold, K.R., et al., *Paraoxonase activity in the serum and hepatic mRNA levels decrease during the acute phase response.* Atherosclerosis, 1998. **139**(2): p. 307-315.
50. Lindhorst, E., et al., *Acute inflammation, acute phase serum amyloid A and cholesterol metabolism in the mouse.* Biochimica et Biophysica Acta, 1997. **1339**(1): p. 143-54.
51. Kunimoto, S., et al., *Steryl glucoside is a lipid mediator in stress-responsive signal transduction.* Cell Structure and Function, 2002. **27**(3): p. 157-162.
52. Ridker, P.M., *On evolutionary biology, inflammation, infection, and the causes of atherosclerosis.* Circulation, 2002. **105**(1): p. 2-4.

53. Cabana, V.G., J.N. Siegel, and S.M. Sabesin, *Effects of the Acute Phase Response on the Concentration and Density Distribution of Plasma-Lipids and Apolipoproteins.* Journal of Lipid Research, 1989. **30**(1): p. 39-49.
54. Navab, K.D., et al., *Chronic Inflammatory Disorders and Accelerated Atherosclerosis: Chronic Kidney Disease.* Current Pharmaceutical Design, 2011. **17**(1): p. 17-20.
55. Vaziri, N.D., M. Navab, and A.M. Fogelman, *HDL metabolism and activity in chronic kidney disease.* Nature Reviews Nephrology, 2010. **6**(5): p. 287-296.
56. Broedl, U.C., W. Jin, and D.J. Rader, *Endothelial lipase: a modulator of lipoprotein metabolism upregulated by inflammation.* Trends in Cardiovascular Medicine, 2004. **14**(5): p. 202-6.
57. Blades, B., G.L. Vega, and S.M. Grundy, *Activities of Lipoprotein-Lipase and Hepatic Triglyceride Lipase in Postheparin Plasma of Patients with Low Concentrations of Hdl Cholesterol.* Arteriosclerosis and Thrombosis, 1993. **13**(8): p. 1227-1235.
58. Jaye, M., et al., *A novel endothelial-derived lipase that modulates HDL metabolism.* Nature Genetics, 1999. **21**(4): p. 424-428.
59. Jin, W., et al., *Inhibition of endothelial lipase causes increased HDL cholesterol levels in vivo.* Journal of Clinical Investigation, 2003. **111**(3): p. 357-62.
60. Ross, R., *Mechanisms of disease - Atherosclerosis - An inflammatory disease.* New England Journal of Medicine, 1999. **340**(2): p. 115-126.
61. Navab, M., et al., *Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein: step 1.* Journal of Lipid Research, 2000. **41**(9): p. 1481-1494.
62. Kelesidis, T., et al., *A biochemical fluorometric method for assessing the oxidative properties of HDL.* Journal of Lipid Research, 2011. **52**(12): p. 2341-2351.
63. Festa, A., et al., *The relation of body fat mass and distribution to markers of chronic inflammation.* International Journal of Obesity, 2001. **25**(10): p. 1407-1415.
64. Tchernof, A., et al., *Weight loss reduces C-reactive protein levels in obese postmenopausal women.* Circulation, 2002. **105**(5): p. 564-569.
65. Taylor, C., et al., *Hematologic, Iron-Related, and Acute-Phase Protein Responses to Sustained Strenuous Exercise.* Journal of Applied Physiology, 1987. **62**(2): p. 464-469.
66. Siegel, A.J., et al., *Effect of marathon running on inflammatory and hemostatic markers.* American Journal of Cardiology, 2001. **88**(8): p. 918-+.
67. Strachan, A.F., et al., *C Reactive Protein Concentrations during Long-Distance Running.* British Medical Journal, 1984. **289**(6454): p. 1249-1251.
68. Pedersen, B.K., et al., *Muscle-derived interleukin-6: lipolytic, anti-inflammatory and immune regulatory effects.* Pflugers Archiv-European Journal of Physiology, 2003. **446**(1): p. 9-16.
69. Starkie, R.L., et al., *Carbohydrate ingestion attenuates the increase in plasma interleukin-6, but not skeletal muscle interleukin-6 mRNA, during exercise in humans.* Journal of Physiology-London, 2001. **533**(2): p. 585-591.
70. Nieman, D.C., et al., *Influence of mode and carbohydrate on the cytokine response to heavy exertion.* Medicine and Science in Sports and Exercise, 1998. **30**(5): p. 671-678.
71. Nielsen, H.B., et al., *Lymphocytes and NK cell activity during repeated bouts of maximal exercise.* American Journal of Physiology, 1996. **271**(1 Pt 2): p. R222-7.
72. Ostrowski, K., et al., *Chemokines are elevated in plasma after strenuous exercise in humans.* European Journal of Applied Physiology, 2001. **84**(3): p. 244-245.
73. Pedersen, B.K. and M.A. Febbraio, *Muscle as an endocrine organ: Focus on muscle-derived interleukin-6.* Physiological Reviews, 2008. **88**(4): p. 1379-1406.
74. Coyle, E.F., et al., *Muscle Glycogen Utilization during Prolonged Strenuous Exercise When Fed Carbohydrate.* Journal of Applied Physiology, 1986. **61**(1): p. 165-172.

75. Margeli, A., et al., *Dramatic elevations of interleukin-6 and acute-phase reactants in athletes participating in the ultradistance foot race spartathlon: severe systemic inflammation and lipid and lipoprotein changes in protracted exercise.* Journal of Clinical Endocrinology & Metabolism, 2005. **90**(7): p. 3914-8.
76. Matthys, P., et al., *Anti-gamma interferon and anti-interleukin-6 antibodies affect staphylococcal enterotoxin B-induced weight loss, hypoglycemia, and cytokine release in D-galactosamine-sensitized and unsensitized mice.* Infection and Immunity, 1995. **63**(4): p. 1158-64.
77. Starkie, R., et al., *Exercise and IL-6 infusion inhibit endotoxin-induced TNF-alpha production in humans.* Faseb Journal, 2003. **17**(3): p. 884-+.
78. Gleeson, M., B. McFarlin, and M. Flynn, *Exercise and Toll-like receptors.* Exercise Immunology Review, 2006. **12**: p. 34-53.
79. McFarlin, B.K., et al., *Physical activity status, but not age, influences inflammatory biomarkers and toll-like receptor 4.* Journals of Gerontology Series a-Biological Sciences and Medical Sciences, 2006. **61**(4): p. 388-393.
80. Powers, S.K., L.L. Ji, and C. Leeuwenburgh, *Exercise training-induced alterations in skeletal muscle antioxidant capacity: a brief review.* Medicine and Science in Sports and Exercise, 1999. **31**(7): p. 987-997.
81. van Oostrom, A.J.H.H.M., et al., *Postprandial recruitment of neutrophils may contribute to endothelial dysfunction.* Journal of Lipid Research, 2003. **44**(3): p. 576-583.
82. Plotnick, G.D., M.C. Corretti, and R.A. Vogel, *Effect of antioxidant vitamins on the transient impairment of endothelium-dependent brachial artery vasoactivity following a single high-fat meal.* The Journal of the American Medical Association, 1997. **278**(20): p. 1682-1686.
83. Booth, F.W. and K.M. Baldwin, *Muscle Plasticity: Energy Demand and Supply Processes*, in *Comprehensive Physiology*2010, John Wiley & Sons, Inc.
84. Zhang, J.Q., et al., *Changes in LPLa and reverse cholesterol transport variables during 24-h postexercise period.* American Journal of Physiology-Endocrinology and Metabolism, 2002. **283**(2): p. E267-E274.
85. Gupta, A.K., et al., *Increased Reverse Cholesterol Transport in Athletes.* Metabolism-Clinical and Experimental, 1993. **42**(6): p. 684-690.
86. Olchawa, B., et al., *Physical fitness and reverse cholesterol transport.* Arteriosclerosis Thrombosis and Vascular Biology, 2004. **24**(6): p. 1087-1091.
87. Iborra, R.T., et al., *Aerobic exercise training improves the role of high-density lipoprotein antioxidant and reduces plasma lipid peroxidation in type 2 diabetes mellitus.* Scandinavian Journal of Medicine & Science in Sports, 2008. **18**(6): p. 742-750.
88. Leon, A.S. and O.A. Sanchez, *Response of blood lipids to exercise training alone or combined with dietary intervention.* Medicine and Science in Sports and Exercise, 2001. **33**(6 Suppl): p. S502-15; discussion S528-9.
89. Kokkinos, P.F., et al., *Miles Run Per Week and High-Density-Lipoprotein Cholesterol Levels in Healthy, Middle-Aged Men - a Dose-Response Relationship.* Archives of Internal Medicine, 1995. **155**(4): p. 415-420.
90. Williams, P.T., *Relationship of distance run per week to coronary heart disease risk factors in 8283 male runners - The national runners' health study.* Archives of Internal Medicine, 1997. **157**(2): p. 191-198.
91. Drygas, W., et al., *Long-term effects of different physical activity levels on coronary heart disease risk factors in middle-aged men.* International Journal of Sports Medicine, 2000. **21**(4): p. 235-241.

92. Kodama, S., et al., *Effect of aerobic exercise training on serum levels of high-density lipoprotein cholesterol - A meta-analysis.* Archives of Internal Medicine, 2007. **167**(10): p. 999-1008.
93. Farrell, P.A. and J. Barboriak, *The Time Course of Alterations in Plasma-Lipid and Lipoprotein Concentrations during 8 Weeks of Endurance Training.* Atherosclerosis, 1980. **37**(2): p. 231-238.
94. Stein, R.A., et al., *Effects of Different Exercise Training Intensities on Lipoprotein Cholesterol Fractions in Healthy Middle-Aged Men.* American Heart Journal, 1990. **119**(2): p. 277-283.
95. Williams, P.T., *Relationships of heart disease risk factors to exercise quantity and intensity.* Archives of Internal Medicine, 1998. **158**(3): p. 237-245.
96. Slentz, C.A., et al., *Inactivity, exercise training and detraining, and plasma lipoproteins. STRRIDE: a randomized, controlled study of exercise intensity and amount.* Journal of Applied Physiology, 2007. **103**(2): p. 432-42.
97. Thomas, T.R., et al., *Effects of interval and continuous running on HDL-cholesterol, apoproteins A-1 and B, and LCAT.* Canadian Journal of Applied Sports Science, 1985. **10**(1): p. 52-9.
98. Gaesser, G.A. and R.G. Rich, *Effects of High-Intensity and Low-Intensity Exercise Training on Aerobic Capacity and Blood-Lipids.* Medicine and Science in Sports and Exercise, 1984. **16**(3): p. 269-274.
99. Nybo, L., et al., *High-Intensity Training versus Traditional Exercise Interventions for Promoting Health.* Medicine and Science in Sports and Exercise, 2010. **42**(10): p. 1951-1958.
100. Billat, L.V., *Interval training for performance: A scientific and empirical practice - Special recommendations for middle- and long-distance running. Part II: Anaerobic interval training.* Sports Medicine, 2001. **31**(2): p. 75-90.
101. Laursen, P.B. and D.G. Jenkins, *The scientific basis for high-intensity interval training - Optimising training programmes and maximising performance in highly trained endurance athletes.* Sports Medicine, 2002. **32**(1): p. 53-73.
102. Warburton, D.E., et al., *Effectiveness of high-intensity interval training for the rehabilitation of patients with coronary artery disease.* American Journal of Cardiology, 2005. **95**(9): p. 1080-4.
103. Laursen, P.B., et al., *Interval training program optimization in highly trained endurance cyclists.* Medicine and Science in Sports and Exercise, 2002. **34**(11): p. 1801-1807.
104. Londeree, B.R., *Effect of training on lactate/ventilatory thresholds: A meta-analysis.* Medicine and Science in Sports and Exercise, 1997. **29**(6): p. 837-843.
105. Esfarjani, F. and P.B. Laursen, *Manipulating high-intensity interval training: Effects on VO2max, the lactate threshold and 3000 m running performance in moderately trained mates.* Journal of Science and Medicine in Sport, 2007. **10**(1): p. 27-35.
106. Cornish, A.K., S. Broadbent, and B.S. Cheema, *Interval training for patients with coronary artery disease: a systematic review.* European Journal of Applied Physiology, 2011. **111**(4): p. 579-589.
107. Gibala, M.J. and S.L. Mcgee, *Metabolic adaptations to short-term high-intensity interval training: A little pain for a lot of gain?* Exercise and Sport Sciences Reviews, 2008. **36**(2): p. 58-63.
108. Burgomaster, K.A., et al., *Divergent response of metabolite transport proteins in human skeletal muscle after sprint interval training and detraining.* American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 2007. **292**(5): p. R1970-6.
109. Burgomaster, K.A., et al., *Similar metabolic adaptations during exercise after low volume sprint interval and traditional endurance training in humans.* Journal of Physiology, 2008. **586**(1): p. 151-60.
110. Rakobowchuk, M., et al., *Sprint interval and traditional endurance training induce similar improvements in peripheral arterial stiffness and flow-mediated dilation in healthy humans.*

American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 2008. **295**(1): p. R236-R242.
111. Burgomaster, K.A., G.J.F. Heigenhauser, and M.J. Gibala, *Effect of short-term sprint interval training on human skeletal muscle carbohydrate metabolism during exercise and time-trial performance.* Journal of Applied Physiology, 2006. **100**(6): p. 2041-2047.
112. Hazell, T.J., et al., *10 or 30-s sprint interval training bouts enhance both aerobic and anaerobic performance.* European Journal of Applied Physiology, 2010. **110**(1): p. 153-160.
113. Guiraud, T., et al., *Optimization of high intensity interval exercise in coronary heart disease.* European Journal of Applied Physiology, 2010. **108**(4): p. 733-740.
114. Moholdt, T., et al., *Aerobic interval training increases peak oxygen uptake more than usual care exercise training in myocardial infarction patients: a randomized controlled study.* Clinical Rehabilitation, 2012. **26**(1): p. 33-44.
115. Harmer, A.R., et al., *Sprint Training Increases Muscle Oxidative Metabolism During High-Intensity Exercise in Patients With Type 1 Diabetes.* Diabetes Care, 2008. **31**(11): p. 2097-2102.
116. Little, J.P., et al., *Low-volume high-intensity interval training reduces hyperglycemia and increases muscle mitochondrial capacity in patients with type 2 diabetes.* Journal of Applied Physiology, 2011. **111**(6): p. 1554-1560.
117. Whyte, L.J., J.M.R. Gill, and A.J. Cathcart, *Effect of 2 weeks of sprint interval training on health-related outcomes in sedentary overweight/obese men.* Metabolism-Clinical and Experimental, 2010. **59**(10): p. 1421-1428.
118. Musa, D.I., et al., *The Effect of a High-Intensity Interval Training Program on High-Density Lipoprotein Cholesterol in Young Men.* Journal of Strength and Conditioning Research, 2009. **23**(2): p. 587-592.
119. Cohn, J.S., *Postprandial lipemia: Emerging evidence for atherogenicity of remnant lipoproteins.* Canadian Journal of Cardiology, 1998. **14**: p. 18b-27b.
120. Nappo, F., et al., *Postprandial endothelial activation in healthy subjects and in type 2 diabetic patients: Role of fat and carbohydrate meals.* Journal of the American College of Cardiology, 2002. **39**(7): p. 1145-1150.
121. Gill, J.M.R., et al., *Effects of prior moderate exercise on postprandial metabolism and vascular function in lean and centrally obese men.* Journal of the American College of Cardiology, 2004. **44**(12): p. 2375-2382.

# CHAPTER III

## JOURNAL MANUSCRIPT


### ABSTRACT

INTRODUCTION: High-density lipoprotein (HDL) plays an important role in the prevention of atherosclerosis and coronary heart disease (CHD), which is the leading cause of death in the United States. While the importance of adequate concentrations of HDL is recognized, HDL must exhibit anti-inflammatory properties and participate in reverse cholesterol transport to be beneficial. Chronic C-reactive protein (CRP) levels may indicate whether HDL can function in this manner, with reductions in CRP being associated with greater HDL function and thus protection from CHD. Traditional endurance training (ET) is an effective way to reduce chronic CRP concentration, increase HDL function, and increase HDL concentration. However, High-intensity Interval Training (HIIT) is equal or superior to ET for improving measures of cardiovascular health across a wide range of populations, and may be more beneficial than ET for improving inflammatory status, HDL function, and HDL concentration. PURPOSE: To compare the effects of duration- and work-matched ET and HIIT on HDL function and concentration, as well as on CRP, the acute IL-6 response to exercise, and cardiovascular fitness. METHODS: Twelve young males (age 21.6 $\pm$ 1.6 years, HDL 34 $\pm$ 8 mg/dL, $VO_2$max 41.6 $\pm$ 5.4 ml/kg/min) divided into 2 matched groups (HIIT or ET) based on HDL concentration and $VO_2$max completed 8 weeks of duration- and work-matched exercise training 3 days per week. Each exercise session lasted 30 min, with a progression in average intensity from 70 to 80% of $VO_2$max. HDL function and concentration, resting CRP, acute IL-6 response to exercise, $VO_2$max, body composition, and

blood pressure were determined before and after the 8 weeks of training. RESULTS: After 8 weeks of training, there were no significant differences in HDL function, resting CRP, HDL concentration, or $VO_2$max within or between groups ($p>0.05$). HIIT lowered plasma triglyceride (TRG) concentrations (-31 ± 28 mg/dL, $p=0.04$) significantly more than ET ($p=0.009$). HIIT also significantly increased treadmill Vmax (0.6 ± 0.5 mph, $p=0.02$) and reduced HRmax (-5 ± 3 bpm, $p=0.01$) from baseline levels, but there was no significant difference between the groups. ET resulted in significant reductions in the percentage of android fat (-2.60 ± 2.41%, $p=0.045$) and TC:HDL ratio (-0.60 ± 0.41, $p=0.02$), but neither were significantly different from HIIT. CONCLUSION: When average intensity, workload, and duration are equal, these results indicate no difference between HIIT and ET for improving HDL function, HDL concentrations, or resting CRP. However, in this time period HIIT was significantly better than ET for reducing plasma TRG concentrations. Also, HIIT alone improved Vmax and reduced HRmax, while only ET was beneficial for reducing the percentage of android fat. A larger sample size, longer training period, or different exercise protocols may be necessary to alter HDL function and chronic inflammation.

# INTRODUCTION

High-density lipoprotein (HDL) plays a vital role in the prevention of atherosclerosis and coronary heart disease (CHD), which according to the CDC is the leading cause of death in the United States, claiming nearly 600,000 lives per year. Atherosclerosis progresses as low-density lipoprotein (LDL) is oxidized (Ox-LDL) before binding to macrophages in endothelial cells. Foam cells are formed as a result, which can accumulate and progress into fatty streaks that eventually form plaque. HDL works not only as an anti-inflammatory molecule to prevent the oxidation of LDL, but also participates in reverse cholesterol transport to remove Ox-LDL from the endothelial cells and dispose of it in the liver. The importance of maintaining sufficient quantities of HDL to perform these functions has been the focus of much research, as evidence suggests an inverse association between HDL concentrations and risk of CHD events.[1, 2] However, the presence of a seemingly adequate concentration of HDL does not necessarily indicate protection from CHD.[15] In the Framingham Heart Study, 44% of men and 43% of women who had a CHD event had HDL concentrations above the recommended levels.[16]

The failure of HDL to protect against a CHD event can be explained by the differences in the functional ability of HDL. Ansell *et al.* compared the functional ability of HDL from healthy donors and individuals with CHD when HDL concentrations were matched, finding that the HDL from CHD patients was pro-inflammatory and less capable of reverse cholesterol transport compared to the HDL from healthy donors.[16] The potential cause of this reduction in HDL function is persistent low-grade inflammation.

In the acute inflammatory response to an endotoxemic challenge, the structure of HDL is modified, losing apolipoprotein A-1 (apoA-1) subunits and gaining serum amyloid A (SAA) subunits, resulting in a pro-inflammatory HDL molecule or an oxidized HDL (Ox-HDL) molecule that enhances the formation of Ox-LDL.[48-50, 55] When the endotoxemic challenge ends, the systemic inflammation subsides and HDL regains its anti-inflammatory properties.[50] However, patients with CHD or at high risk of CHD exhibit chronic low-grade inflammation, which results in persistent pro-inflammatory HDL and Ox-HDL. It is therefore not surprising that chronic inflammation matched with increased pro-inflammatory HDL and Ox-HDL is a strong predictor of CHD mortality, regardless of HDL concentration.[16, 46, 52, 54]

Regular exercise has the beneficial effect of reducing chronic inflammation,[19-21] which could explain how exercise also increases HDL concentration and HDL anti-inflammatory function,[22, 23] helping decrease the risk of CHD.[22] This adaptation is potentially the result of repeated, exercise-induced inflammatory challenges that results in greater anti-oxidant enzyme function.[27-29] High-intensity interval training (HIIT) may be more beneficial than traditional endurance training (ET) for reducing chronic inflammation,[28] as increased exercise intensity generates a greater acute inflammatory challenge.[30, 31] While adaptations to HIIT and ET have been compared for a wide range of variables,[29, 37, 40, 44, 107, 114] no research has directly compared the potential effects of HIIT vs ET on HDL function.

Therefore, the purpose of this study is to compare the effect of 8 weeks of HIIT vs ET on HDL function and concentration, resting inflammation, and the acute inflammatory response. Secondary measures will include comparisons of adaptations in $VO_2$max, running speed at $VO_2$max (Vmax), body composition, and blood pressure.

## METHODS AND PROCEDURES

*Participants*

Participants were recruited by word of mouth from the local community and completed informed consent and screening documentation in accordance with the Auburn University Institutional Review Board. Participants were included if they were not currently engaging in vigorous intensity physical activity on more than two days of the week, as defined by the American College of Sports Medicine,[122] and did not use medications that could increase the risks associated with vigorous intensity physical activity or affect testing variables, such as antihypertensive, antihyperlipidemic, antiarrhythmic, anti-inflammatory, and corticosteroidal medications. A power analysis prior to participant recruitment indicated that at least 5 participants in each group were needed. Fourteen young males volunteered (age 21.4 $\pm$ 1.6) with an average height of 1.81 $\pm$ 0.08 m and average weight of 86.63 $\pm$ 15.82 kg. The average initial HDL concentration, $VO_2$max, and percent fat for participants was 34 $\pm$ 8 mg/dL, 41.6 $\pm$ 5.4 ml/kg/min, and 28.1 $\pm$ 7.17%, respectively. Table 1 provides a complete description of initial participant characteristics. No participants were dyslipidemic (TC>200) or hypertensive (>140/90) at the start of the study. Females were not included due to the variability in blood lipid profile during the menstrual cycle, which is exacerbated by the use of oral contraceptives.[123, 124]

*Testing Days*

Participants were asked to refrain from vigorous activity for 2 days prior to reporting to the lab in order to allow for any inflammatory response to previous activity to return to baseline.[125]

They were also instructed to refrain from consuming alcohol and caffeine for 24 hours before the testing day, and to come to the lab after an overnight fast, allowing the inflammatory response to feeding to pass and the lipid profile to be measured at a fasting level.[81, 121, 126] Height and weight were recorded, and body composition was determined using dual x-ray absorbance (GE Healthcare Lunar, Madison, WI), rendering measures of percent body fat, android fat, gynoid fat, and bone mineral density. Blood pressure was measured using a sphygmomanometer after sitting quietly for 10 min in a semi-reclined position. Blood lipid profile was then determined via whole blood sample acquired by finger prick using the Cholestech LDX system (Alere Inc., Waltham, MA). Another blood sample was then collected via venipuncture to be used for future analysis. Finally, participants completed a $VO_2$max test to determine aerobic fitness. Testing days occurred before the start of training and at the end of 8 weeks of training. The $VO_2$max test was repeated an additional time after 4 weeks of training to track participants' fitness and update the training protocols.

*$VO_2$max and Vmax*

Each participant completed a graded exercise test on a motorized treadmill (Woodway Desmo, Woodway USA, Waukesha, WI) to determine $VO_2$max using a ParvoMedics TrueMax 2400 metabolic testing cart (ParvoMedics, East Sandy, UT), which demonstrates excellent reliability and accuracy.[127, 128] Heart rate was continuously monitored and logged using the Polar Team2 system (Polar Electro Inc., Lake Success, NY). The protocol consisted of four 3 min stages with speeds increasing by 0.5 mph at each stage. If the fourth stage was completed, then the incline was increased 2% every min until $VO_2$max was reached. If no plateau in $VO_2$ was reached at the end of the test, then 2 of the following criteria were met in order to consider it a valid test: 1) heart rate within 10 beats of age-predicted max (220 – age), 2) RER > 1.15, or 3) volitional exhaustion. Values were measured every 15 sec and averaged for each minute. For the safety of the

participants, if Vmax was not determined in the first four stages of the $VO_2$max test, then it was estimated by extrapolating the relationship between speed and $VO_2$ over the first four stages. The average correlation between speed and $VO_2$ over the four submaximal stages used to estimate Vmax was $r^2$=0.9839.

*Blood sample collection*

Blood samples were collected via finger prick and venipuncture using aseptic technique by a trained phlebotomist. Samples to be used for determining HDL function, resting inflammation, and acute inflammatory response were collected in sodium heparin-treated tubes (Vacutainer, Becton Dickenson, Franklin Lakes, NJ). Sodium heparin treated samples were centrifuged within 10 min of sample collection for 10 min at 6000 rpm and 0°C. The plasma was then transferred into 1.5 ml microtubes and stored for later analysis at -80°C.

*Blood Lipid Profile*

A complete blood lipid profile was acquired using the Cholestech LDX system with Lipid Profile + Glu cartridges. This system has excellent accuracy and precision when measuring the lipid profile, with accuracies for TC, HDL, and TRG being within 1.6, 2.74, and 2.11% respectively and precisions of 3.05, 1.05, and 2.65%.[129] A whole blood sample was taken via finger prick after an overnight fast and sampled within 20 sec after initial prick. The profile included measurements of TC, HDL, TC:HDL ratio, TRG, and Glu and a calculation for LDL using the Friedewald formula:[130]

$$LDL = TC - HDL - (TRG/5) \quad \text{(eq. 1)}$$

*HDL function*

HDL function was determined by a cell-free assay as described by Kelesidis *et al.*[62] Iron-free 4-(2-Hydroxyethyl)piperazine-1-ethanesulfonic acid, N-(2-Hydroxyethyl)piperazine-N′-(2-ethanesulfonic acid)(HEPES)-buffered saline (HBS) was made with concentrations of 20 mM

HEPES, 150 mM NaCl, and 0.08 μM deferoxamine mesylate salt (Sigma-Aldrich, St. Louis, MO). 50 mM dihydrorhodamine 123 (DHR) suspended in dimethyl sulfoxide (Life Technologies, Grand Island, NY) was diluted in HBS to 50 μM DHR solution. Five micrograms of HDL isolated from participants' blood samples via precipitation were added to each well of a 96-well, black polypropylene plate (NUNC, Roskilde, Denmark), along with 150 microliters of HBS and 25 microliters of 50 μM DHR solution. Fluorescence was measured every 2 min for 60 min at wavelengths of 485 nm excitation and 538 nm emission. The oxidation rate was determined by the slope of the linear regression between 10 and 50 min measured in fluorescence units per minute (FU/min). Greater HDL function is indicated by a slower rate of oxidation of DHR, and thus a lower slope, over the 10-50 min period.[62]

*Chronic Inflammation*

A traditional laboratory CRP assay was used to measure chronic, resting inflammation levels before the start of training and at the end of the 8 week training period. CRP was determined using a human CRP sandwich enzyme immunoassay (Quantikine ELISA, R&D Systems, Minneapolis, MN). Plasma samples were diluted 100-fold with calibrator diluent, with 50 μL of sample added to each well. After addition of CRP conjugate and incubation, substrate solution was added to each well and allowed to incubate for 30 min. Stop solution was then added and the plate was read at 450 nm with a 570 nm correction. All samples were measured in triplicate.

*Acute Inflammatory Response*

The acute inflammatory response to the different exercise protocols and the change in acute inflammatory response after 8 weeks of exercise were determined by measuring plasma IL-6 concentrations immediately before and within two minutes after the exercise sessions on the first and last day of exercise in the study. Plasma IL-6 concentrations were measured using a human IL-6 enzyme immunoassay (ELISA Ready-Set-Go!, eBioscience, San Diego, CA). Plates were coated

with capture antibody in coating buffer and blocked with assay diluent. 100 µL of plasma was added to each well followed by an overnight incubation. Detection antibody, substrate solution, and, after a 30 min incubation, stop solution were added. The plate was read at 450 nm with a 570 nm correction. All samples were measured in triplicate.

*Groups*

Participants were placed into matched groups based on HDL concentration and aerobic fitness, as determined by $VO_2$max: a high-intensity interval training group (HIIT) and a traditional endurance training group (ET). Baseline values for HDL concentration were 34 ± 9 for the HIIT group and 34 ± 8 for the ET group ($p = 1.00$), and initial $VO_2$max for ET and HIIT groups were 41.4 ± 5.5 and 41.9 ± 5.8 ($p = 0.87$), respectively. There were no significant differences in baseline values between groups for any of the variables measured in this study. Table 2 provides average characteristics for each group.

*Training*

The average Vmax used to generate exercise protocols at the start of the study was 7.5 ± 0.5 mph for the HIIT group and 7.4 ± 1.2 mph for the ET group ($p = 0.88$). The total duration of exercise for both groups was matched, lasting 30 min per day, 3 days per week for 8 weeks. Both groups also exercised at the same average relative intensity on each day, which included a 3 min warm-up and 3 min cool-down at 60% Vmax. The average workload of training was 1075 ± 181 kcal for weeks 1-2, 1139 ± 191 kcal for weeks 3-4, 1215 ± 185 kcal for weeks 5-6, and 1282 ± 195 kcal for weeks 7-8.

The $VO_2$max test and Vmax estimation were repeated after 4 weeks of training in order to ensure the training speeds corresponded to the participants' current fitness, as $VO_2$max adaptations to a particular intensity result in a plateau after 3 weeks, with a half-time of about 11 days.[131] The ET group ran at a constant pace for the duration of each training day, with the intensity set to 70%

of Vmax for weeks 1-2, 75% of Vmax for weeks 3-6, and 80% of Vmax for weeks 7-8. The HIIT group completed 12 intervals, at a ratio of 1 min work to 1 min active recovery. During weeks 1-2, the intervals were 1 min at 90% of Vmax and 1 min at 50% of Vmax (average = 70% of Vmax). For weeks 3-6, the intensity increased to 1 min at 100% of Vmax and 1 min at 50% of Vmax (average = 75% of Vmax), and for weeks 7-8 the intensity again increased to 1 min at 110% Vmax and 1 min at 50% of Vmax (average = 80% of Vmax). All training sessions were completed on laboratory treadmills (Woodway Desmo, Woodway USA, Waukesha, WI) programmed with each participant's training protocol and supervised by a researcher trained in adult CPR/AED. See tables 3 and 4 for individual and group training protocol details.

*Study Design and Statistical Analysis*

This study employed a within-individual and between-groups design using matched groups, based on initial HDL concentration and $VO_2$max. If the baseline values were normally distributed, then t-tests were used to determine if there were any significant initial differences between the two groups. For nonparametric baseline values, a Wilcoxon-Mann-Whitney test was used to evaluate any potential baseline differences. Normality was determined by the Shapiro-Wilk normality test. Two-way repeated-measures ANOVAs were used to examine main effects for training and group on differences in adaptations for data that did not violate assumptions of normality. For nonparametric data, the Wilcoxon sign rank test was used for within-subjects analysis, and the Friedman ranked ANOVA for nonparametric data was used for between-groups analysis. Outliers were included in the analysis unless they were greater than $\pm$ 3 standard deviations away from the mean and exhibited undue influence on the results. Significance was set *a priori* to $\alpha = 0.05$. SAS 9.3 (SAS Institute Inc., Cary, NC) was used to complete all statistical analyses in this study.

# RESULTS

*Participants*

Fourteen participants were recruited and began the study, but one participant in the ET group withdrew from the study due to an ankle injury unrelated to activities involved in the study. In order to maintain a balanced study design, data from the participant with the most similar baseline values in the HIIT group was not analyzed or included in the results. Thus, 12 participants completed the study and were included in the data analysis.

*Training*

All 12 participants completed the 8 weeks of exercise training in the study with an adherence rate of 100%. There were no adverse events during any of the training sessions, and no injuries were incurred as a direct result of participating in the study. Individual and average training velocities for each participant and each group are presented in Table 3 for the HIIT group and Table 4 for the ET group.

*HDL function*

There were two outliers greater than ± 3 standard deviations in the data for HDL function, one from each group, which had inappropriate influence and resulted in a nonparametric distribution of the data. Therefore, the outliers were removed from the analysis, bringing the data back to a normal distribution and leaving 5 results for each group. In the HIIT group, the average slope of the oxidation of DHR with the addition of HDL before training was 45.40 ± 20.06 FU/min. After training, the oxidation slope was 43.66 ± 9.45 FU/min. The change in HDL function did not reach significance (3.8%, p=0.83). In the ET group, the slope of oxidation was

decreased from 50.23 ± 23.11 to 30.14 ± 19.76 FU/min, indicating a 40% increase in HDL function. However, this change in HDL function did not reach significance either (p=0.21) most likely due to dichotomous responses among the participants. The three participants with initially lower HDL function improved, but the two participants with initially greater HDL function regressed. The change in HDL function between the groups was also not significantly different (p=0.27).

*Inflammation*

Data for CRP and IL-6 concentrations violated assumptions of normality and were thus analyzed using the Wilcoxon sign rank test and the Friedman ranked ANOVA for nonparametric data. CRP concentrations were not significantly altered by either HIIT (p=0.84) or ET (p=0.84), and there was no significance in the changes between groups (p=0.88). Interestingly, the plasma IL-6 concentration was not significantly elevated immediately after exercise for either running protocol on either the first (HIIT p=0.84; ET p=0.84) or the last exercise day (HIIT p=0.44; ET p=0.69). The IL-6 response to exercise did not change after the 8 weeks of training (HIIT p=0.22; ET p=0.84), and was not significantly different between groups (p=0.45).

*Lipid profile*

Despite increases in HDL concentration in both groups (HIIT = 34 ± 9 to 40 ± 9, 16%; ET = 34 ± 8 to 41 ± 10, 19.0%), HDL concentration was not significantly increased in either group (HIIT p=0.33; ET p=0.08) and was not significantly different between groups (p=0.84). TC was not significantly different for either group (HIIT p=0.73; ET p=0.0.62; between-groups p=0.99). However, the TC:HDL ratio was significantly lowered by ET, from 4.67 ± 0.88 to 4.07 ± 0.96 (p=0.02). The reduction from 4.37 ± 1.39 to 3.85 ± 1.29 in TC:HDL in the HIIT group did not reach significance (p=0.30). The change in TC:HDL ratio was not significantly different between groups (p=0.87). The change in calculated LDL was not significant for either group (HIIT

p=0.65; ET p=0.75) and was not significant between groups (p=0.57). Plasma TRG concentration was significantly reduced by HIIT, dropping from 92 ± 32 to 61 ± 12 mg/dL (p=0.04). In the ET group, there was an outlier in the results for TRG concentration, greater than ± 3 standard deviations from the mean and causing a nonparametric distribution of the data. Therefore the pre- and post-training TRG results for that participant were excluded from the analysis. TRG concentration was not significantly different following ET, increasing from 87 ± 36 to 97 ± 43 mg/dL (p=0.13). The change in TRG concentration was significantly greater in the HIIT group than the ET group (p=0.009). The change in Glu concentration was not significantly different in either group (HIIT p=0.91; ET p=.029) and was not significantly different between groups (p=0.39).

*Cardiovascular fitness*

For the HIIT group, $VO_2$max increased by 6.6%, from 41.4 ± 5.5 to 44.1 ± 6.8 ml/kg/min but failed to reach significance from initial $VO_2$max (p=0.13). In the ET group, $VO_2$max also was not significantly different, increasing by 3.9%, from 41.9 ± 5.8 to 43.5 ± 7.2 ml/kg/min (p=0.31). There was no significant difference in improvement between the HIIT and ET groups (p=0.62). The Vmax after 8 weeks of HIIT was significantly different from baseline (p=0.02), increasing from 7.5 ± 0.5 to 8.1 ± 0.8 mph (8.5%), but the increase in the ET was not significant (p=0.26), increasing from 7.4 ± 1.2 to 7.7 ± 1.4 mph (4.5%). The increase in Vmax was not significantly different between groups (p=0.38). As a result of training, HRmax was significantly reduced in the HIIT group, from 198 ± 11 to 193 ± 12 bpm (p=0.01), but not in the ET group (p=0.41), with no significant difference between groups (p=0.27).

*Body composition*

After the 8 weeks of exercise training, the ET group had significantly reduced the percentage of android fat from 36.78 ± 9.60 to 34.18 ± 11.39% (p=0.046). There was no

significant difference seen in the HIIT group ($p=0.24$) and no significant difference between the groups ($p=0.67$). There were no significant decreases in weight, percent fat, or gynoid fat within- or between-groups. For a complete overview of the results for the HIIT group, ET group, and group comparisons see Tables 5, 6, and 7.

# DISCUSSION

The primary goal of this study was to examine the potential differences in the extent of HDL function improvement after 8 weeks of high-intensity interval training or traditional endurance training that was matched for average intensity, workload, and duration. As chronic inflammation, $VO_2$max, the lipid profile, and body composition are potentially correlated with HDL function, these factors were included to evaluate their potential contribution to any changes in HDL function. The main strengths of this study were matched average intensity, workload, and duration exercise protocols and equally matched groups based on HDL concentration ($p=1.00$) and $VO_2$max ($p=0.87$). The results of this study indicate that there was no significant change in HDL function in the HIIT group or in the ET group over the 8 week of exercise training, and the HDL function response to training was not significantly different between groups. Chronic CRP and acute IL-6 responses to exercise were no different after training as well, again with no differences between the groups. There were also no significant changes in $VO_2$max as a result of 8 weeks of either training protocol, with no differences between the two groups. The HIIT group was able to significantly increase their Vmax and significantly decrease their HRmax, but with no significant effect between groups. HIIT did result in a significantly greater reduction in fasting TRG concentration after the training period compared to traditional ET. The ET group significantly decreased the percentage of android fat and the TC:HDL ratio, however again with no difference between groups.

*HDL function*

The results of this study indicate that HDL function was not improved by 8 weeks of exercise training. However, previous research has shown this adaptation to be possible. Roberts *et al.* witnessed improvements in HDL function after only 3 weeks of training and dietary intervention in patients at risk for metabolic syndrome, but their participants exercised daily for 3 weeks for 45-60 min per session at an intensity of 70-85% of HRmax.[25] So while the intensity was similar to that completed in this study, the weekly volume was much greater. Also, most of the participants in the Roberts *et al.* study began in a diseased state, and their exercise was accompanied by a strict dietary intervention, neither of which was present in this research effort. Iborra *et al.* also demonstrated increased HDL function after exercise training at similar intensities to this study, but there were significant differences between the two as well, namely, an 18 week training period and middle-aged participants with diabetes mellitus and low initial $VO_2$max (17.8 ml/kg/min), compared to the young, healthy participants with only below-average $VO_2$max (41.6 ml/kg/min) in our study.[87]

The potential discrepancy between our results and past research could be due to the lack of an increase in $VO_2$max and decrease in CRP in this study. In the studies just mentioned, increases in HDL function were accompanied by increases in VO2max and decreases in CRP,[25, 87] and research has indicated a strong association between increases in physical fitness, decreases in CRP, and increases in HDL function.[19, 22, 54, 86] Thus, the lack of any increase in HDL function in this study could be partially explained by the absence of adaptations in $VO_2$max and CRP.

*Inflammation*

Exercise physiology literature has indicated that chronic inflammation is inversely associated with physical fitness, with athletes exhibiting significantly lower levels of CRP than their sedentary counterparts.[20] In fact only 200-599 kcal of exercise per week is sufficient to reduce

CRP, with further reductions when energy expenditure is increased to 600-1499 kcal/week.[22] This adaptation is thought to be the result of repeated inflammatory challenges arising from regular exercise, which can be seen by the acute phase response of plasma IL-6.[125] This increase in IL-6 is intensity dependent,[31] but chronic exercise training can result in a blunted response, despite increases in the absolute workload of an exercise session.[27, 132] This adaptive response may be a result of increased glycogen storage and a reduced reliance on carbohydrate metabolism after HIIT.[111] IL-6 is significantly increased when carbohydrate metabolism is limited by depleted carbohydrate stores,[30, 69] and HIIT relies more heavily on carbohydrate oxidation than ET even when workloads are matched.[133]

The results from this study did not show any decrease in resting CRP concentrations, despite weekly energy expenditure ranging from 1075 kcal/week in weeks 1-2 to 1282 kcal/week in weeks 7-8. While this result is surprising, it is potentially explained by the lack of a significant acute IL-6 response in either group in response to exercise, both on the first and last training days of the study. This was also unexpected, as acute increases in IL-6 after exercise are common in the literature.[68-72] Nielsen *et al.* reported a 2-fold increase in systemic IL-6 after just 6 min of maximal rowing exercise,[71] and participants in our HIIT group spent 12 min near or above Vmax during each exercise session. However, the difference in response may be due to a lack of glycogen depletion and carbohydrate supply-demand mismatch in the exercise protocols employed in this study, consisting of only 1 min work intervals at 100% of Vmax or just 24 min of running at a moderate intensity (70-80% Vmax). This carbohydrate supply-demand mismatch is at least partially responsible for increases in IL-6 during exercise.[69] Studies that found significant acute IL-6 responses to exercise employed activities that result in severe glycogen depletion and carbohydrate supply being a limiting factor,[134, 135] whether it be 6 min of maximal effort[71] or moderate-intensity exercise lasting greater than one hour.[136-138]

The interplay between HIIT interval intensity and duration is complicated,[113, 139] and may be more so when the goal is to reduce resting CRP via acute inflammatory challenges that likely arise from glycogen depletion. When the intensity of exercise is normalized, the IL-6 response to increased exercise duration is exponential, with small increases in time resulting in large increases in IL-6.[140] The results of this study, using 1 min work intervals at 100% Vmax, were in agreement with Hovanloo *et al.*, who did not see any IL-6 or CRP adaptations following a training period using 30 sec "all-out" intervals.[141] However, when the high intensity intervals consist of 1000m maximal effort runs or last 3 min at 90% of $VO_2$max, significant acute IL-6 responses occur, resulting in reductions in IL-6 response to exercise after a period of exercise training.[132, 142] If acute increases in IL-6 are driven by the inhibition of carbohydrate utilization that accompanies glycogen depletion, then seeking a HIIT protocol that results in the greatest glycogen reduction would be beneficial. Several different HIIT protocols significantly reduce muscle glycogen content, including 30 sec Wingate sprints,[143] 1 min intervals at 130% Vmax,[144] and 5 min intervals at 85% of VO2max.[145] The nature of these three studies do not lend themselves to easy comparisons due to the different training statuses of the subject populations and the implication of reduced glycogen use in more highly trained individuals.[146] However, with adjustments made for potential confounding factors, the greatest depletion of glycogen appears to occur in more prolonged, 5 min intervals. This would be in agreement with the exponential increase in IL-6 response with increasing duration and the significant IL-6 response after 6 min of maximal exercise.[71, 140] Based on the limited evidence presented, it seems logical to conclude that longer high-intensity intervals may result in greater glycogen reduction, a greater acute IL-6 response, and thus greater reductions in chronic inflammation. However, this conclusion is merely speculative; much more research is needed specifically addressing this relationship before any definitive conclusion can be reached.

*Lipid profile*

While the concentration of HDL was increased in both the HIIT and ET groups (16% and 19%, respectively), this increase did not reach significance in either group (HIIT p=0.33; ET p=0.08). Our results were very similar to the results seen by Musa *et al.*, who had participants complete an extremely similar training period: 8 weeks of HIIT 3 days per week, with interval intensities of 90% of $VO_2$max, a 1:1 work to rest ratio, and an energy expenditure of 1269 kcal/week.[118] They found an 18% increase in HDL concentration, which was significant from baseline values likely due to their larger sample size (20 participants completing HIIT compared to our 6 participants). Farrell and Barboriak also found significant increases in HDL concentration (7%) with a larger sample size (16 participants) with 8 weeks of exercise, training 3-4 days per week at an average intensity of 70% of $VO_2$max.[93] Measuring HDL every week, they found a decrease in HDL concentration over the first 2 weeks, followed by a linear 1 mg/dL/week increase in HDL over the next 6 weeks. While our protocol only involved measuring HDL concentration before and after the 8 weeks of training, our results appear to agree with this pattern, with participants averaging a 6 mg/dL increase over the 8 weeks. With such similar results, it seems that the increases in our study would reach significance if a larger sample had been used.

The TC:HDL ratio was significantly reduced only in the ET group in this study (p=0.02), with the reduction failing to reach significance in the HIIT group (p=0.30). These results are similar to those seen by Nybo *et al.*, in which neither the HIIT nor ET group significantly altered HDL or TC, but the ET group significantly reduced the TC:HDL ratio.[99] However, in the Nybo *et al.* study, the reduction in TC:HDL could be explained by greater reductions in TC in the ET group compared to the HIIT group, which was not the case in our study. In our study, the significant reduction in TC:HDL in the ET group seems to be due to a slightly greater increase in HDL and no difference in TC compared to the HIIT group. In fact, neither of our groups saw a

significant change in TC as a result of training (HIIT p=0.73; ET p=0.62). It is worth mentioning that the Nybo *et al.* results cannot be directly compared to our study, due to distinct differences in their training protocols, such as mismatched workloads between their HIIT and ET groups. Regardless, it does appear that ET is more beneficial than HIIT for improving the TC:HDL ratio.

Plasma concentration of TRG is an independent risk factor for future cardiovascular incidents.[147] The response of plasma TRG to exercise training is varied, with factors such as initial TRG concentration and initial HDL concentration possibly playing a role in the response.[148] The response of plasma TRG to HIIT is also varied, with some finding significantly reduced TRG concentrations,[41] reductions that were not significant,[32, 149] and insignificant increases.[40] However, the results of our study indicate significant reductions in plasma concentrations of TRG (p=0.04) that were significantly greater than that of ET (p=0.009). This is especially interesting since ET resulted in significant reductions in the percentage of android fat (p=0.045), and both groups saw decreases in percent fat, though insignificant (HIIT = -3.3%, p=.21; ET = -5.5%, p=0.10). Therefore, it seems that when HDL concentrations and percent fat are standardized between groups, HIIT is significantly more beneficial for reducing plasma TRG concentrations than ET, even in normotriglyceridemic men. This lowering of plasma TRG could be indicative of increased lipoprotein lipase (LPL) enzyme activity, as the two are inversely associated.[57] LPL not only plays a role in the breakdown of triglycerides, but is also directly associated with HDL concentration by being involved in the maturation of HDL molecules by adding additional apoA-1 subunits to HDL.[12, 56] This enzyme is secreted from metabolically active tissue and inhibited by increased inflammation, and while we did not witness decreases in inflammation, the exercise training certainly could have resulted in increased LPL release. These ideas are somewhat confounded, however, as the ET group showed increased HDL concentrations along with increased TRG, though neither were significant.

*Cardiovascular fitness*

The most surprising result from this study was the lack of a significant increase in $VO_2$max as a result of 8 weeks of aerobic endurance training. Increases in $VO_2$max are almost ubiquitously seen in training studies throughout the literature, with studies demonstrating HIIT to be superior to ET for generating this adaptation.[32, 37, 44, 99] Increases in $VO_2$max are seen in as little as 2 weeks in some studies, with other studies showing a 0.63% increase in $VO_2$max per HIIT session.[37, 109] A study employing the same amount of time at the same intensity using either short (15 sec) intervals or long (4 min) intervals, with the same frequency of training over the same time period as this research effort resulted in a significant 8% increase in $VO_2$max.[32] However, in this study, there were 2 participants in each group who either did not increase $VO_2$max or decreased slightly over the course of the study, despite 100% adherence to the exercise sessions over the 8 weeks of training. If these participants could be removed from the analysis, the average increase in $VO_2$max would reach significance in the HIIT group (11% increase as opposed to 6.6%), despite the small sample size. However, the participants cannot be excluded, as this is a well-documented phenomenon among the population.[150] "Non-responders" or "low-responders," as they are often called, made up between 15-20% of participants in the large HERITAGE Family Study, showing less than a 200 ml/min increase in $VO_2$max after 20 weeks of endurance training.[151] The ability to increase $VO_2$max as a result of exercise training is a genetically inherited trait, with genetics accounting for 47% of the variability and resulting in significant variations in response in members of separate families.[151] While there were no participants in this study that were members of the same family, and having 33% of participants in the study be non-responders is unusually high, it is not an impossible scenario.

The Vmax was significantly increased after 8 weeks of HIIT, but not ET (HIIT = 8.5%, $p=0.02$; ET = 4.5%, $p=0.26$), with no significant difference between the two ($p=0.38$). This

adaptation was not unexpected, as many studies have shown greater increases in Vmax, cycling power, time to exhaustion, and time trial performance to be greater after HIIT.[44, 100, 101, 105, 152] The performance increases are most likely a product of the specificity of high-speed training when completing HIIT.[139]

Aerobic endurance training often results in a reduction in HRmax due to increases in ventricular volume and stroke volume. It has been documented that HIIT can result in this type of significant cardiovascular remodeling, increasing stroke volume and maximal cardiac output to a greater extent than ET.[32, 35, 36, 40, 41] Therefore, although stroke volume and ventricular remodeling were not measured, it is understandable why the HIIT group would significantly reduce HRmax over the course of this study ($p=0.01$), while no significant decrease would be present in the ET group ($p=0.41$).

# CONCLUSIONS

These high-intensity interval training and traditional endurance training protocols are not sufficient for significantly improving HDL function or concentration. A larger sample size, greater length of training, or longer HIIT intervals may be necessary to reduce chronic inflammation and increase HDL function. HIIT is significantly better than ET for reducing plasma triglyceride concentrations over a short, 8 week training period. Also only HIIT was able to increase Vmax and reduce HRmax over the 8 week training period, while ET alone resulted in decreases in TC:HDL ratio and percentage of android fat. As HIIT and ET are both beneficial for cardiovascular health in different respects, it may be advantageous to include both training styles to maximize the positive benefits of exercise training.

REFERENCES


1. Boden, W.E., *High-density lipoprotein cholesterol as an independent risk factor in cardiovascular disease: Assessing the data from Framingham to the Veterans Affairs high-density lipoprotein intervention trial.* American Journal of Cardiology, 2000. **86**(12A): p. 19l-22l.
2. Ridker, P.M., et al., *C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women.* New England Journal of Medicine, 2000. **342**(12): p. 836-843.
12. Lewis, G.F. and D.J. Rader, *New insights into the regulation of HDL metabolism and reverse cholesterol transport.* Circulation, 2005. **96**(12): p. 1221-32.
15. Navab, M., et al., *High-density lipoprotein: antioxidant and anti-inflammatory properties.* Current Atherosclerosis Reports, 2007. **9**(3): p. 244-8.
16. Ansell, B.J., et al., *Inflammatory/antiinflammatory properties of high-density lipoprotein distinguish patients from control subjects better than high-density lipoprotein cholesterol levels and are favorably affected by simvastatin treatment.* Circulation, 2003. **108**(22): p. 2751-2756.
19. Mattusch, F., et al., *Reduction of the plasma concentration of C-reactive protein following nine months of endurance training.* International Journal of Sports Medicine, 2000. **21**(1): p. 21-24.
20. Aronson, D., et al., *C-Reactive protein is inversely related to physical fitness in middle-aged subjects.* Atherosclerosis, 2004. **176**(1): p. 173-9.
21. Wannamethee, S.G., et al., *Physical activity and hemostatic and inflammatory variables in elderly men.* Circulation, 2002. **105**(15): p. 1785-1790.
22. Mora, S., et al., *Physical activity and reduced risk of cardiovascular events - Potential mediating mechanisms.* Circulation, 2007. **116**(19): p. 2110-2118.
23. Durstine, J.L., et al., *Blood lipid and lipoprotein adaptations to exercise - A quantitative analysis.* Sports Medicine, 2001. **31**(15): p. 1033-1062.
25. Roberts, C.K., et al., *Effect of a short-term diet and exercise intervention on oxidative stress, inflammation, MMP-9, and monocyte chemotactic activity in men with metabolic syndrome factors.* Journal of Applied Physiology, 2006. **100**(5): p. 1657-65.
27. Fischer, C.P., et al., *Endurance training reduces the contraction-induced interleukin-6 mRNA expression in human skeletal muscle.* American Journal of Physiology-Endocrinology and Metabolism, 2004. **287**(6): p. E1189-E1194.
28. Hellsten, Y., F.S. Apple, and B. Sjodin, *Effect of sprint cycle training on activities of antioxidant enzymes in human skeletal muscle.* Journal of Applied Physiology, 1996. **81**(4): p. 1484-1487.
29. Tyldum, G.A., et al., *Endothelial Dysfunction Induced by Post-Prandial Lipemia Complete Protection Afforded by High-Intensity Aerobic Interval Exercise.* Journal of the American College of Cardiology, 2009. **53**(2): p. 200-206.
30. Febbraio, M.A. and B.K. Pedersen, *Muscle-derived interleukin-6: mechanisms for activation and possible biological roles.* Faseb Journal, 2002. **16**(11): p. 1335-1347.
31. Ostrowski, K., P. Schjerling, and B.K. Pedersen, *Physical activity and plasma interleukin-6 in humans - effect of intensity of exercise.* European Journal of Applied Physiology, 2000. **83**(6): p. 512-515.

32. Helgerud, J., et al., *Aerobic high-intensity intervals improve VO2max more than moderate training.* Medicine and Science in Sports and Exercise, 2007. **39**(4): p. 665-671.
35. Moholdt, T.T., et al., *Aerobic interval training versus continuous moderate exercise after coronary artery bypass surgery: A randomized study of cardiovascular effects and quality of life.* American Heart Journal, 2009. **158**(6): p. 1031-1037.
36. Munk, P.S., et al., *High-intensity interval training may reduce in-stent restenosis following percutaneous coronary intervention with stent implantation: A randomized controlled trial evaluating the relationship to endothelial function and inflammation.* American Heart Journal, 2009. **158**(5): p. 734-741.
37. Rognmo, O., et al., *High intensity aerobic interval exercise is superior to moderate intensity exercise for increasing aerobic capacity in patients with coronary artery disease.* European Journal of Cardiovascular Prevention & Rehabilitation, 2004. **11**(3): p. 216-222.
40. Tjonna, A.E., et al., *Aerobic interval training versus continuous moderate exercise as a treatment for the metabolic syndrome - A pilot study.* Circulation, 2008. **118**(4): p. 346-354.
41. Wisloff, U., et al., *Superior cardiovascular effect of aerobic interval training versus moderate continuous training in heart failure patients - A randomized study.* Circulation, 2007. **115**(24): p. 3086-3094.
44. Daussin, F.N., et al., *Effect of interval versus continuous training on cardiorespiratory and mitochondrial functions: relationship to aerobic performance improvements in sedentary subjects.* American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 2008. **295**(1): p. R264-R272.
46. Honda, H., et al., *Oxidized high-density lipoprotein as a risk factor for cardiovascular events in prevalent hemodialysis patients.* Atherosclerosis, 2012. **220**(2): p. 493-501.
48. VanLenten, B.J., et al., *Anti-inflammatory HDL becomes pro-inflammatory during the acute phase response - Loss of protective effect of HDL against LDL oxidation in aortic wall cell cocultures.* Journal of Clinical Investigation, 1995. **96**(6): p. 2758-2767.
49. Feingold, K.R., et al., *Paraoxonase activity in the serum and hepatic mRNA levels decrease during the acute phase response.* Atherosclerosis, 1998. **139**(2): p. 307-315.
50. Lindhorst, E., et al., *Acute inflammation, acute phase serum amyloid A and cholesterol metabolism in the mouse.* Biochimica et Biophysica Acta, 1997. **1339**(1): p. 143-54.
52. Ridker, P.M., *On evolutionary biology, inflammation, infection, and the causes of atherosclerosis.* Circulation, 2002. **105**(1): p. 2-4.
54. Navab, K.D., et al., *Chronic Inflammatory Disorders and Accelerated Atherosclerosis: Chronic Kidney Disease.* Current Pharmaceutical Design, 2011. **17**(1): p. 17-20.
55. Vaziri, N.D., M. Navab, and A.M. Fogelman, *HDL metabolism and activity in chronic kidney disease.* Nature Reviews Nephrology, 2010. **6**(5): p. 287-296.
56. Broedl, U.C., W. Jin, and D.J. Rader, *Endothelial lipase: a modulator of lipoprotein metabolism upregulated by inflammation.* Trends in Cardiovascular Medicine, 2004. **14**(5): p. 202-6.
57. Blades, B., G.L. Vega, and S.M. Grundy, *Activities of Lipoprotein-Lipase and Hepatic Triglyceride Lipase in Postheparin Plasma of Patients with Low Concentrations of Hdl Cholesterol.* Arteriosclerosis and Thrombosis, 1993. **13**(8): p. 1227-1235.
62. Kelesidis, T., et al., *A biochemical fluorometric method for assessing the oxidative properties of HDL.* Journal of Lipid Research, 2011. **52**(12): p. 2341-2351.
68. Pedersen, B.K., et al., *Muscle-derived interleukin-6: lipolytic, anti-inflammatory and immune regulatory effects.* Pflugers Archiv-European Journal of Physiology, 2003. **446**(1): p. 9-16.

69. Starkie, R.L., et al., *Carbohydrate ingestion attenuates the increase in plasma interleukin-6, but not skeletal muscle interleukin-6 mRNA, during exercise in humans.* Journal of Physiology-London, 2001. **533**(2): p. 585-591.
70. Nieman, D.C., et al., *Influence of mode and carbohydrate on the cytokine response to heavy exertion.* Medicine and Science in Sports and Exercise, 1998. **30**(5): p. 671-678.
71. Nielsen, H.B., et al., *Lymphocytes and NK cell activity during repeated bouts of maximal exercise.* American Journal of Physiology, 1996. **271**(1 Pt 2): p. R222-7.
72. Ostrowski, K., et al., *Chemokines are elevated in plasma after strenuous exercise in humans.* European Journal of Applied Physiology, 2001. **84**(3): p. 244-245.
81. van Oostrom, A.J.H.H.M., et al., *Postprandial recruitment of neutrophils may contribute to endothelial dysfunction.* Journal of Lipid Research, 2003. **44**(3): p. 576-583.
86. Olchawa, B., et al., *Physical fitness and reverse cholesterol transport.* Arteriosclerosis Thrombosis and Vascular Biology, 2004. **24**(6): p. 1087-1091.
87. Iborra, R.T., et al., *Aerobic exercise training improves the role of high-density lipoprotein antioxidant and reduces plasma lipid peroxidation in type 2 diabetes mellitus.* Scandinavian Journal of Medicine & Science in Sports, 2008. **18**(6): p. 742-750.
93. Farrell, P.A. and J. Barboriak, *The Time Course of Alterations in Plasma-Lipid and Lipoprotein Concentrations during 8 Weeks of Endurance Training.* Atherosclerosis, 1980. **37**(2): p. 231-238.
99. Nybo, L., et al., *High-Intensity Training versus Traditional Exercise Interventions for Promoting Health.* Medicine and Science in Sports and Exercise, 2010. **42**(10): p. 1951-1958.
100. Billat, L.V., *Interval training for performance: A scientific and empirical practice - Special recommendations for middle- and long-distance running. Part II: Anaerobic interval training.* Sports Medicine, 2001. **31**(2): p. 75-90.
101. Laursen, P.B. and D.G. Jenkins, *The scientific basis for high-intensity interval training - Optimising training programmes and maximising performance in highly trained endurance athletes.* Sports Medicine, 2002. **32**(1): p. 53-73.
105. Esfarjani, F. and P.B. Laursen, *Manipulating high-intensity interval training: Effects on VO2max, the lactate threshold and 3000 m running performance in moderately trained mates.* Journal of Science and Medicine in Sport, 2007. **10**(1): p. 27-35.
107. Gibala, M.J. and S.L. Mcgee, *Metabolic adaptations to short-term high-intensity interval training: A little pain for a lot of gain?* Exercise and Sport Sciences Reviews, 2008. **36**(2): p. 58-63.
109. Burgomaster, K.A., et al., *Similar metabolic adaptations during exercise after low volume sprint interval and traditional endurance training in humans.* Journal of Physiology, 2008. **586**(1): p. 151-60.
111. Burgomaster, K.A., G.J.F. Heigenhauser, and M.J. Gibala, *Effect of short-term sprint interval training on human skeletal muscle carbohydrate metabolism during exercise and time-trial performance.* Journal of Applied Physiology, 2006. **100**(6): p. 2041-2047.
113. Guiraud, T., et al., *Optimization of high intensity interval exercise in coronary heart disease.* European Journal of Applied Physiology, 2010. **108**(4): p. 733-740.
114. Moholdt, T., et al., *Aerobic interval training increases peak oxygen uptake more than usual care exercise training in myocardial infarction patients: a randomized controlled study.* Clinical Rehabilitation, 2012. **26**(1): p. 33-44.
118. Musa, D.I., et al., *The Effect of a High-Intensity Interval Training Program on High-Density Lipoprotein Cholesterol in Young Men.* Journal of Strength and Conditioning Research, 2009. **23**(2): p. 587-592.

121. Gill, J.M.R., et al., *Effects of prior moderate exercise on postprandial metabolism and vascular function in lean and centrally obese men.* Journal of the American College of Cardiology, 2004. **44**(12): p. 2375-2382.
122. Whaley, M.H., et al., *ACSM's guidelines for exercise testing and prescription*. 7th ed2006, Philadelphia, Pa.: Lippincott Williams & Wilkins. xxi, 366 p.
123. Cullinane, E.M., et al., *Variations in Plasma-Volume Affect Total and Low-Density-Lipoprotein Cholesterol Concentrations during the Menstrual-Cycle.* Metabolism-Clinical and Experimental, 1995. **44**(8): p. 965-971.
124. Demacker, P.N.M., et al., *Influence of Contraceptive Pill and Menstrual-Cycle on Serum-Lipids and High-Density Lipoprotein Cholesterol Concentrations.* British Medical Journal, 1982. **284**(6324): p. 1213-1215.
125. Kasapis, C. and P.D. Thompson, *The effects of physical activity on serum C-reactive protein and inflammatory markers: a systematic review.* Journal of the American College of Cardiology, 2005. **45**(10): p. 1563-9.
126. Goldberg, C.S., A.R. Tall, and S. Krumholz, *Acute Inhibition of Hepatic Lipase and Increase in Plasma-Lipoproteins after Alcohol Intake.* Journal of Lipid Research, 1984. **25**(7): p. 714-720.
127. Crouter, S.E., et al., *Accuracy and reliability of the ParvoMedics TrueOne 2400 and MedGraphics VO2000 metabolic systems.* European Journal of Applied Physiology, 2006. **98**(2): p. 139-51.
128. Macfarlane, D.J. and H.L. Wu, *Inter-unit variability in two ParvoMedics TrueOne 2400 automated metabolic gas analysis systems.* European Journal of Applied Physiology, 2013. **113**(3): p. 753-62.
129. Issa, J.S., et al., *[Precision and accuracy of blood lipid analyses by a portable device (Cholestech-LDX)].* Arquivos Brasileiros de Cardiologia, 1996. **66**(6): p. 339-42.
130. Friedewald, W.T., R.I. Levy, and D.S. Fredrickson, *Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge.* Clinical Chemistry, 1972. **18**(6): p. 499-502.
131. Hickson, R.C., et al., *Time Course of the Adaptive Responses of Aerobic Power and Heart-Rate to Training.* Medicine and Science in Sports and Exercise, 1981. **13**(1): p. 17-20.
132. Croft, L., et al., *High-intensity interval training attenuates the exercise-induced increase in plasma IL-6 in response to acute exercise.* Applied Physiology Nutrition and Metabolism-Physiologie Appliquee Nutrition Et Metabolisme, 2009. **34**(6): p. 1098-1107.
133. Malatesta, D., et al., *Effect of High-Intensity Interval Exercise on Lipid Oxidation during Postexercise Recovery.* Medicine and Science in Sports and Exercise, 2009. **41**(2): p. 364-374.
134. Saltin, B. and J. Karlsson, *Muscle glycogen utilization during work of different intensities*, in *Muscle metabolism during exercise*1971, Springer. p. 289-299.
135. Karlsson, J. and B. Saltin, *Diet, Muscle Glycogen, and Endurance Performance.* Journal of Applied Physiology, 1971. **31**(2): p. 203-&.
136. Sprenger, H., et al., *Enhanced Release of Cytokines, Interleukin-2 Receptors, and Neopterin after Long-Distance Running.* Clinical Immunology and Immunopathology, 1992. **63**(2): p. 188-195.
137. Drenth, J.P.H., et al., *Endurance Run Increases Circulating Il-6 and Il-1ra but down-Regulates Ex-Vivo Tnf-Alpha and Il-1-Beta Production.* Journal of Applied Physiology, 1995. **79**(5): p. 1497-1503.
138. NehlsenCannarella, S.L., et al., *Carbohydrate and the cytokine response to 2.5 h of running.* Journal of Applied Physiology, 1997. **82**(5): p. 1662-1667.

139. Denadai, B.S., et al., *Interval training at 95% and 100% of the velocity at VO2 max: effects on aerobic physiological indexes and running performance.* Applied Physiology, Nutrition, and Metabolism, 2006. **31**(6): p. 737-43.
140. Fischer, C.P., *Interleukin-6 in acute exercise and training: what is the biological relevance?* Exercise Immunology Review, 2006. **12**: p. 6-33.
141. Hovanloo, F., T. Arefirad, and S. Ahmadizad, *Effects of sprint interval and continuous endurance training on serum levels of inflammatory biomarkers.* Journal of Diabetes & Metabolic Disorders, 2013. **12**(1): p. 22.
142. Niess, A.M., et al., *Evaluation of stress responses to interval training at low and moderate altitudes.* Medicine and Science in Sports and Exercise, 2003. **35**(2): p. 263-269.
143. Bogdanis, G.C., et al., *Contribution of phosphocreatine and aerobic metabolism to energy supply during repeated sprint exercise.* Journal of Applied Physiology, 1996. **80**(3): p. 876-884.
144. Krustrup, P., Y. Hellsten, and J. Bangsbo, *Intense interval training enhances human skeletal muscle oxygen uptake in the initial phase of dynamic exercise at high but not at low intensities.* Journal of Physiology-London, 2004. **559**(1): p. 335-345.
145. Stepto, N.K., et al., *Metabolic demands of intense aerobic interval training in competitive cyclists.* Medicine and Science in Sports and Exercise, 2001. **33**(2): p. 303-310.
146. Parra, J., et al., *The distribution of rest periods affects performance and adaptations of energy metabolism induced by high-intensity training in human muscle.* Acta Physiologica Scandinavica, 2000. **169**(2): p. 157-165.
147. Hokanson, J.E. and M.A. Austin, *Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a metaanalysis of population-based prospective studies.* Journal of cardiovascular risk, 1996. **3**(2): p. 213-219.
148. Couillard, C., et al., *Effects of endurance exercise training on plasma HDL cholesterol levels depend on levels of triglycerides - Evidence from men of the health, risk factors, exercise training and genetics (HERITAGE) family study.* Arteriosclerosis Thrombosis and Vascular Biology, 2001. **21**(7): p. 1226-1232.
149. Stensvold, D., et al., *Strength training versus aerobic interval training to modify risk factors of metabolic syndrome.* Journal of Applied Physiology, 2010. **108**(4): p. 804-810.
150. Bouchard, C. and T. Rankinen, *Individual differences in response to regular physical activity.* Medicine and Science in Sports and Exercise, 2001. **33**(6): p. S446-S451.
151. Bouchard, C., et al., *Familial aggregation of VO2max response to exercise training: results from the HERITAGE Family Study.* Journal of Applied Physiology, 1999. **87**(3): p. 1003-1008.
152. Stepto, N.K., et al., *Effects of different interval-training programs on cycling time-trial performance.* Medicine and Science in Sports and Exercise, 1999. **31**(5): p. 736-741.

FIGURES

**Figure 1 –** *Overview of HDL function in reverse cholesterol transport.*

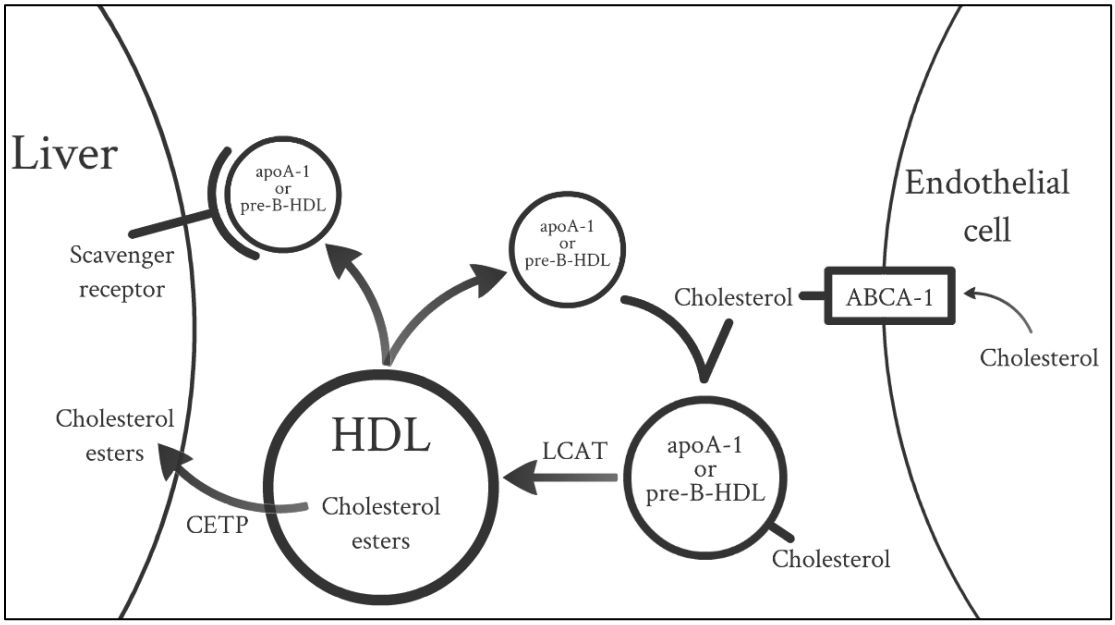


**Figure 2 –** *Individual and average change in HDL function for HIIT group.*

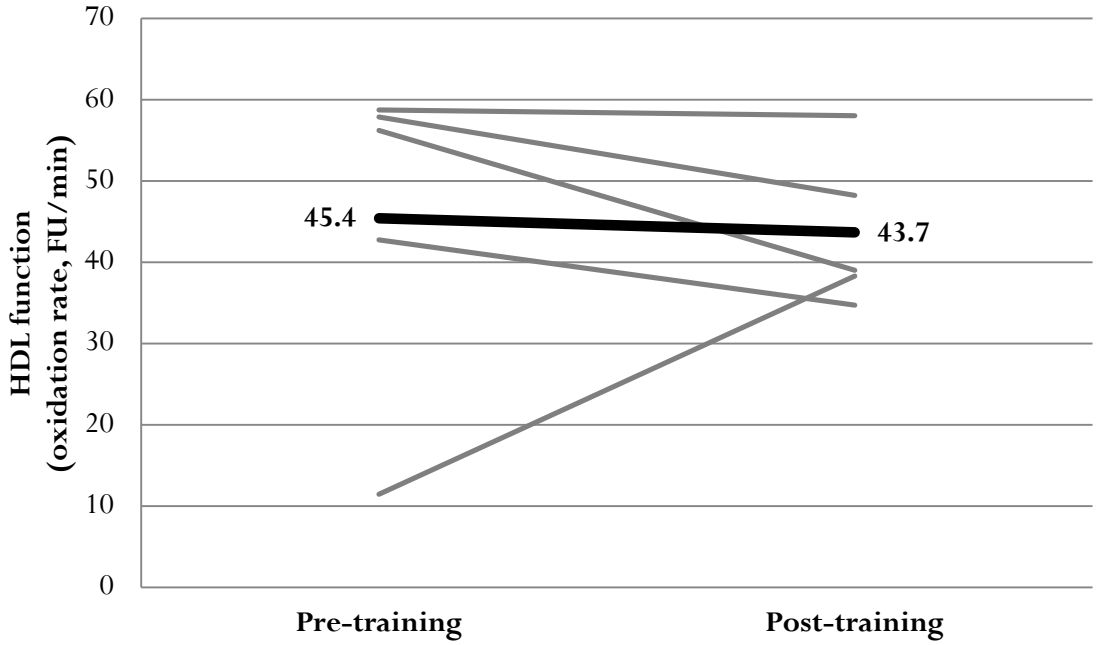


**Figure 3 –** *Individual and average change in HDL function for ET group.*

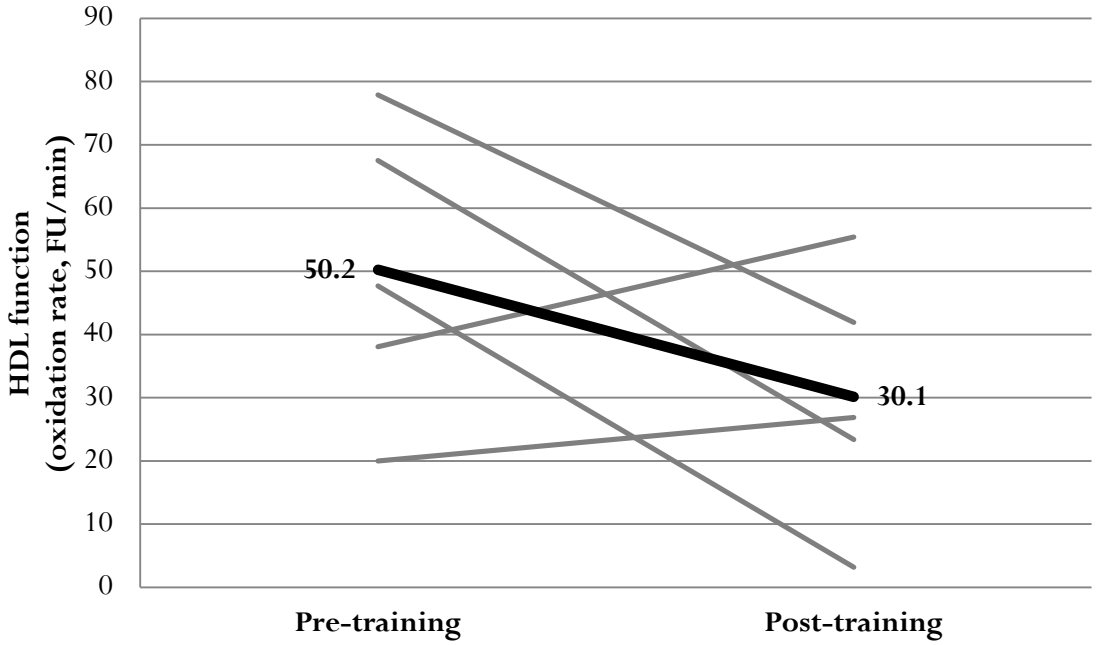


**Figure 4 –** *Individual and average change in [CRP] for HIIT group.*

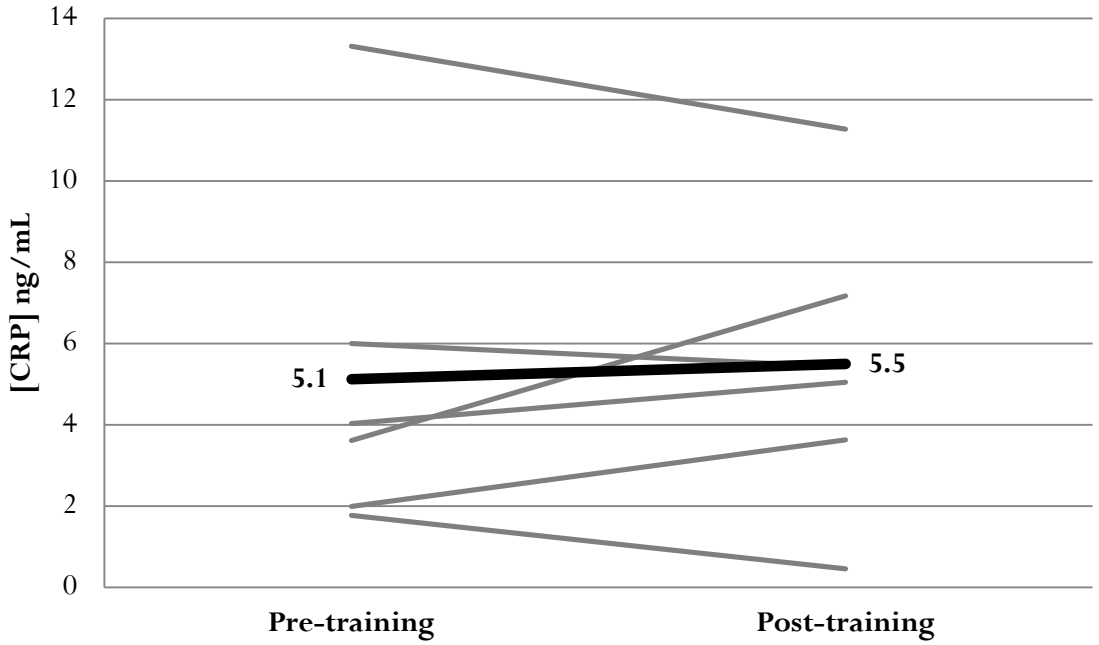


**Figure 5 –** *Individual and average change in [CRP] for ET group.*

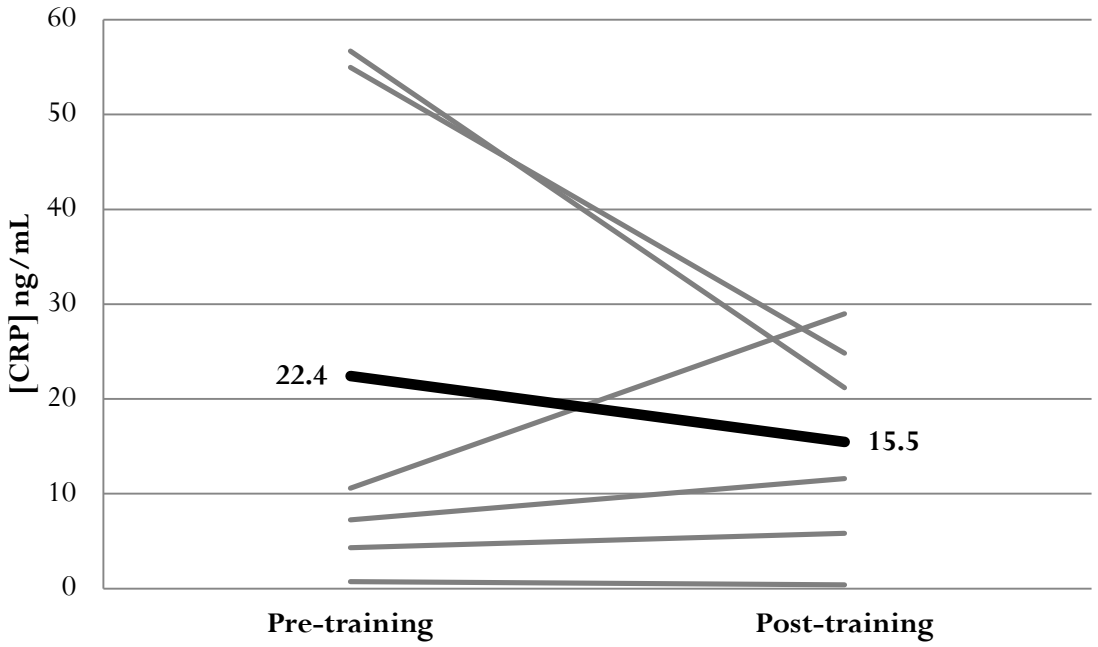


**Figure 6 –** *Individual and average change in [HDL] for HIIT group.*

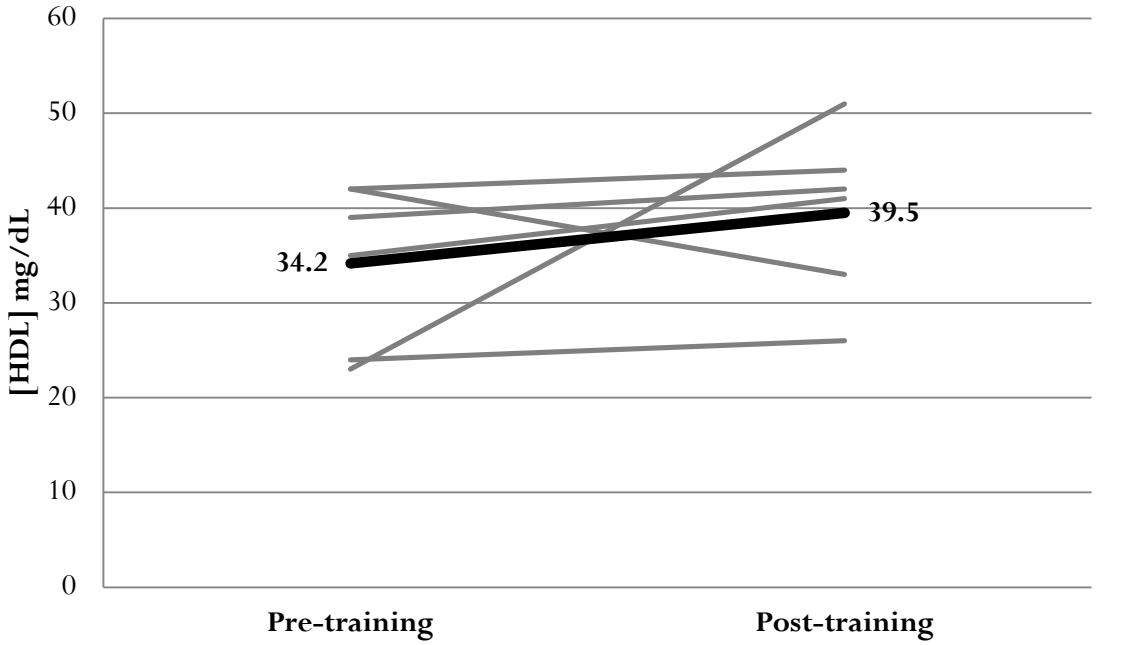


**Figure 7 –** *Individual and average change in [HDL] for ET group.*

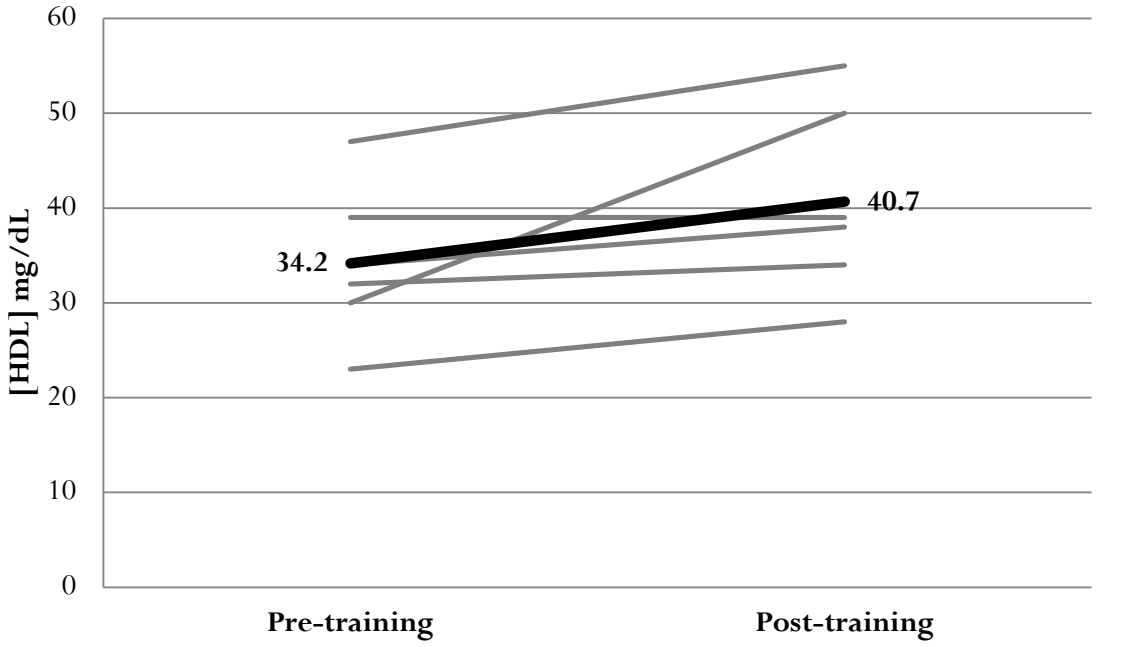


**Figure 8 –** *Individual and average change in $VO_2$max for HIIT group.*

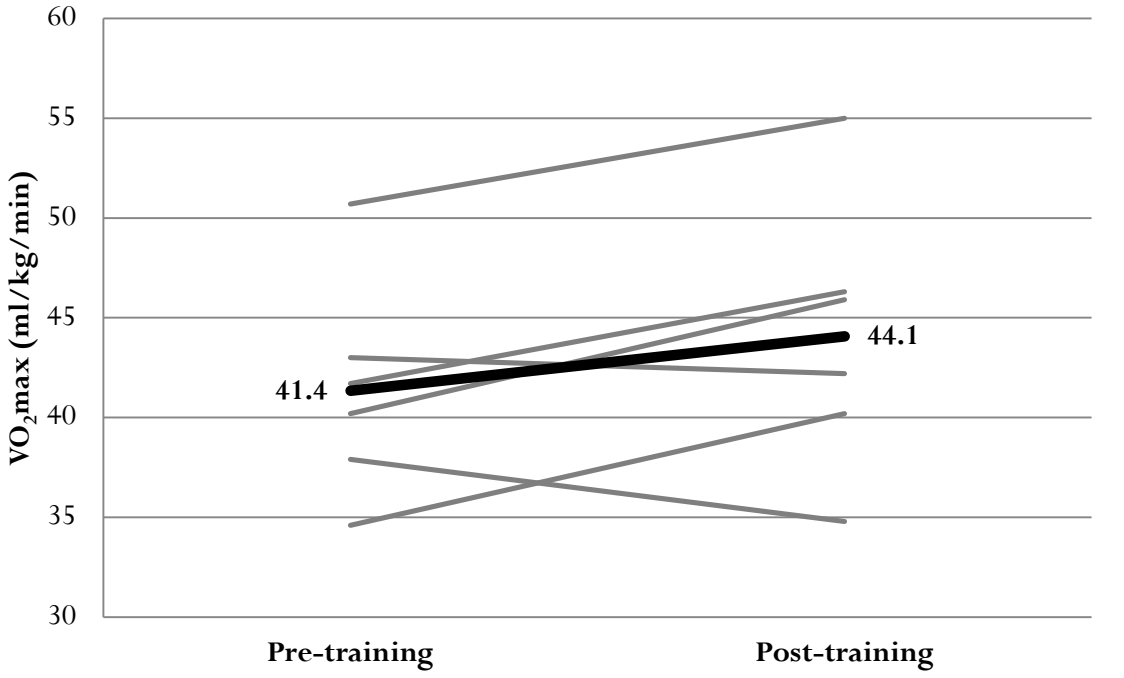


**Figure 9 –** *Individual and average change in $VO_2$max for ET group.*

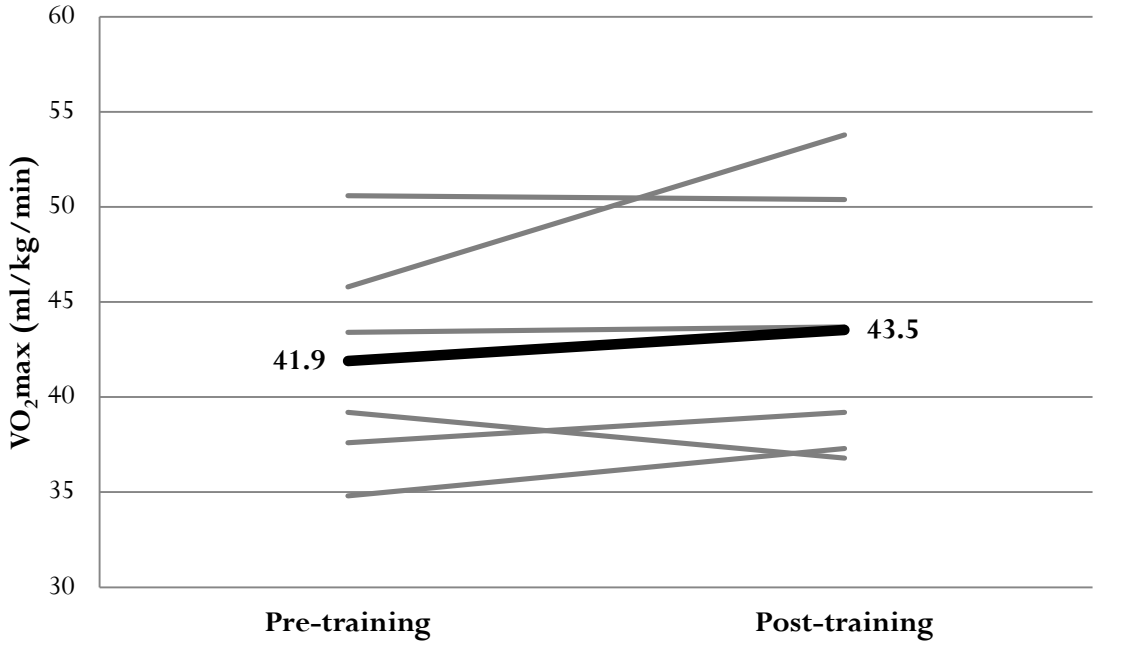
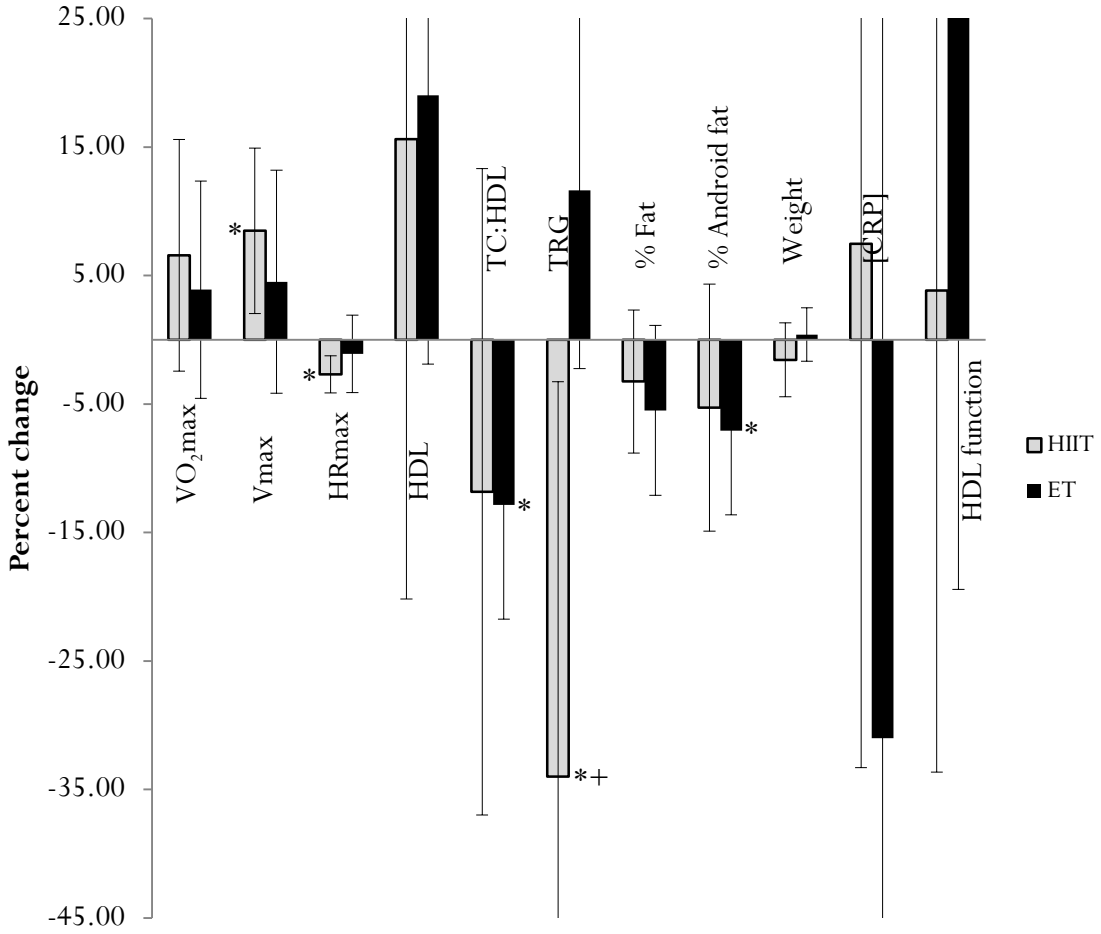


**Figure 10 –** *Percent change of relevant variables.*




*Percent change after 8 weeks of either HIIT or ET. * indicates significantly different from baseline ($p<0.05$). $^{+}$ indicates significant difference between groups ($p<0.05$).*

TABLES

**Table 1** – *Baseline physiological characteristics.*

| Variable | Mean ± SD |
|---|---|
| **Anthropometrics** | |
| *Age (years)* | *21.6 ± 1.6* |
| *Height (m)* | *1.81 ± 0.08* |
| *Weight (kg)* | *86.05 ± 16.30* |
| **Body composition** | |
| *Percent fat (%)* | *28.07 ± 7.17* |
| *Android fat (%)* | *35.88 ± 8.58* |
| *Gynoid fat (%)* | *33.06 ± 6.80* |
| **Cardiovascular characteristics** | |
| *$VO_2$max (ml/kg/min)* | *41.6 ± 5.4* |
| *Vmax (mph)* | *7.4 ± 0.9* |
| *HRmax (bpm)* | *197 ± 9* |
| *Systolic blood pressure (mmHg)* | *118 ± 7* |
| *Diastolic blood pressure (mmHg)* | *73 ± 8* |
| **Lipid profile** | |
| *Total cholesterol (mg/dl)* | *147 ± 20* |
| *HDL concentration (mg/dl)* | *34 ± 8* |
| *TC:HDL ratio* | *4.52 ± 1.12* |
| *LDL concentration (mg/dl)* | *94 ± 17* |
| *TRG (mg/dl)* | *90 ± 32* |
| *Glu (mg/dl)* | *88 ± 7* |
| *Plasma CRP (ng/mL)* | *13.77 ± 19.98* |
| *HDL function (oxidation slope, FU/min)* | *47.81 ± 20.56* |

*All values presented as Mean ± SD. BMD = bone mineral density; $VO_2$max = maximal oxygen uptake; Vmax = estimated running velocity at $VO_2$max; Hrmax = maximum recorded heart rate.*

**Table 2** – *Baseline comparison between groups.*

| Variable | Group | | p-value |
|---|---|---|---|
| **Anthropometrics** | **HIIT** | **ET** | |
| *Age (years)* | *21.4 ± 1.1* | *21.8 ± 2.1* | *0.66* |
| *Height (m)* | *1.80 ± 0.05* | *1.83 ± 0.10* | *0.61* |
| *Weight (kg)* | *81.90 ± 10.00* | *90.20 ± 21.05* | *0.40* |
| **Body composition** | | | |
| *Percent fat (%)* | *28.23 ± 7.03* | *27.90 ± 7.97* | *0.94* |
| *Android fat (%)* | *34.98 ± 8.23* | *36.78 ± 9.60* | *0.74* |
| *Gynoid fat (%)* | *33.97 ± 7.38* | *32.15 ± 7.63* | *0.67* |
| **Cardiovascular characteristics** | | | |
| *$^{+}VO_2max$ (ml/kg/min)* | *41.4 ± 5.5* | *41.9 ± 5.8* | *0.87* |
| *Vmax (mph)* | *7.5 ± 0.5* | *7.4 ± 1.2* | *0.88* |
| *HRmax (bpm)* | *198 ± 11* | *196 ± 7* | *0.77* |
| *Systolic blood pressure (mmHg)* | *120 ± 7* | *115 ± 7* | *0.30* |
| *Diastolic blood pressure (mmHg)* | *76 ± 9* | *71 ± 7* | *0.36* |
| **Lipid profile** | | | |
| *Total cholesterol (mg/dl)* | *140 ± 17* | *155 ± 22* | *0.22* |
| **HDL concentration (mg/dl)* | *34 ± 9* | *34 ± 8* | *1.00* |
| *TC:HDL ratio* | *4.37 ± 1.39* | *4.67 ± 0.85* | *0.66* |
| *LDL concentration (mg/dl)* | *88 ± 17* | *101 ± 15* | *0.19* |
| *TRG (mg/dl)* | *92 ± 32* | *87 ± 36* | *0.41* |
| *Glu (mg/dl)* | *89 ± 6* | *87 ± 7* | *0.53* |
| *Plasma CRP (ng/mL)* | *5.12 ± 4.30* | *22.41 ± 26.09* | *0.23* |
| *HDL function (oxidation slope, FU/min)* | *45.40 ± 20.06* | *50.23 ± 23.11* | *0.81* |

*Values presented as Mean ± SD. *Indicates primary criteria used to match groups. $^{+}$Indicates secondary criteria used to match groups.*

**Table 3** – *HIIT group training data.*

| Participant | Weeks | Warm-up/Cool-down | Protocol |
|---|---|---|---|
| *1* | | | |
| | *1 – 2* | *3 min at 4.6 mph* | *(1 min at 6.8 mph; 1 min at 3.8 mph) x12* |
| | *3 – 4* | *3 min at 4.6 mph* | *(1 min at 7.6 mph; 1 min at 3.8 mph) x12* |
| | *5 – 6* | *3 min at 4.6 mph* | *(1 min at 7.7 mph; 1 min at 3.8 mph) x12* |
| | *7 – 8* | *3 min at 4.6 mph* | *(1 min at 8.5 mph; 1 min at 3.8 mph) x12* |
| *4* | | | |
| | *1 – 2* | *3 min at 5.1 mph* | *(1 min at 7.6 mph; 1 min at 4.2 mph) x12* |
| | *3 – 4* | *3 min at 5.1 mph* | *(1 min at 8.5 mph; 1 min at 4.2 mph) x12* |
| | *5 – 6* | *3 min at 5.3 mph* | *(1 min at 8.8 mph; 1 min at 4.4 mph) x12* |
| | *7 – 8* | *3 min at 5.3 mph* | *(1 min at 9.7 mph; 1 min at 4.4 mph) x12* |
| *6* | | | |
| | *1 – 2* | *3 min at 4.3 mph* | *(1 min at 6.5 mph; 1 min at 3.6 mph) x12* |
| | *3 – 4* | *3 min at 4.3 mph* | *(1 min at 7.2 mph; 1 min at 3.6 mph) x12* |
| | *5 – 6* | *3 min at 4.4 mph* | *(1 min at 7.4 mph; 1 min at 3.7 mph) x12* |
| | *7 – 8* | *3 min at 4.4 mph* | *(1 min at 8.1 mph; 1 min at 3.7 mph) x12* |
| *9* | | | |
| | *1 – 2* | *3 min at 4.2 mph* | *(1 min at 6.3 mph; 1 min at 3.5 mph) x12* |
| | *3 – 4* | *3 min at 4.2 mph* | *(1 min at 7.0 mph; 1 min at 3.5 mph) x12* |
| | *5 – 6* | *3 min at 4.6 mph* | *(1 min at 7.6 mph; 1 min at 3.8 mph) x12* |
| | *7 – 8* | *3 min at 4.6 mph* | *(1 min at 8.4 mph; 1 min at 3.8 mph) x12* |
| *11* | | | |
| | *1 – 2* | *3 min at 4.3 mph* | *(1 min at 6.4 mph; 1 min at 3.6 mph) x12* |
| | *3 – 4* | *3 min at 4.3 mph* | *(1 min at 7.2 mph; 1 min at 3.6 mph) x12* |
| | *5 – 6* | *3 min at 4.5 mph* | *(1 min at 7.6 mph; 1 min at 3.8 mph) x12* |
| | *7 – 8* | *3 min at 4.5 mph* | *(1 min at 8.3 mph; 1 min at 3.8 mph) x12* |
| *13* | | | |
| | *1 – 2* | *3 min at 4.4 mph* | *(1 min at 6.5 mph; 1 min at 3.6 mph) x12* |
| | *3 – 4* | *3 min at 4.4 mph* | *(1 min at 7.3 mph; 1 min at 3.6 mph) x12* |
| | *5 – 6* | *3 min at 4.4 mph* | *(1 min at 7.3 mph; 1 min at 3.6 mph) x12* |
| | *7 – 8* | *3 min at 4.4 mph* | *(1 min at 7.9 mph; 1 min at 3.6 mph) x12* |
| *Average (n=6)* | | | |
| | *1 – 2* | *4.5 ± 0.3 mph* | *6.7 ± 0.5 mph; 3.7 ± 0.3 mph* |
| | *3 – 4* | *4.5 ± 0.3 mph* | *7.5 ± 0.5 mph; 3.7 ± 0.3 mph* |
| | *5 – 6* | *4.6 ± 0.3 mph* | *7.7 ± 0.5 mph; 3.9 ± 0.3 mph* |
| | *7 – 8* | *4.6 ± 0.3 mph* | *8.5 ± 0.6 mph; 3.9 ± 0.3 mph* |

*Individual training protocols for HIIT group. Weeks 1-2 average 70% Vmax, weeks 3-6 average 75% Vmax, and weeks 7-8 average 80% Vmax. Average values presented as Mean ± SD.*

**Table 4** – *ET group training data.*

| Participant | Weeks | Warm-up/Cool-down | Protocol |
|---|---|---|---|
| *2* | | | |
| | *1 – 2* | *3 min at 4.2 mph* | *24 min at 4.9 mph* |
| | *3 – 4* | *3 min at 4.2 mph* | *24 min at 5.2 mph* |
| | *5 – 6* | *3 min at 4.3 mph* | *24 min at 5.4 mph* |
| | *7 – 8* | *3 min at 4.3 mph* | *24 min at 5.7 mph* |
| *5* | | | |
| | *1 – 2* | *3 min at 4.5 mph* | *24 min at 5.2 mph* |
| | *3 – 4* | *3 min at 4.5 mph* | *24 min at 5.6 mph* |
| | *5 – 6* | *3 min at 4.7 mph* | *24 min at 5.9 mph* |
| | *7 – 8* | *3 min at 4.7 mph* | *24 min at 6.3 mph* |
| *8* | | | |
| | *1 – 2* | *3 min at 3.7 mph* | *24 min at 4.3 mph* |
| | *3 – 4* | *3 min at 3.7 mph* | *24 min at 4.6 mph* |
| | *5 – 6* | *3 min at 3.9 mph* | *24 min at 4.8 mph* |
| | *7 – 8* | *3 min at 3.9 mph* | *24 min at 5.2 mph* |
| *10* | | | |
| | *1 – 2* | *3 min at 5.0 mph* | *24 min at 5.8 mph* |
| | *3 – 4* | *3 min at 5.0 mph* | *24 min at 6.2 mph* |
| | *5 – 6* | *3 min at 5.0 mph* | *24 min at 6.2 mph* |
| | *7 – 8* | *3 min at 5.0 mph* | *24 min at 6.6 mph* |
| *12* | | | |
| | *1 – 2* | *3 min at 5.5 mph* | *24 min at 6.4 mph* |
| | *3 – 4* | *3 min at 5.5 mph* | *24 min at 6.9 mph* |
| | *5 – 6* | *3 min at 5.9 mph* | *24 min at 7.4 mph* |
| | *7 – 8* | *3 min at 5.9 mph* | *24 min at 7.9 mph* |
| *14* | | | |
| | *1 – 2* | *3 min at 3.6 mph* | *24 min at 4.2 mph* |
| | *3 – 4* | *3 min at 3.6 mph* | *24 min at 4.5 mph* |
| | *5 – 6* | *3 min at 3.9 mph* | *24 min at 4.9 mph* |
| | *7 – 8* | *3 min at 3.9 mph* | *24 min at 5.2 mph* |
| *Average (n=6)* | | | |
| | *1 – 2* | *4.4 ± 0.7 mph* | *5.1 ± 0.9 mph* |
| | *3 – 4* | *4.4 ± 0.7 mph* | *5.5 ± 0.9 mph* |
| | *5 – 6* | *4.6 ± 0.8 mph* | *5.8 ± 1.0 mph* |
| | *7 – 8* | *4.6 ± 0.8 mph* | *6.2 ± 1.0 mph* |

*Individual training protocols for ET group. Weeks 1-2 average 70% Vmax, weeks 3-6 average 75% Vmax, and weeks 7-8 average 80% Vmax. Average values presented as Mean ± SD.*

**Table 5** – *Training adaptations in the HIIT group.*

| Variable | Pre-training | Post-training | p-value |
|---|---|---|---|
| **Body composition** | | | |
| *Weight (kg)* | *81.90 ± 10.00* | *80.62 ± 9.51* | *0.24* |
| *Percent fat (%)* | *28.23 ± 7.03* | *27.32 ± 7.66* | *0.21* |
| *Android fat (%)* | *34.98 ± 8.23* | *33.13 ± 9.87* | *0.24* |
| *Gynoid fat (%)* | *33.97 ± 6.45* | *33.97 ± 7.38* | *1.00* |
| **Cardiovascular characteristics** | | | |
| *$VO_2$max (ml/kg/min)* | *41.4 ± 5.5* | *44.1 ± 6.8* | *0.13* |
| *Vmax (mph)* | *7.5 ± 0.5* | *8.1 ± 0.8* | *0.02** |
| *HRmax (bpm)* | *198 ± 11* | *193 ± 12* | *0.01** |
| *Systolic blood pressure (mmHg)* | *120 ± 7* | *122 ± 12* | *0.68* |
| *Diastolic blood pressure (mmHg)* | *76 ± 9* | *78 ± 7* | *0.54* |
| **Lipid profile** | | | |
| *Total cholesterol (mg/dl)* | *140 ± 17* | *143 ± 24* | *0.73* |
| *HDL concentration (mg/dl)* | *34 ± 9* | *40 ± 9* | *0.33* |
| *TC:HDL ratio* | *4.37 ± 1.39* | *3.85 ± 1.29* | *0.30* |
| *LDL concentration (mg/dl)* | *88 ± 17* | *92 ± 28* | *0.65* |
| *TRG (mg/dl)* | *92 ± 32* | *61 ± 12* | *0.04** |
| *Glu (mg/dl)* | *89 ± 6* | *90 ± 8* | *0.91* |
| **Inflammation** | | | |
| *Plasma CRP (ng/mL)* | *5.12 ± 4.30* | *5.50 ± 3.61* | *0.84* |
| *Plasma IL-6 response (ng/mL)* | *-0.09 ± 0.48* | *5.92 ± 9.14* | *0.22* |
| *HDL function (oxidation slope, FU/min)* | *45.40 ± 20.06* | *43.66 ± 9.45* | *0.63* |

*Adaptations to 8 weeks of exercise using HIIT. Values are presented as Mean ± SD. * indicates significantly different from baseline ($p<0.05$).*

**Table 6** – *Training adaptations in the ET group.*

| Variable | Pre-training | Post-training | p-value |
|---|---|---|---|
| **Body composition** | | | |
| *Weight (kg)* | *90.20 ± 21.05* | *90.57 ± 21.88* | *0.65* |
| *Percent fat (%)* | *27.90 ± 7.97* | *26.37 ± 9.01* | *0.10* |
| *Android fat (%)* | *36.78 ± 9.60* | *34.18 ± 11.39* | *0.045** |
| *Gynoid fat (%)* | *32.15 ± 7.63* | *31.12 ± 8.43* | *0.55* |
| **Cardiovascular characteristics** | | | |
| *$VO_2$max (ml/kg/min)* | *41.9 ± 5.8* | *43.5 ± 7.2* | *0.31* |
| *Vmax (mph)* | *7.4 ±1.2* | *7.7 ± 1.4* | *0.26* |
| *HRmax (bpm)* | *196 ± 7* | *194 ± 6* | *0.41* |
| *Systolic blood pressure (mmHg)* | *115 ± 7* | *115 ± 8* | *0.96* |
| *Diastolic blood pressure (mmHg)* | *71 ± 7* | *78 ± 7* | *0.20* |
| **Lipid profile** | | | |
| *Total cholesterol (mg/dl)* | *155 ± 22* | *159 ± 24* | *0.62* |
| *HDL concentration (mg/dl)* | *34 ± 8* | *41 ± 10* | *0.08* |
| *TC:HDL ratio* | *4.67 ± 0.85* | *4.07 ± 0.96* | *0.02** |
| *LDL concentration (mg/dl)* | *101 ± 15* | *99 ± 17* | *0.75* |
| *TRG (mg/dl)* | *87 ± 36* | *97 ± 43* | *0.13* |
| *Glu (mg/dl)* | *87 ± 7* | *92 ± 5* | *0.29* |
| **Inflammation** | | | |
| *Plasma CRP (ng/mL)* | *22.41 ± 26.09* | *15.46 ± 11.30* | *0.84* |
| *Plasma IL-6 response (ng/mL)* | *-1.99 ± 4.00* | *0.70 ± 2.21* | *0.84* |
| *HDL function (oxidation slope, FU/min)* | *50.23 ± 23.11* | *30.14 ± 19.76* | *0.21* |

*Adaptations to 8 weeks of exercise using ET. Values are presented as Mean ± SD. * indicates significantly different from baseline ($p<0.05$).*

**Table 7** – *Comparison of adaptations between the HIIT and ET groups.*

| Variable | Average change | | p-value |
|---|---|---|---|
| **Body composition** | **HIIT** | **ET** | |
| *Weight (kg)* | *-1.28 ± 2.36* | *0.37 ± 1.87* | *0.21* |
| *Percent fat (%)* | *-0.92 ± 1.57* | *-1.53 ± 1.85* | *0.55* |
| *Android fat (%)* | *-1.85 ± 3.36* | *-2.60 ± 2.41** | *0.67* |
| *Gynoid fat (%)* | *0.00 ± 1.17* | *-1.03 ± 3.92* | *0.55* |
| **Cardiovascular characteristics** | | | |
| *$VO_2$max (ml/kg/min)* | *2.72 ± 3.73* | *1.63 ± 3.54* | *0.62* |
| *Vmax (mph)* | *0.63 ± 0.48** | *0.33 ± 0.64* | *0.38* |
| *HRmax (bpm)* | *-5.33 ± 2.88** | *-2.17 ± 5.91* | *0.27* |
| *Systolic blood pressure (mmHg)* | *2.17 ± 11.94* | *-0.17 ± 8.50* | *0.71* |
| *Diastolic blood pressure (mmHg)* | *2.17 ± 7.96* | *7.17 ± 11.84* | *0.41* |
| **Lipid profile** | | | |
| *Total cholesterol (mg/dl)* | *3.17 ± 20.93* | *3.33 ± 15.45* | *0.99* |
| *HDL concentration (mg/dl)* | *5.33 ± 12.23* | *6.50 ± 7.15* | *0.84* |
| *TC:HDL ratio* | *-0.52 ± 1.10* | *-0.60 ± 0.41** | *0.87* |
| *LDL concentration (mg/dl)* | *4.33 ± 22.19* | *-2.00 ± 14.55* | *0.57* |
| *TRG (mg/dl)* | *-31.33 ± 28.32** | *19.00 ± 22.66* | *0.009$^+$* |
| *Glu (mg/dl)* | *0.33 ± 7.12* | *4.83 ± 10.07* | *0.39* |
| **Inflammation** | | | |
| *Plasma CRP (ng/mL)* | *0.38 ± 2.09* | *-6.95 ± 21.16* | *0.88* |
| *Plasma IL-6 response (ng/mL)* | *5.38 ± 9.34* | *2.69 ± 5.59* | *0.35* |
| *HDL function (oxidation slope, FU/min)* | *1.74 ± 17.02* | *20.09 ± 29.84* | *0.27* |

*Average change after 8 weeks of either HIIT or ET. Values are presented as Mean ± SD. * indicates significantly different from baseline ($p < 0.05$). $^+$ indicates HIIT significantly different from ET ($p < 0.05$).*

CUMULATIVE REFERENCES


1. Boden, W.E., *High-density lipoprotein cholesterol as an independent risk factor in cardiovascular disease: Assessing the data from Framingham to the Veterans Affairs high-density lipoprotein intervention trial.* American Journal of Cardiology, 2000. **86**(12A): p. 19l-22l.
2. Ridker, P.M., et al., *C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women.* New England Journal of Medicine, 2000. **342**(12): p. 836-843.
3. Blair, S.N., et al., *Physical fitness and all-cause mortality. A prospective study of healthy men and women.* The Journal of the American Medical Association, 1989. **262**(17): p. 2395-401.
4. Kavanagh, T., et al., *Prediction of long-term prognosis in 12 169 men referred for cardiac rehabilitation.* Circulation, 2002. **106**(6): p. 666-671.
5. Blair, S.N., et al., *Physical fitness and all-cause mortality in hypertensive men.* Annals of Internal Medicine, 1991. **23**(3): p. 307-12.
6. Blair, S.N., et al., *Changes in physical fitness and all-cause mortality. A prospective study of healthy and unhealthy men.* The Journal of the American Medical Association, 1995. **273**(14): p. 1093-8.
7. Keteyian, S.J., et al., *Peak aerobic capacity predicts prognosis in patients with coronary heart disease.* American Heart Journal, 2008. **156**(2): p. 292-300.
8. Wilson, P.W.F., et al., *Prediction of coronary heart disease using risk factor categories.* Circulation, 1998. **97**(18): p. 1837-1847.
9. Cleeman, J.I., et al., *Executive summary of the Third Report of the National Cholesterol Education Program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III).* The Journal of the American Medical Association, 2001. **285**(19): p. 2486-2497.
10. Aviram, M. and B. Fuhrman, *LDL oxidation by arterial wall macrophages depends on the oxidative status in the lipoprotein and in the cells: Role of prooxidants vs. antioxidants.* Molecular and Cellular Biochemistry, 1998. **188**(1-2): p. 149-159.
11. SanchezQuesada, J.L., et al., *Increase of LDL susceptibility to oxidation occurring after intense, long duration aerobic exercise.* Atherosclerosis, 1995. **118**(2): p. 297-305.
12. Lewis, G.F. and D.J. Rader, *New insights into the regulation of HDL metabolism and reverse cholesterol transport.* Circulation, 2005. **96**(12): p. 1221-32.
13. Rader, D.J., et al., *Markedly Accelerated Catabolism of Apolipoprotein a-Ii (Apoa-Ii) and High-Density-Lipoproteins Containing Apoa-Ii in Classic Lecithin - Cholesterol Acyltransferase Deficiency and Fish-Eye Disease.* Journal of Clinical Investigation, 1994. **93**(1): p. 321-330.
14. Assmann, G. and A.M. Gotto, *HDL cholesterol and protective factors in atherosclerosis.* Circulation, 2004. **109**(23): p. 8-14.
15. Navab, M., et al., *High-density lipoprotein: antioxidant and anti-inflammatory properties.* Current Atherosclerosis Reports, 2007. **9**(3): p. 244-8.
16. Ansell, B.J., et al., *Inflammatory/antiinflammatory properties of high-density lipoprotein distinguish patients from control subjects better than high-density lipoprotein cholesterol levels and are favorably affected by simvastatin treatment.* Circulation, 2003. **108**(22): p. 2751-2756.

17. Navab, M., et al., *A cell-free assay for detecting HDL that is dysfunctional in preventing the formation of or inactivating oxidized phospholipids.* Journal of Lipid Research, 2001. **42**(8): p. 1308-1317.
18. Navab, M., et al., *The role of dysfunctional HDL in atherosclerosis.* Journal of Lipid Research, 2009. **50**: p. S145-S149.
19. Mattusch, F., et al., *Reduction of the plasma concentration of C-reactive protein following nine months of endurance training.* International Journal of Sports Medicine, 2000. **21**(1): p. 21-24.
20. Aronson, D., et al., *C-Reactive protein is inversely related to physical fitness in middle-aged subjects.* Atherosclerosis, 2004. **176**(1): p. 173-9.
21. Wannamethee, S.G., et al., *Physical activity and hemostatic and inflammatory variables in elderly men.* Circulation, 2002. **105**(15): p. 1785-1790.
22. Mora, S., et al., *Physical activity and reduced risk of cardiovascular events - Potential mediating mechanisms.* Circulation, 2007. **116**(19): p. 2110-2118.
23. Durstine, J.L., et al., *Blood lipid and lipoprotein adaptations to exercise - A quantitative analysis.* Sports Medicine, 2001. **31**(15): p. 1033-1062.
24. SanchezQuesada, J.L., et al., *LDL from aerobically-trained subjects shows higher resistance to oxidative modification than LDL from sedentary subjects.* Atherosclerosis, 1997. **132**(2): p. 207-213.
25. Roberts, C.K., et al., *Effect of a short-term diet and exercise intervention on oxidative stress, inflammation, MMP-9, and monocyte chemotactic activity in men with metabolic syndrome factors.* Journal of Applied Physiology, 2006. **100**(5): p. 1657-65.
26. Church, T.S., et al., *Associations between cardiorespiratory fitness and C-reactive protein in men.* Arteriosclerosis Thrombosis and Vascular Biology, 2002. **22**(11): p. 1869-1876.
27. Fischer, C.P., et al., *Endurance training reduces the contraction-induced interleukin-6 mRNA expression in human skeletal muscle.* American Journal of Physiology-Endocrinology and Metabolism, 2004. **287**(6): p. E1189-E1194.
28. Hellsten, Y., F.S. Apple, and B. Sjodin, *Effect of sprint cycle training on activities of antioxidant enzymes in human skeletal muscle.* Journal of Applied Physiology, 1996. **81**(4): p. 1484-1487.
29. Tyldum, G.A., et al., *Endothelial Dysfunction Induced by Post-Prandial Lipemia Complete Protection Afforded by High-Intensity Aerobic Interval Exercise.* Journal of the American College of Cardiology, 2009. **53**(2): p. 200-206.
30. Febbraio, M.A. and B.K. Pedersen, *Muscle-derived interleukin-6: mechanisms for activation and possible biological roles.* Faseb Journal, 2002. **16**(11): p. 1335-1347.
31. Ostrowski, K., P. Schjerling, and B.K. Pedersen, *Physical activity and plasma interleukin-6 in humans - effect of intensity of exercise.* European Journal of Applied Physiology, 2000. **83**(6): p. 512-515.
32. Helgerud, J., et al., *Aerobic high-intensity intervals improve VO2max more than moderate training.* Medicine and Science in Sports and Exercise, 2007. **39**(4): p. 665-671.
33. Makrides, L., G.J. Heigenhauser, and N.L. Jones, *High-intensity endurance training in 20- to 30- and 60- to 70-yr-old healthy men.* Journal of Applied Physiology, 1990. **69**(5): p. 1792-8.
34. Amundsen, B.H., et al., *High-intensity aerobic exercise improves diastolic function in coronary artery disease.* Scandinavian Cardiovascular Journal, 2008. **42**(2): p. 110-117.
35. Moholdt, T.T., et al., *Aerobic interval training versus continuous moderate exercise after coronary artery bypass surgery: A randomized study of cardiovascular effects and quality of life.* American Heart Journal, 2009. **158**(6): p. 1031-1037.

36. Munk, P.S., et al., *High-intensity interval training may reduce in-stent restenosis following percutaneous coronary intervention with stent implantation: A randomized controlled trial evaluating the relationship to endothelial function and inflammation.* American Heart Journal, 2009. **158**(5): p. 734-741.
37. Rognmo, O., et al., *High intensity aerobic interval exercise is superior to moderate intensity exercise for increasing aerobic capacity in patients with coronary artery disease.* European Journal of Cardiovascular Prevention & Rehabilitation, 2004. **11**(3): p. 216-222.
38. Schjerve, I.E., et al., *Both aerobic endurance and strength training programmes improve cardiovascular health in obese adults.* Clinical Science, 2008. **115**(9-10): p. 283-293.
39. Tjonna, A.E., et al., *Aerobic interval training reduces cardiovascular risk factors more than a multitreatment approach in overweight adolescents.* Clinical Science, 2009. **116**(3-4): p. 317-326.
40. Tjonna, A.E., et al., *Aerobic interval training versus continuous moderate exercise as a treatment for the metabolic syndrome - A pilot study.* Circulation, 2008. **118**(4): p. 346-354.
41. Wisloff, U., et al., *Superior cardiovascular effect of aerobic interval training versus moderate continuous training in heart failure patients - A randomized study.* Circulation, 2007. **115**(24): p. 3086-3094.
42. Gormley, S.E., et al., *Effect of Intensity of Aerobic Training on VO2max.* Medicine and Science in Sports and Exercise, 2008. **40**(5): p. S42-S42.
43. Laursen, P.B., *Training for intense exercise performance: high-intensity or high-volume training?* Scandinavian Journal of Medicine & Science in Sports, 2010. **20**: p. 1-10.
44. Daussin, F.N., et al., *Effect of interval versus continuous training on cardiorespiratory and mitochondrial functions: relationship to aerobic performance improvements in sedentary subjects.* American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 2008. **295**(1): p. R264-R272.
45. Ciolac, E.G., et al., *Effects of high-intensity aerobic interval training vs. moderate exercise on hemodynamic, metabolic and neuro-humoral abnormalities of young normotensive women at high familial risk for hypertension.* Hypertension Research, 2010. **33**(8): p. 836-843.
46. Honda, H., et al., *Oxidized high-density lipoprotein as a risk factor for cardiovascular events in prevalent hemodialysis patients.* Atherosclerosis, 2012. **220**(2): p. 493-501.
47. Honda, H., et al., *Serum albumin, C-reactive protein, interleukin 6, and fetuin A as predictors of malnutrition, cardiovascular disease, and mortality in patients with ESRD.* American Journal of Kidney Diseases, 2006. **47**(1): p. 139-148.
48. VanLenten, B.J., et al., *Anti-inflammatory HDL becomes pro-inflammatory during the acute phase response - Loss of protective effect of HDL against LDL oxidation in aortic wall cell cocultures.* Journal of Clinical Investigation, 1995. **96**(6): p. 2758-2767.
49. Feingold, K.R., et al., *Paraoxonase activity in the serum and hepatic mRNA levels decrease during the acute phase response.* Atherosclerosis, 1998. **139**(2): p. 307-315.
50. Lindhorst, E., et al., *Acute inflammation, acute phase serum amyloid A and cholesterol metabolism in the mouse.* Biochimica et Biophysica Acta, 1997. **1339**(1): p. 143-54.
51. Kunimoto, S., et al., *Steryl glucoside is a lipid mediator in stress-responsive signal transduction.* Cell Structure and Function, 2002. **27**(3): p. 157-162.
52. Ridker, P.M., *On evolutionary biology, inflammation, infection, and the causes of atherosclerosis.* Circulation, 2002. **105**(1): p. 2-4.
53. Cabana, V.G., J.N. Siegel, and S.M. Sabesin, *Effects of the Acute Phase Response on the Concentration and Density Distribution of Plasma-Lipids and Apolipoproteins.* Journal of Lipid Research, 1989. **30**(1): p. 39-49.

54. Navab, K.D., et al., *Chronic Inflammatory Disorders and Accelerated Atherosclerosis: Chronic Kidney Disease.* Current Pharmaceutical Design, 2011. **17**(1): p. 17-20.
55. Vaziri, N.D., M. Navab, and A.M. Fogelman, *HDL metabolism and activity in chronic kidney disease.* Nature Reviews Nephrology, 2010. **6**(5): p. 287-296.
56. Broedl, U.C., W. Jin, and D.J. Rader, *Endothelial lipase: a modulator of lipoprotein metabolism upregulated by inflammation.* Trends in Cardiovascular Medicine, 2004. **14**(5): p. 202-6.
57. Blades, B., G.L. Vega, and S.M. Grundy, *Activities of Lipoprotein-Lipase and Hepatic Triglyceride Lipase in Postheparin Plasma of Patients with Low Concentrations of Hdl Cholesterol.* Arteriosclerosis and Thrombosis, 1993. **13**(8): p. 1227-1235.
58. Jaye, M., et al., *A novel endothelial-derived lipase that modulates HDL metabolism.* Nature Genetics, 1999. **21**(4): p. 424-428.
59. Jin, W., et al., *Inhibition of endothelial lipase causes increased HDL cholesterol levels in vivo.* Journal of Clinical Investigation, 2003. **111**(3): p. 357-62.
60. Ross, R., *Mechanisms of disease - Atherosclerosis - An inflammatory disease.* New England Journal of Medicine, 1999. **340**(2): p. 115-126.
61. Navab, M., et al., *Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein: step 1.* Journal of Lipid Research, 2000. **41**(9): p. 1481-1494.
62. Kelesidis, T., et al., *A biochemical fluorometric method for assessing the oxidative properties of HDL.* Journal of Lipid Research, 2011. **52**(12): p. 2341-2351.
63. Festa, A., et al., *The relation of body fat mass and distribution to markers of chronic inflammation.* International Journal of Obesity, 2001. **25**(10): p. 1407-1415.
64. Tchernof, A., et al., *Weight loss reduces C-reactive protein levels in obese postmenopausal women.* Circulation, 2002. **105**(5): p. 564-569.
65. Taylor, C., et al., *Hematologic, Iron-Related, and Acute-Phase Protein Responses to Sustained Strenuous Exercise.* Journal of Applied Physiology, 1987. **62**(2): p. 464-469.
66. Siegel, A.J., et al., *Effect of marathon running on inflammatory and hemostatic markers.* American Journal of Cardiology, 2001. **88**(8): p. 918-+.
67. Strachan, A.F., et al., *C Reactive Protein Concentrations during Long-Distance Running.* British Medical Journal, 1984. **289**(6454): p. 1249-1251.
68. Pedersen, B.K., et al., *Muscle-derived interleukin-6: lipolytic, anti-inflammatory and immune regulatory effects.* Pflugers Archiv-European Journal of Physiology, 2003. **446**(1): p. 9-16.
69. Starkie, R.L., et al., *Carbohydrate ingestion attenuates the increase in plasma interleukin-6, but not skeletal muscle interleukin-6 mRNA, during exercise in humans.* Journal of Physiology-London, 2001. **533**(2): p. 585-591.
70. Nieman, D.C., et al., *Influence of mode and carbohydrate on the cytokine response to heavy exertion.* Medicine and Science in Sports and Exercise, 1998. **30**(5): p. 671-678.
71. Nielsen, H.B., et al., *Lymphocytes and NK cell activity during repeated bouts of maximal exercise.* American Journal of Physiology, 1996. **271**(1 Pt 2): p. R222-7.
72. Ostrowski, K., et al., *Chemokines are elevated in plasma after strenuous exercise in humans.* European Journal of Applied Physiology, 2001. **84**(3): p. 244-245.
73. Pedersen, B.K. and M.A. Febbraio, *Muscle as an endocrine organ: Focus on muscle-derived interleukin-6.* Physiological Reviews, 2008. **88**(4): p. 1379-1406.
74. Coyle, E.F., et al., *Muscle Glycogen Utilization during Prolonged Strenuous Exercise When Fed Carbohydrate.* Journal of Applied Physiology, 1986. **61**(1): p. 165-172.
75. Margeli, A., et al., *Dramatic elevations of interleukin-6 and acute-phase reactants in athletes participating in the ultradistance foot race spartathlon: severe systemic inflammation and lipid and*

*lipoprotein changes in protracted exercise.* Journal of Clinical Endocrinology & Metabolism, 2005. **90**(7): p. 3914-8.
76. Matthys, P., et al., *Anti-gamma interferon and anti-interleukin-6 antibodies affect staphylococcal enterotoxin B-induced weight loss, hypoglycemia, and cytokine release in D-galactosamine-sensitized and unsensitized mice.* Infection and Immunity, 1995. **63**(4): p. 1158-64.
77. Starkie, R., et al., *Exercise and IL-6 infusion inhibit endotoxin-induced TNF-alpha production in humans.* Faseb Journal, 2003. **17**(3): p. 884-+.
78. Gleeson, M., B. McFarlin, and M. Flynn, *Exercise and Toll-like receptors.* Exercise Immunology Review, 2006. **12**: p. 34-53.
79. McFarlin, B.K., et al., *Physical activity status, but not age, influences inflammatory biomarkers and toll-like receptor 4.* Journals of Gerontology Series a-Biological Sciences and Medical Sciences, 2006. **61**(4): p. 388-393.
80. Powers, S.K., L.L. Ji, and C. Leeuwenburgh, *Exercise training-induced alterations in skeletal muscle antioxidant capacity: a brief review.* Medicine and Science in Sports and Exercise, 1999. **31**(7): p. 987-997.
81. van Oostrom, A.J.H.H.M., et al., *Postprandial recruitment of neutrophils may contribute to endothelial dysfunction.* Journal of Lipid Research, 2003. **44**(3): p. 576-583.
82. Plotnick, G.D., M.C. Corretti, and R.A. Vogel, *Effect of antioxidant vitamins on the transient impairment of endothelium-dependent brachial artery vasoactivity following a single high-fat meal.* The Journal of the American Medical Association, 1997. **278**(20): p. 1682-1686.
83. Booth, F.W. and K.M. Baldwin, *Muscle Plasticity: Energy Demand and Supply Processes*, in *Comprehensive Physiology*2010, John Wiley & Sons, Inc.
84. Zhang, J.Q., et al., *Changes in LPLa and reverse cholesterol transport variables during 24-h postexercise period.* American Journal of Physiology-Endocrinology and Metabolism, 2002. **283**(2): p. E267-E274.
85. Gupta, A.K., et al., *Increased Reverse Cholesterol Transport in Athletes.* Metabolism-Clinical and Experimental, 1993. **42**(6): p. 684-690.
86. Olchawa, B., et al., *Physical fitness and reverse cholesterol transport.* Arteriosclerosis Thrombosis and Vascular Biology, 2004. **24**(6): p. 1087-1091.
87. Iborra, R.T., et al., *Aerobic exercise training improves the role of high-density lipoprotein antioxidant and reduces plasma lipid peroxidation in type 2 diabetes mellitus.* Scandinavian Journal of Medicine & Science in Sports, 2008. **18**(6): p. 742-750.
88. Leon, A.S. and O.A. Sanchez, *Response of blood lipids to exercise training alone or combined with dietary intervention.* Medicine and Science in Sports and Exercise, 2001. **33**(6 Suppl): p. S502-15; discussion S528-9.
89. Kokkinos, P.F., et al., *Miles Run Per Week and High-Density-Lipoprotein Cholesterol Levels in Healthy, Middle-Aged Men - a Dose-Response Relationship.* Archives of Internal Medicine, 1995. **155**(4): p. 415-420.
90. Williams, P.T., *Relationship of distance run per week to coronary heart disease risk factors in 8283 male runners - The national runners' health study.* Archives of Internal Medicine, 1997. **157**(2): p. 191-198.
91. Drygas, W., et al., *Long-term effects of different physical activity levels on coronary heart disease risk factors in middle-aged men.* International Journal of Sports Medicine, 2000. **21**(4): p. 235-241.
92. Kodama, S., et al., *Effect of aerobic exercise training on serum levels of high-density lipoprotein cholesterol - A meta-analysis.* Archives of Internal Medicine, 2007. **167**(10): p. 999-1008.

93. Farrell, P.A. and J. Barboriak, *The Time Course of Alterations in Plasma-Lipid and Lipoprotein Concentrations during 8 Weeks of Endurance Training.* Atherosclerosis, 1980. **37**(2): p. 231-238.
94. Stein, R.A., et al., *Effects of Different Exercise Training Intensities on Lipoprotein Cholesterol Fractions in Healthy Middle-Aged Men.* American Heart Journal, 1990. **119**(2): p. 277-283.
95. Williams, P.T., *Relationships of heart disease risk factors to exercise quantity and intensity.* Archives of Internal Medicine, 1998. **158**(3): p. 237-245.
96. Slentz, C.A., et al., *Inactivity, exercise training and detraining, and plasma lipoproteins. STRRIDE: a randomized, controlled study of exercise intensity and amount.* Journal of Applied Physiology, 2007. **103**(2): p. 432-42.
97. Thomas, T.R., et al., *Effects of interval and continuous running on HDL-cholesterol, apoproteins A-1 and B, and LCAT.* Canadian Journal of Applied Sports Science, 1985. **10**(1): p. 52-9.
98. Gaesser, G.A. and R.G. Rich, *Effects of High-Intensity and Low-Intensity Exercise Training on Aerobic Capacity and Blood-Lipids.* Medicine and Science in Sports and Exercise, 1984. **16**(3): p. 269-274.
99. Nybo, L., et al., *High-Intensity Training versus Traditional Exercise Interventions for Promoting Health.* Medicine and Science in Sports and Exercise, 2010. **42**(10): p. 1951-1958.
100. Billat, L.V., *Interval training for performance: A scientific and empirical practice - Special recommendations for middle- and long-distance running. Part II: Anaerobic interval training.* Sports Medicine, 2001. **31**(2): p. 75-90.
101. Laursen, P.B. and D.G. Jenkins, *The scientific basis for high-intensity interval training - Optimising training programmes and maximising performance in highly trained endurance athletes.* Sports Medicine, 2002. **32**(1): p. 53-73.
102. Warburton, D.E., et al., *Effectiveness of high-intensity interval training for the rehabilitation of patients with coronary artery disease.* American Journal of Cardiology, 2005. **95**(9): p. 1080-4.
103. Laursen, P.B., et al., *Interval training program optimization in highly trained endurance cyclists.* Medicine and Science in Sports and Exercise, 2002. **34**(11): p. 1801-1807.
104. Londeree, B.R., *Effect of training on lactate/ventilatory thresholds: A meta-analysis.* Medicine and Science in Sports and Exercise, 1997. **29**(6): p. 837-843.
105. Esfarjani, F. and P.B. Laursen, *Manipulating high-intensity interval training: Effects on VO2max, the lactate threshold and 3000 m running performance in moderately trained mates.* Journal of Science and Medicine in Sport, 2007. **10**(1): p. 27-35.
106. Cornish, A.K., S. Broadbent, and B.S. Cheema, *Interval training for patients with coronary artery disease: a systematic review.* European Journal of Applied Physiology, 2011. **111**(4): p. 579-589.
107. Gibala, M.J. and S.L. Mcgee, *Metabolic adaptations to short-term high-intensity interval training: A little pain for a lot of gain?* Exercise and Sport Sciences Reviews, 2008. **36**(2): p. 58-63.
108. Burgomaster, K.A., et al., *Divergent response of metabolite transport proteins in human skeletal muscle after sprint interval training and detraining.* American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 2007. **292**(5): p. R1970-6.
109. Burgomaster, K.A., et al., *Similar metabolic adaptations during exercise after low volume sprint interval and traditional endurance training in humans.* Journal of Physiology, 2008. **586**(1): p. 151-60.
110. Rakobowchuk, M., et al., *Sprint interval and traditional endurance training induce similar improvements in peripheral arterial stiffness and flow-mediated dilation in healthy humans.* American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 2008. **295**(1): p. R236-R242.

111. Burgomaster, K.A., G.J.F. Heigenhauser, and M.J. Gibala, *Effect of short-term sprint interval training on human skeletal muscle carbohydrate metabolism during exercise and time-trial performance.* Journal of Applied Physiology, 2006. **100**(6): p. 2041-2047.
112. Hazell, T.J., et al., *10 or 30-s sprint interval training bouts enhance both aerobic and anaerobic performance.* European Journal of Applied Physiology, 2010. **110**(1): p. 153-160.
113. Guiraud, T., et al., *Optimization of high intensity interval exercise in coronary heart disease.* European Journal of Applied Physiology, 2010. **108**(4): p. 733-740.
114. Moholdt, T., et al., *Aerobic interval training increases peak oxygen uptake more than usual care exercise training in myocardial infarction patients: a randomized controlled study.* Clinical Rehabilitation, 2012. **26**(1): p. 33-44.
115. Harmer, A.R., et al., *Sprint Training Increases Muscle Oxidative Metabolism During High-Intensity Exercise in Patients With Type 1 Diabetes.* Diabetes Care, 2008. **31**(11): p. 2097-2102.
116. Little, J.P., et al., *Low-volume high-intensity interval training reduces hyperglycemia and increases muscle mitochondrial capacity in patients with type 2 diabetes.* Journal of Applied Physiology, 2011. **111**(6): p. 1554-1560.
117. Whyte, L.J., J.M.R. Gill, and A.J. Cathcart, *Effect of 2 weeks of sprint interval training on health-related outcomes in sedentary overweight/obese men.* Metabolism-Clinical and Experimental, 2010. **59**(10): p. 1421-1428.
118. Musa, D.I., et al., *The Effect of a High-Intensity Interval Training Program on High-Density Lipoprotein Cholesterol in Young Men.* Journal of Strength and Conditioning Research, 2009. **23**(2): p. 587-592.
119. Cohn, J.S., *Postprandial lipemia: Emerging evidence for atherogenicity of remnant lipoproteins.* Canadian Journal of Cardiology, 1998. **14**: p. 18b-27b.
120. Nappo, F., et al., *Postprandial endothelial activation in healthy subjects and in type 2 diabetic patients: Role of fat and carbohydrate meals.* Journal of the American College of Cardiology, 2002. **39**(7): p. 1145-1150.
121. Gill, J.M.R., et al., *Effects of prior moderate exercise on postprandial metabolism and vascular function in lean and centrally obese men.* Journal of the American College of Cardiology, 2004. **44**(12): p. 2375-2382.
122. Whaley, M.H., et al., *ACSM's guidelines for exercise testing and prescription*. 7th ed2006, Philadelphia, Pa.: Lippincott Williams & Wilkins. xxi, 366 p.
123. Cullinane, E.M., et al., *Variations in Plasma-Volume Affect Total and Low-Density-Lipoprotein Cholesterol Concentrations during the Menstrual-Cycle.* Metabolism-Clinical and Experimental, 1995. **44**(8): p. 965-971.
124. Demacker, P.N.M., et al., *Influence of Contraceptive Pill and Menstrual-Cycle on Serum-Lipids and High-Density Lipoprotein Cholesterol Concentrations.* British Medical Journal, 1982. **284**(6324): p. 1213-1215.
125. Kasapis, C. and P.D. Thompson, *The effects of physical activity on serum C-reactive protein and inflammatory markers: a systematic review.* Journal of the American College of Cardiology, 2005. **45**(10): p. 1563-9.
126. Goldberg, C.S., A.R. Tall, and S. Krumholz, *Acute Inhibition of Hepatic Lipase and Increase in Plasma-Lipoproteins after Alcohol Intake.* Journal of Lipid Research, 1984. **25**(7): p. 714-720.
127. Crouter, S.E., et al., *Accuracy and reliability of the ParvoMedics TrueOne 2400 and MedGraphics VO2000 metabolic systems.* European Journal of Applied Physiology, 2006. **98**(2): p. 139-51.

128. Macfarlane, D.J. and H.L. Wu, *Inter-unit variability in two ParvoMedics TrueOne 2400 automated metabolic gas analysis systems.* European Journal of Applied Physiology, 2013. **113**(3): p. 753-62.
129. Issa, J.S., et al., *[Precision and accuracy of blood lipid analyses by a portable device (Cholestech-LDX)].* Arquivos Brasileiros de Cardiologia, 1996. **66**(6): p. 339-42.
130. Friedewald, W.T., R.I. Levy, and D.S. Fredrickson, *Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge.* Clinical Chemistry, 1972. **18**(6): p. 499-502.
131. Hickson, R.C., et al., *Time Course of the Adaptive Responses of Aerobic Power and Heart-Rate to Training.* Medicine and Science in Sports and Exercise, 1981. **13**(1): p. 17-20.
132. Croft, L., et al., *High-intensity interval training attenuates the exercise-induced increase in plasma IL-6 in response to acute exercise.* Applied Physiology Nutrition and Metabolism-Physiologie Appliquee Nutrition Et Metabolisme, 2009. **34**(6): p. 1098-1107.
133. Malatesta, D., et al., *Effect of High-Intensity Interval Exercise on Lipid Oxidation during Postexercise Recovery.* Medicine and Science in Sports and Exercise, 2009. **41**(2): p. 364-374.
134. Saltin, B. and J. Karlsson, *Muscle glycogen utilization during work of different intensities*, in *Muscle metabolism during exercise*1971, Springer. p. 289-299.
135. Karlsson, J. and B. Saltin, *Diet, Muscle Glycogen, and Endurance Performance.* Journal of Applied Physiology, 1971. **31**(2): p. 203-&.
136. Sprenger, H., et al., *Enhanced Release of Cytokines, Interleukin-2 Receptors, and Neopterin after Long-Distance Running.* Clinical Immunology and Immunopathology, 1992. **63**(2): p. 188-195.
137. Drenth, J.P.H., et al., *Endurance Run Increases Circulating Il-6 and Il-1ra but down-Regulates Ex-Vivo Tnf-Alpha and Il-1-Beta Production.* Journal of Applied Physiology, 1995. **79**(5): p. 1497-1503.
138. NehlsenCannarella, S.L., et al., *Carbohydrate and the cytokine response to 2.5 h of running.* Journal of Applied Physiology, 1997. **82**(5): p. 1662-1667.
139. Denadai, B.S., et al., *Interval training at 95% and 100% of the velocity at VO2 max: effects on aerobic physiological indexes and running performance.* Applied Physiology, Nutrition, and Metabolism, 2006. **31**(6): p. 737-43.
140. Fischer, C.P., *Interleukin-6 in acute exercise and training: what is the biological relevance?* Exercise Immunology Review, 2006. **12**: p. 6-33.
141. Hovanloo, F., T. Arefirad, and S. Ahmadizad, *Effects of sprint interval and continuous endurance training on serum levels of inflammatory biomarkers.* Journal of Diabetes & Metabolic Disorders, 2013. **12**(1): p. 22.
142. Niess, A.M., et al., *Evaluation of stress responses to interval training at low and moderate altitudes.* Medicine and Science in Sports and Exercise, 2003. **35**(2): p. 263-269.
143. Bogdanis, G.C., et al., *Contribution of phosphocreatine and aerobic metabolism to energy supply during repeated sprint exercise.* Journal of Applied Physiology, 1996. **80**(3): p. 876-884.
144. Krustrup, P., Y. Hellsten, and J. Bangsbo, *Intense interval training enhances human skeletal muscle oxygen uptake in the initial phase of dynamic exercise at high but not at low intensities.* Journal of Physiology-London, 2004. **559**(1): p. 335-345.
145. Stepto, N.K., et al., *Metabolic demands of intense aerobic interval training in competitive cyclists.* Medicine and Science in Sports and Exercise, 2001. **33**(2): p. 303-310.
146. Parra, J., et al., *The distribution of rest periods affects performance and adaptations of energy metabolism induced by high-intensity training in human muscle.* Acta Physiologica Scandinavica, 2000. **169**(2): p. 157-165.

147. Hokanson, J.E. and M.A. Austin, *Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a metaanalysis of population-based prospective studies.* Journal of cardiovascular risk, 1996. **3**(2): p. 213-219.
148. Couillard, C., et al., *Effects of endurance exercise training on plasma HDL cholesterol levels depend on levels of triglycerides - Evidence from men of the health, risk factors, exercise training and genetics (HERITAGE) family study.* Arteriosclerosis Thrombosis and Vascular Biology, 2001. **21**(7): p. 1226-1232.
149. Stensvold, D., et al., *Strength training versus aerobic interval training to modify risk factors of metabolic syndrome.* Journal of Applied Physiology, 2010. **108**(4): p. 804-810.
150. Bouchard, C. and T. Rankinen, *Individual differences in response to regular physical activity.* Medicine and Science in Sports and Exercise, 2001. **33**(6): p. S446-S451.
151. Bouchard, C., et al., *Familial aggregation of VO2max response to exercise training: results from the HERITAGE Family Study.* Journal of Applied Physiology, 1999. **87**(3): p. 1003-1008.
152. Stepto, N.K., et al., *Effects of different interval-training programs on cycling time-trial performance.* Medicine and Science in Sports and Exercise, 1999. **31**(5): p. 736-741.

# APPENDICES

*Appendix A*

Recruitment Script
"Effect of 8 Weeks of High-intensity Interval Training versus Traditional Endurance Training on the Blood Lipid Profile in Humans"

**Purpose:**
You are invited to participate in a research study comparing the effects of 8 weeks of high-intensity interval training to traditional endurance training on cholesterol levels and general cardiovascular health. To accomplish this, we are looking to recruit healthy individuals that are not currently exercising on a regular basis to complete 8 weeks of exercise training. We will measure cholesterol levels and cardiovascular fitness before and after the training period.

**Participant Qualifications:**
-Males, age 19 to 35
-Healthy (as determined by the Physical Activity Readiness Questionnaire)
-Blood pressure below 140/90
-Not taking anti-inflammatory, blood pressure, cholesterol, or heart rate altering medications
-Not currently engaging in vigorous physical activity more than 2 times per week (e.g. running, swimming laps, playing singles tennis, etc.)

**Requirements:**
If you decide to participate, you will be asked to read and sign Informed Consent documentation and complete the Physical Activity Readiness Questionnaire (PAR-Q). Preliminary testing before the start of training will consist of: a body composition assessment, consisting of measuring height, weight, and fat percentage using a measuring tape, scale, and DEXA machine; a blood pressure test, using a standard blood pressure cuff; a 10ml venous blood sample, taken by a trained phlebotomist using a small butterfly needle and syringe, to determine your cholesterol levels; and a VO2max test, consisting of running on a treadmill at progressively increasing speeds and inclines until you reach your maximal capability. After initial testing, you will either begin 8 weeks of high-intensity interval training, comprised of running at alternating low and high intensities, or 8 weeks of traditional endurance training, which involves running at a continuous, moderate pace. The initial testing session will be repeated after 4 weeks and after 8 weeks of training to examine the effects of the exercise training.

***YOUR PARTICIPATION IS COMPLETELY VOLUNTARY!***

*If you choose to volunteer for this study you have the right to stop any trial at any time.* Your participation or lack of participation in this study will in no way affect your relationship with the researchers or the Department of Kinesiology. The data recorded will only be identifiable by participant number.

**Contact Information:** Please contact David Elmer via email at elmerdj@auburn.edu or by telephone at 334-844-1479.

*Appendix B*

**Modified Physical Activity Readiness Questionnaire (PAR-Q)**

Please read each question carefully and answer honestly. If you do not understand the question, please ask the investigator for clarification. Check the appropriate answer.

**Any "yes" response will disqualify you for the study.**

| **NO** | **YES** | |
|---|---|---|
| _____ | _____ | 1. Are you under the age of 19 or over the age of 35? |
| _____ | _____ | 2. Do you presently smoke or have you ever been a regular smoker? |
| _____ | _____ | 3. Has your doctor ever said you have heart trouble? |
| _____ | _____ | 4. Do you have a family history of early cardiovascular death before the age of 50? |
| _____ | _____ | 5. Have you ever had a heart murmur, rheumatic fever or respiratory problems? |
| _____ | _____ | 6. Have you ever been told that you have a fast resting heart rate? |
| _____ | _____ | 7. Have you ever been told by your doctor or nurse that your blood pressure is too high? |
| _____ | _____ | 8. Have you ever been told that your cholesterol is too high? |
| _____ | _____ | 9. Have you ever been told that you have a kidney disorder? |
| _____ | _____ | 10. Have you been told that you have diabetes or that your blood sugar is too high? |
| _____ | _____ | 11. Have you been told that your electrocardiogram (EKG), 12 lead EKG or stress test is not normal? |
| _____ | _____ | 12. Have you been hospitalized in the last year? |

13. Please list any prescription medicine you are currently taking:

_____

***Note that taking certain medications may cause you to be excluded, including those that cause increases in heart rate and/or blood pressure, increase inflammation, alter cholesterol levels, or have other effects that may increase the risks associated with strenuous exercise.**

14. Do you have any reason to believe that your participation in this investigative effort may put your health or well-being at risk? If so, please state reason:

_____

**Signature of participant**: _____ **Date**: _____

*Appendix C*

# AUBURN UNIVERSITY

Auburn University, Alabama 36849-5323
Department of Kinesiology:
*phone* – (334) 844-4483; *fax* – (334) 844-1467
2050 Memorial Coliseum, Thermal Lab (room 2118)
*phone* – (334) 844-1479

**(NOTE: DO NOT SIGN THIS DOCUMENT UNLESS AN IRB APPROVAL STAMP WITH CURRENT DATES HAS BEEN APPLIED TO THIS DOCUMENT.)**

## INFORMED CONSENT
**For a research study entitled:**
**"Effect of 8 Weeks of High-intensity Interval Training versus Traditional Endurance Training on the Blood Lipid Profile in Humans"**

***Project overview:*** You are invited to participate in a study that seeks to determine whether 8 weeks of running repeated bouts of high-intensity intervals is more effective at improving cholesterol levels than 8 weeks of using the traditional method of running at a continuous pace for a certain amount of time. David Elmer, Dr. David Pascoe, and the Department of Kinesiology are recruiting participants to complete preliminary, intermediate, and post-training testing, as well as either 8 weeks of exercise training 3 days per week. Exercise sessions will either consist of running at alternating low and high intensities for 24 minutes, or running at a continuous pace for 24 minutes.

***Purpose:*** The purpose of this study is to determine whether running at alternating low and high intensities improves cholesterol levels, and whether this strategy is more effective at improving cholesterol levels than the traditional method of running at a constant speed.

***Participation Requirements:*** To be eligible to participate in this study, you must meet the following requirements:

1. Male, age 19 to 35
2. Healthy (as determined by the Physical Activity Readiness Questionnaire)
3. Resting blood pressure below 140/90
4. Not taking anti-inflammatory, blood pressure, cholesterol, or heart rate altering medications
5. Not currently engaging in vigorous physical activity more than 2 times per week (e.g. running, swimming laps, playing singles tennis, etc.)

***Testing Procedures:***

*Testing Day 1:* Hydration will be measured upon arrival to the lab, followed by a body composition assessment using DEXA, a measuring tape, and a scale. After this, your resting blood pressure will be measured using a standard blood pressure cuff. You will then be asked to give a 10ml venous blood sample, taken by a trained phlebotomist using a small butterfly needle and syringe, in order to determine your cholesterol levels before the start of exercise training. If you have a resting systolic blood pressure greater than 140 mmHg or diastolic pressure greater than 90 mmHg, you will not be allowed to participate in the study and will be advised to seek

medical treatment. After the blood sample is taken, you will complete a VO2max test, consisting of running on a treadmill at progressively higher speeds and inclines until you can no longer continue, to determine your maximal oxygen consumption and to formulate your training protocol. After testing, you will be placed into either the HIIT or ET group.

*Estimated time commitment:* ***about 1 hour***

Participants will come to the lab 3 times per week with at least one day between visits to complete their training sessions. Training for the HIIT group will consist of completing 12 intervals, with each interval comprised of 1 min of running at a low intensity and 1 min of running at a high intensity, resulting in a total of 24 min of running. Specific intensities will progress in the following manner:

Weeks 1-2 – 1 min at 50% of $VO_2$max, 1 min at 90% of $VO_2$max
Weeks 3-6 – 1 min at 50% of $VO_2$max, 1 min at 100% of $VO_2$max
Weeks 7-8 – 1 min at 50% of $VO_2$max, 1 min at 110% of $VO_2$max

Training for the ET group will consist of running at a constant pace of moderate intensity for 24 min, and will progress as follows:

Weeks 1-2 – 24 min at 70% of $VO_2$max
Weeks 3-6 – 24 min at 75% of $VO_2$max
Weeks 7-8 – 24 min at 80% of $VO_2$max

Training for both HIIT and ET groups averages the same relative intensity and volume, as well as duration, meaning each group will complete the exact same amount of exercise.

Before each exercise session, questions will be asked to determine if there is any health reason that you should not participate in exercise on that day. The exercise will be canceled for the day if it is determined that it would be unsafe for you to continue. On the first and last exercise days only, a small (5 ml) blood sample will be collected before and after the exercise session to determine the changes in inflammation as a result of the exercise.

*Estimated time commitment per day*: ***about 30 min (45 min on first day)***

*Estimated time commitment of 8 weeks of exercise training:* ***about 12 hours and 30 minutes***

*Testing Day 2:* After completing the first 4 weeks of exercise training, you will report to the lab to complete the same battery of tests as testing day 1.

*Estimated time commitment:* ***about 1 hour***

*Testing Day 3:* All participants will report to the lab at the end of the 8 week training period to complete the same battery of tests as testing days 1 and 2.

*Estimated time commitment:* ***about 1 hour***

*Estimated total time commitment:* ***about 15 hours, 30 min***

***Testing Variables:*** We will be measuring the change in total blood cholesterol levels (lipid profile) resulting from 8 weeks of either high-intensity interval training or traditional endurance training. We will also track the change in maximal oxygen consumption, chronic inflammation, and vascular health as determined by VO2max, blood sample analysis, and blood pressure.

***Potential Risks:***

1. While performing any type of exercise there is the risk of muscle strains/pulls, dehydration, nausea, dizziness, and even death. $VO_2$max testing with a metabolic

cart may cause additional discomfort, but this discomfort is minimal in this routine test.

2. The American College of Sports Medicine rates the risk of death at 0.05 per 10,000 individuals

**Note: You are responsible for any costs incurred as a result of injury*

***Precautions:***

1) Some of the testing and exercise sessions in this experiment will be at a high exercise intensity which may be uncomfortable. However, each trial will be conducted in a controlled laboratory setting, monitored by a researcher certified in adult CPR by the Red Cross. Also, our pre-screening via PAR-Q helps reduce the risk of injury during exercise.
2) It is recommended that you consume 500ml (about 1 pint) of water an hour before reporting to the lab for each session to ensure hydration. You will be allowed a complete warm-up and warm-down period before and after exercise as needed.

***Benefits:*** You will benefit from the physiologic testing conducted during this study that would not normally be available to you. This information gives you information on your physiologic and cardiopulmonary function capabilities, your cholesterol levels, and your fitness status, which has implications for your overall health. You will also benefit from the increases in health and physical fitness associated with regular exercise, such as decreased risk of death, reduced risk of cardiovascular disease, reduced risk of diabetes, increased bone and muscle health, reduced feelings of anxiety and depression, and better control of body weight.

***Compensation:*** For students in PHED jogging or walking classes, participation in this study will replace your class participation for the duration of the study (8 weeks). You will still be held accountable to the Departmental Attendance Policy explained in the class syllabus and will be responsible for any coursework assigned by your instructor. If you wish to stop participating in the study, then you may simply resume attending class at its scheduled time. Students in other PHED or KINE classes will receive 5 points toward their final grades, as verified by their instructors.

***Voluntary Participation:*** YOUR PARTICIPATION IS COMPLETELY **VOLUNTARY**. You have the right to stop any trial at any time, for any reason. Your complete participation or termination of participation will not affect your relationship with the researchers, the Thermal Lab, or the Department of Kinesiology.

***Confidentiality:*** All identifiable data collected or information recorded during this study will be kept purely confidential. All measurements will be recorded using a participant number coded to a master list of participants. Once data collection has ended, the master list will be destroyed, removing any identifiable link between you and the data. The non-identifiable data may be published or presented if results from the study warrant this.

**If you have any questions about this study,** please contact David Elmer by phone (334)-844-1479 or email at elmerdj@auburn.edu or Dr. David Pascoe by phone (334)-844-1479 or email at pascodd@auburn.edu.

**If you have questions about your rights as a research participant,** you may contact the Auburn University Office of Human Subjects Research or the Institutional Review Board by phone (334)-844-5966 or email at hsubjec@auburn.edu or IRBChair@auburn.edu.

**HAVING READ THE INFORMATION PROVIDED, YOU MUST DECIDE WHETHER YOU WISH TO PARTICIPATE IN THIS RESEARCH STUDY. YOUR SIGNATURE INDICATES YOUR WILLINGNESS TO PARTICIPATE. A copy of this consent will be given to you to keep.**

| | | |
|---|---|---|
| _____ | | _____ |
| Participant's Signature | Date | Print Name |
| _____ | | _____ |
| Investigator's Signature | Date | Print Name |
| _____ | | _____ |
| Witness | Date | Print Name |