Kehmeier et al. (2020) *Int J Res Ex Phys*. 15(2):65-90.

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International Journal of Research in Exercise Physiology

Original Research Article

Effects of Acute Reduced-Exertion High-Intensity Interval Training and Vigorous-Intensity Exercise on the Advanced Lipid Profile

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ABSTRACT

Introduction: Coronary Heart Disease (CHD) is the leading cause of death in the United States. In recent years, it has been reported that traditional lipid measurements may not be adequate to identify plaque accumulation and subsequent vessel occlusion in spite of lipid profiles falling within normal ranges (Shaheen et al 2014). Physical activity is undoubtedly associated with lowering risks of cardiovascular disease, however, the ideal intensity or amount of exercise has yet to be determined. The purpose of this crossover study was to examine the acute effects of exercise on apolipoproteins and particle size. **Methods**: Four male participants (age 40.25±13.1 yr, Ht 193.6±6.16 cm, Wt 102.93±11.54 kg) volunteered for this study, participating in pre-testing and two acute exercise bouts (ventilatory threshold (VT2) and reduced exertion high intensity interval training (REHIT)). Biological variability was calculated via coefficients of variation, and P value was set a <0.05. **Results**: Although no statistical significance was found, the REHIT session elicited greater effects than the VT2 bout. Decreases in TC, apoB, LDLC were seen following both acute exercise sessions. HDLC increased with both interventions and insulin increased with REHIT. **Conclusion**: This study showed REHIT is an effective exercise intervention in acutely altering lipids and increasing insulin. Biological variability permitted increased confidence that changes in the key outcome were attributable to the acute bouts of exercise rather than day-to-day variability and/or measurement error. Increases in insulin shows to be a key to favor lipid changes acutely.

Key words: Apolipoproteins, Coronary Heart Disease, Physical Activity.

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Introduction

Coronary Heart Disease (CHD) is the leading cause of death in the United States. Elevated total cholesterol (TC), low density lipoprotein (LDL-C), and triglycerides (TG) are highly associated with an increased risk for CHD[1,2]. Alternatively, higher levels of high-density lipoprotein (HDL) lower the risk of CHD. In recent years, it has been reported that traditional lipid measurements may not be adequate to identify plaque accumulation and subsequent vessel occlusion in spite of lipid profiles falling within normal ranges[3]. Freedman et al. [4] suggest there is a limitation to using a normal lipid panel. Information about particle sizes and the number of particles is lacking. According to Gerber et al.[5] small dense LDL particles are linked to an increase in atherosclerosis. HDL particle size may be atherogenic although this notion has been questions more recently. However, small HDL particles[6] are not atherogenic as once thought. According to Varady et al.[7], small HDL are potent in upregulating a key protein expressed on foam cells, in which cholesterol is effluxes from the foam cell to the immature HDL[8].

Another measure vital in lipid measures often overlooked are apolipoproteins. Apolipoproteins come in a wide variety, specific ones discussed in this paper however will be apolipoprotein A (apoA) and apolipoprotein B (apoB). ApoA is attached to all HDL cholesterol, giving us a more accurate measure of the number of HDL particles. ApoB's are attached on all atherogenic cholesterol. Thus meaning, chylomicrons, VLDL, IDL, and LDL. These measures are more accurate in depicting the number of atherogenic particles than compared to a standard lipid profile.

Extensive research has shown that exercise positively affects the lipid profile[9]. Physical activity is undoubtedly associated with lowering risks of cardiovascular disease; however, the ideal intensity or amount of exercise has yet to be determined. Various studies have investigated the optimal amount and intensity of exercise to elicit optimal effects on blood lipids[10,2,11,7,12]. A common theme across these studies was that an effective intensity and dose-response relationship needs to be established before implementing these practices into a clinical population. Kraus et al [10] argues that high-amount high-intensity (20 miles/wk, 65-80% of VO2max elicits the greatest and most beneficial effects to advanced lipid measures as evidenced by the following adaptations: LDL particle size increased, small LDL particles decreased, HDL particle size and concentration increased.

Exercise is often prescribed as a one size fits all concept, although individuals do not always benefit from this uniform method of prescription. Alternatively, an emerging way of prescribing exercise is off an individual's ventilatory thresholds. A study done by Weatherwax et al.[13], demonstrated that there was a significant effect of exercise intensity prescription method on the incidence of VO2max responders occurred

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with the individualized group eliciting 100% responsiveness, whereas the standardized group had a 60% incidence of response. HR intensity was prescribed off ventilatory threshold 1 (VT1) and ventilatory threshold 2 (VT2). We want to determine if personalizing the exercise prescription will be just as successful with eliciting positive responses in other areas beyond VO2max. To our knowledge, no studies have investigated particle size, or apolipoproteins using this exercise prescription method.

Another relatively new training paradigm is high-intensity interval training (HIIT). This fad has captivated the attention of the scientific community due to its ability to improve health and fitness in less of a time commitment[14]. However, HIIT protocols are not as time friendly as once thought to be. A time commitment of ~120 min/wk[15] is similar to those of ACSM's [16] guidelines of 150 min/wk of moderate intensity exercise. Emerging research suggests REHIT, characterized as minimal sprint durations (2x20 sec) elicits substantial cardiometabolic health benefits in a more time efficient manner[17,18]. A study done by Cuddy et al.[14], investigated an 8-week intervention of REHIT vs standardized moderate intensity. The REHIT protocol showed beneficial at a 3-minute warm up, 20 second sprint, 3-minute recovery (slow pedal), 20 second sprint, and then a 3-minute cool down. REHIT protocol showed more beneficial for waist circumference, systolic blood pressure, HDL cholesterol, and triglycerides. The HDL-C was increased for both exercise groups, however the REHIT group experienced a greater increase of HDL-C compared to the moderate intensity group. This suggests 1) exercise is beneficial at increasing HDL specifically and 2) REHIT protocol surpasses the threshold needed to elicit beneficial changes in cholesterol.

To our knowledge this is the first study to investigate the effects of REHIT and VT2 protocols on advanced lipid profiles. This study investigated the effect of these two protocols and their effect on particle size and particle count. Therefore, the purpose of the study is a crossover study that will examine the acute effects of apolipoproteins and particle size pre vs. post single exercise bout. A vast amount of studies has shown the positive benefits to the standard and advanced lipid profiles. This study will be the first to demonstrate acute effects REHIT and VT2 have on the lipid profile.

Methods

Subjects

Four male participants and sedentary individuals volunteered for this study. In order to participate in this study individuals had to obtain two or more risk factors according to ACSM guidelines pertaining to their lipid profile or obtain a high triglyceride (TG)/High density lipoprotein (HDL) ratio (>3.0). Participants were males from ages 30-55 years old, considered sedentary (according to the PACE [patient-centered assessment and counseling for exercise] questionnaire) and were able to perform vigorous exercise bouts on a stationary bike.

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If subjects were pregnant, under the age of 30 or over the age of 55, unable to exercise, not sedentary, had a low TG/HDL ratio, or did not possess two or more risk factors, they were excluded from the study.

All subjects filled out an informed consent and were provided with ample time to ask questions during the screening appointment. Once it was determined if the participant qualified for the study, they were given 24 hours to decide if they wish to participate in the study. Screening appointments took place at the High-Altitude Performance Lab (HAPLab) at Western Colorado University (WCU). Exercise bouts were performed in the HAPLab as well, and advanced lipid profile (ADLP) measures were taken at Gunnison Valley Hospital (GVH), then sent to Mayo Clinic in Denver, CO for analyses. This study was reviewed and approved by the Institutional Review Board at Western Colorado University [HRC2019-01-03-R75].

Experimental Design

In this randomized experimental crossover design, subjects were randomly assigned to either REHIT or VT2 group. Prior to the exercise interventions ADLP measures which were 48-hours apart to determine biological variability of the individual. After that, the individuals rested for 20-minutes while their resting metabolic rate was observed, then the participants performed a VO_2max test on the stationary bike. Participants then performed 3 consecutive REHIT familiarization on the HOR/CAROL bike. Following the fourth familiarization was a 30-minute bout of excess post exercise oxygen consumption (EPOC). A 7-day washout period occurred before and between each exercise intervention. See Figure 1 for a schematic representation of the design.

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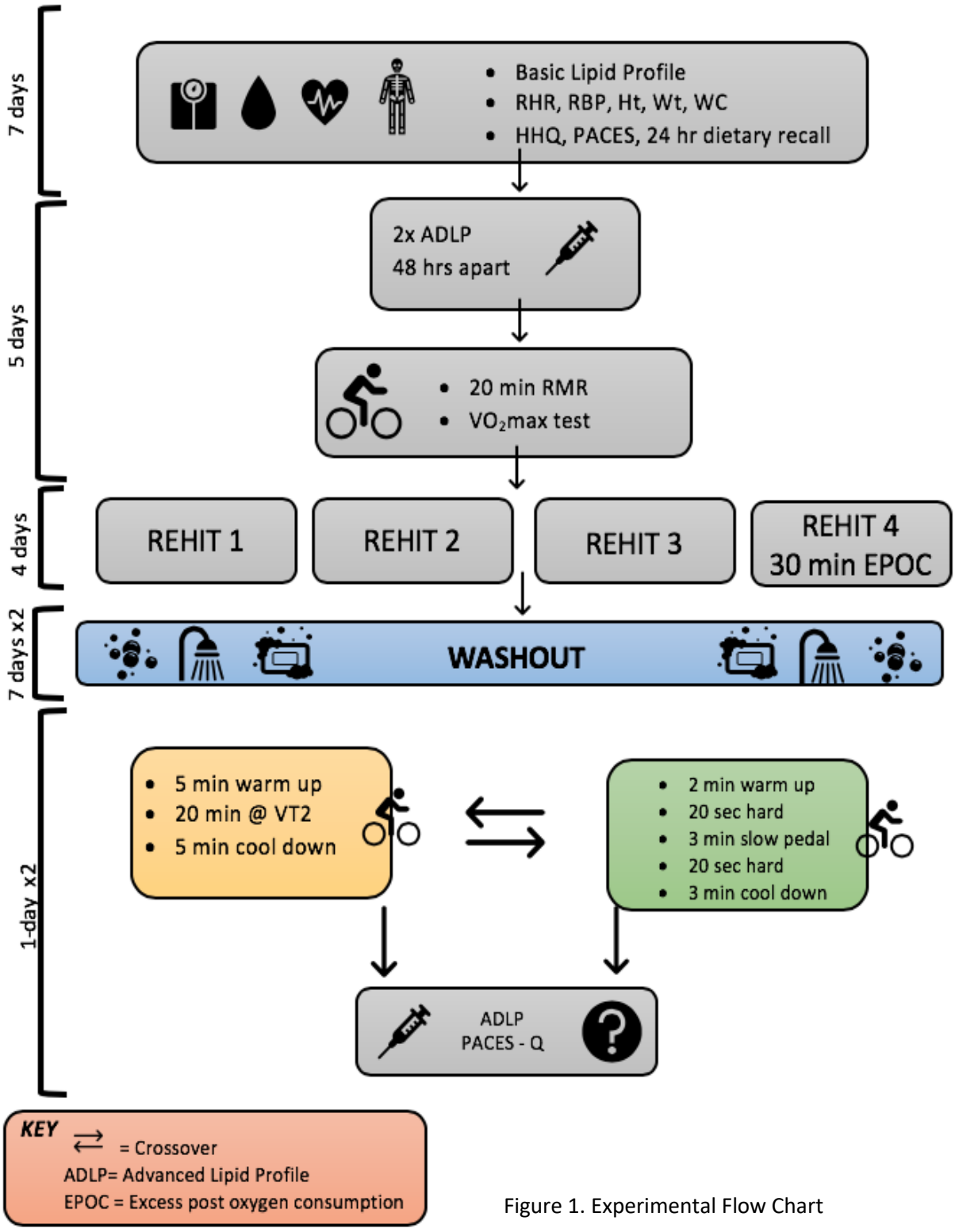

Figure 1. Experimental Flow Chart

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Procedures

Screening measures took place first, this included: blood lipid/glucose testing, resting heart rate, resting blood pressure, height, weight, waist circumference, and questionnaires. Second, two ADLP were taking 48 hours apart, followed by a VO2max. Third, the REHIT familiarization was performed followed by 30 minutes of EPOC analysis. Finally, the experiment was conducted with REHIT protocol and VT2 protocol as a crossover design, followed by ADLP measures. For a more detailed description of each measure see below.

Screening

An email was sent out to the campus of WCU. Individuals interested in the study were instructed to contact the PI (principal investigator) to set up a screening appointment. Potential subjects arrived at the HAPLab in the morning at a predetermined time, fasted for 12 hours to sign a written informed consent. Following the informed consent individuals went through testing of the blood lipid profile to ensure they had the appropriate TG/HDL ratio, along with other risk factors. The tests included: PACE questionnaire, waist circumference, resting heart rate (RHR) and blood pressure (BP), height (Ht), weight (Wt), and a 24-hour dietary recall.

Blood Lipid/ Glucose Testing

Participants were asked to fast for at least 12 hours, refrain exercise, caffeine intake, and smoking during that twelve hours. When the participant arrived, they were given an informed consent that was thoroughly explained to them. Following written informed consent, participants were instructed to wash their hands with warm water. The PI then wiped a chosen fingertip with an alcohol wipe and sat until their hands were dry. The tip of their finger was punctured using a lancet (Medipurpose, Brussels, Belgium) and a 40 µl sample was collected into a heparin-coated capillary tube (Abbott, Abbott Park, IL). Samples were distributed on to a cassette (Abbott, Abbott Park, IL), and inserted into the LDX Cholestech analyzer (Abbott, Abbott Park, IL) where the RBC's are separated from the plasma and reactions occur to give specific values. Measures recorded from the LDX Cholestech are total cholesterol (TC), low density lipoprotein (LDL) cholesterol, high density lipoprotein (HDL) cholesterol, triglycerides (TG), and blood glucose. This process took approximately ten minutes, and after the participants were offered a snack and/or juice to end their fast. Following the results of the lipid panel, and informed if they qualified for the study or not. If they qualified, the rest of the screening measures were completed, and the participant was allotted 24 hours to decide if they wanted to participate in the study. However, if the individuals did not meet inclusion criteria they were informed immediately. If they wished to complete the screening measures, they were allowed to do so understanding that they did not qualify for the study. All data was recorded on a data collection sheet.

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PACE/HHQ/Dietary Recall

Once the participants completed the lipid measures they were given a health history questionnaire (HHQ), 24-hour dietary recall form, and a sedentary behavior questionnaire (PACE). See attached forms for example of the questionnaires. These three questionnaires took approximately 10-15 minutes and were completed in the HAPLab at WCU. The PACE questionnaire was to quantify how sedentary the participant was. The HHQ is to confirm the individual will not have a negative response to exercise. The dietary recall was filled out in order to limit cofounding factors by instructing the participants to repeat the same diet before each blood test.

Resting Blood Pressure and Resting Heart Rate

Following the questionnaires, where the participant sat for at least 5 minutes, the participant will get two resting blood pressures. A blood pressure cuff (Mabis Healthcare Inc., Lake Forest, IL) was placed approximately 1 inch above the antecubital fossa on the subjects left arm. The cuff will be inflated to 220 mmHg, and then release while the PI will listen for the first and last Korotkoff sounds with a stethoscope (Littman Inc., Subury, MA). The heart rate was taken simultaneously, taking about six minutes in the HAPLab. After the participant was done filling out the questionnaires, remained seated to measure their resting heart rate taken. This was done by placing a pulse oximeter (Concord Health Supply, Skokie, IL) on the index/ring finger on the opposite hand in which blood was taken. This took about six minutes, taking place in the HAPLab. Heart rate and blood pressure will be recorded three times and then averaged.

Waist Circumference

A measuring tape (DJO Global, Vista, CA) was wrapped around the participants torso at the narrowest point (between the umbilicus and xyphoid process), this is where the measurement was performed three times and then averaged to ensure accuracy. The participant was instructed to inhale and exhale while the measurement was taken to ensure the participant did not suck in his/her stomach. This process took approximately 2 minutes in the HAPLab.

Height and Weight

Participants were asked to remove their shoes and any other bulky items they were wearing at the time, stepping on the scale with a connected measuring stick (Tanita, Arlington Heights, IL). Participants were asked to step on the scale with heels facing the wall, standing straight up, instructed to take a deep breath. The researcher then measured height and noted weight. This occurred in the HAPLab and took approximately one minute.

Comprehensive Testing

VO_2max/EPOC

This appointment took place in the morning, at least one day after completing ADLP measures. Subjects were fitted to a mask, attached to the falconia tubing that was

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attached to the metabolic cart (TrueOne 2400, PARVOMedics, Sandy, UT). Participants first sat still for 20 minutes attached to the metabolic cart to observe their resting metabolic rate. A 5-minute break was taken, and the participant was advised to hydrate with water as they wished.

After the 5-minute break, the participant was refitted to the mask, attached to the tube and metabolic cart. The subject was instructed to start a 3-4 minute warm up on a stationary bike at 50 watts (Lode Excalibur Sport, Groningen, the Netherlands).
After a proper warm-up, the participant began to pedal as fast as the can above 50 rpm. The intensity increased by 10 watts every minute, and once the individual dropped below 50 rpm the test was terminated and a proper cool down began. The cool down lasted approximately 5 minutes. The test and its entirety lasted approximately 15 minutes.

Throughout the test the participant used a chest strap to record their heart rate (Polar Electro, Woodbury, NY, USA). Prior to each test the metabolic cart was calibrated per manufacturer guidelines with a calibration gas mixture (16.00 % O2 and 4.00 % CO2) and room air (20.93 % O2 and 0.003 % CO2). Gas exchange data were averaged 15 sec, and VO2max was determined by averaging the final two 15-sec VO2 average data during the maximal test. The highest achieved HR during the GXT was considered the maximal HR (HRmax).

Lipoprotein Metabolism Profile (LMPP)
Fasting plasma samples were analyzed by ultracentrifugation, electrophoresis, automated enzymatic, and colorimetric analysis. Each measurement includes the etiology of cholesterol and or triglyceride elevation. Measurements of intermediate-density lipoprotein (IDL), very-low density lipoprotein (VLDL), lipoprotein a (Lp[a]), or even the abnormal lipoprotein complex LpX. Also received are LDL and HDL cholesterol, triglycerides in isolated lipoprotein fractions. LMPP measures require 5mL of a blood sample. Hyperlipoproteinemia's were classified into phenotypes to make a distinction of the individual's predisposition to the disease. After the sample was collected via cuboidal vein, blood was stored at -80 degrees Celsius and sent to Mayo Clinic in Denver, CO. Multiple freezes and thaws were avoided to ensure accuracy of measurements and decrease the variability of measures. Analyses were conducted and then results were sent back to GVH and given to the PI. This process will occur a total of four times per participant and will take approximately 10-15 minutes at GVH. Venipunctures were performed by certified phlebotomists in the blood lab at the hospital.

Insulin (INS)
Insulin serum levels were collected simultaneously the LMPP. 1mL of blood was taking from the fasting individual. Insulin was taken to correlate it effect on the LMPP measures. Particularly its effect on

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lipoprotein size and count. After the sample was collected via cuboidal vein, blood was stored at -80 degrees Celsius and sent to Mayo Clinic in Denver, CO. Multiple freezes and thaws were avoided to ensure accuracy of measurements and decrease the variability of measures. Analyses were conducted and then results were sent back to GVH and given to the PI. This process will occur a total of four times per participant and will take approximately 10-15 minutes at GVH. Venipunctures were performed by phlebotomists in the blood lab at the hospital.

Cholesteryl Esters (CHLE)

Cholesteryl Ester serum levels were observed with another 1mL of blood collected at the same time as the INS and LMPP tests. This test was performed at the via cuboidal vein, stored properly and sent to Mayo Clinic in Denver, CO. This test evaluates the deficiency and regulation of the enzyme lecithin-cholesterol acyltransferase (LCAT). LCAT is the prime enzyme in HDL development. Again, all venipunctures were performed by phlebotomists in the blood lab at GVH.

C-reactive protein (CRP)

CRP is measured in the blood and increases when there is inflammation in your body. Blood was obtained at the same time as the other blood measures via antecubital vein. These measures were taken to observed if the individuals were in an inflamed state or became in an inflamed state post exercise.

HbA1C

A1C reflects your average blood sugar over the past 2-3 months. A1C test measures what percent of hemoglobin is glycated. A normal A1C level is 5.7 or lower. Anything between 5.7 and 6.4 you have prediabetes. Blood was taken by the phlebotomist and analyzed in the lab in Gunnison.

Hematocrit

Hematocrit was measured to account for a decrease in plasma volume during workout. This decrease could affect cholesterol measures and could explain why we would potentially see fluctuation in some cases. Hematocrit is the total of RBC's to the volume of blood.

Intervention

Subjects were randomly assigned to REHIT or VT2 exercise group. Before each exercise session participants showed up fasted at least 12 hours. The day prior to the exercise testing, participants were asked to follow a 24-hr dietary recall from the initial screening appointment. Subjects in the REHIT group partook in a single session on the High-Octane Ride (HOR) bike (CAROL, Kensington, London) at the HAPLab. Each subject had his/her login to the bike accounting for their weight and max workload that was calculated prior to the exercise bout during the familiarization appointments. The exercise bout lasted 9 minutes and 40 seconds and consisted of a 3-minute warm-up, 20 second all-out sprint, 3-minute slow pedal, 20 second all-out sprint, and a 3-minute cool down. The session was

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supervised by the PI/research assistants; however, they did not encourage the participant in either scenario, they were supervising for individual's safety and to answer any questions that surfaced.

VT2 was prescribed off of the individuals VO_2max. VT2 is indicated by the second slope change on the graph provided from their VO_2max test. The individual warmed up at a self-selected pace for 5 minutes. Following the warm-up, the participant pedaled at their VT2 for a 20-minute bout, which was completed with a 5-minute cool down. VT2 is considered vigorous intensity in relation to recommended ACSM guidelines. After each exercise bout the participants were taken over to GVH and their blood samples were collected and sent for NMR analyses.

Statistical Analyses

All statistical analysis was performed on Prism version 8.4.5 (San Diego, CA, USA). Data were reported as a mean ± standard deviation (SD). A two tailed paired samples t-test was conducted to observe exercise overall effect on blood measures. A repeated measures one-way ANOVA was used to analyze with-in group differences. Effect sizes were calculated to measure the magnitude of the study and measures. Coefficient of variation (CV) was calculated to examine the changes among biological variability. Biological variability was used to determine responsiveness of the interventions. Chi-square (χ^2) tests were used to analyze the incidence of responders and non-responders for advanced lipid profile measures following the REHIT group separated by VT2 group. Significance value was set at $p<0.05$.

Results

Baseline Physiological and Lipid Characteristics

A total of seven males (age 40.3±13.1 yr) were screened for this study, four of which qualified (n=4), meeting inclusion criteria (possessing two or more cardiometabolic risk factors on the initial screening). All four participants elected to participate in the study. See table 1 for baseline physiological variables. All participants were asked to keep their diet the same and fast 12 hours before blood measures. To the best of our knowledge each individual was fasted and maintained their diet through the course of the study. Screening and intervention data are presented in table 1 and 2 respectively.

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Table 1. Baseline Physiological Variables.

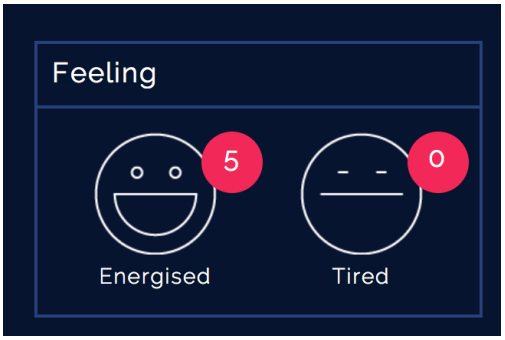
Variable	n=4
Age (yr)	40.25±13.1
Ht (cm)	193.6±6.2
Wt (kg)	102.9±11.54
BMI (kg/m^2)	27.5±3.9
TC (mg/dL)	222.1±31.0
ApoB (mg/dL)	126.9±23.9
LDLC (mg/dL)	151.8±29.8
LDLTG (mg/dL)	39±6.9
TG (mg/dL)	142.6±32.6
HDLC (mg/dL)	38.6±8.9
VLDLC (mg/dL)	28.1±11.6
VLDLTG (mg/dL)	86.3±24.7
Insulin (mcIU/mL)	11.5±5.3
A1C (%)	5.4±0.3
CE (%)	74.9±0.8
Hematocrit (%)	48.8±2.1
TG/HDL ratio (%)	3.94±1.4
RHR (bpm)	72.5±3.9
Systolic (mmHg)	123±10.9
Diastolic (mmHg)	84±6.5
VO_2max (ml/kg/m)	32.4±3.5
RMR (kcal)	2177.5±522.3
30 min EPOC (kcal)	66.75±5.7

Note: Data are presented as mean ± SD. Abbreviations: Height (Ht), weight (Wt), body mass index (BMI), total cholesterol (TC), apolipoprotein B (apoB), low density lipoprotein (LDLC), low density lipoprotein triglycerides (LDLTG), triglycerides (TG), high density lipoprotein (HDLC), very low density lipoprotein (VLDL), very low density lipoprotein triglycerides (VLDLTG), hemoglobin A1C (A1C), cholesterol esters (CE), resting heart rate (RHR), resting metabolic rate (RMR), excess post oxygen consumption (EPOC).

Safety and Enjoyment

Participants reported feeling energized after both works outs and said they felt good. N=18 rides (including familiarization bouts) report feeling energized. N=2 reported feeling tired after a total of 20 rides. No injuries due to the intervention or testing were reported during the study. All participants shared a sense of enjoyment from the interventions sharing they are likely to repeat the activity on their own.

Figure 2. Screen shot from CAROL bike regarding individuals feeling after REHIT bout.



Measurement Issues

One participant had to repeat his VT2 bout due to complications from the blood lab. Three out of four participants consistently had no trace of CRP and Lp(a) in their blood labs.

Table 2: Coefficients of variation (CV (%)), Typical error, and Pearson's R (α) for individual variability in advanced lipid measures

Variable	CV (%)	Typical Error	Pearson's R α
TC (mg/dL)	7.5	0.24	1
TG (mg/dL)	12.7	0.38	0.99
ApoB (mg/dL)	3.7	0.15	1
HDLC (mg/dL)	1.8	0.2	1
LDLC (mg/dL)	5.5	0.18	1
LDLTG (mg/dL)	2.5	0.37	0.99
VLDLC (mg/dL)	1.1	0.09	1
VLDLTG (mg/dL)	13.8	0.56	0.96
A1C (%)	0.1	0.29	1
Insulin (mcIU/mL)	2.1	0.42	0.97
CE (%)	0.4	0.43	0.97
Hematocrit (%)	0.9	0.46	0.96

Biological Variability

When analyzing for biological variability among initial lipid measures, coefficient of variation (CV), typical error (TE), and Pearson's R were calculated. See table 2 for individual variability CV, TE, and Pearson's R values.

VT2 and REHIT

See tables 3 and 4 for exercise specific data regarding power in watts, heart rate, RPE, peak power, peak power per pound, and total power.

Table 3. Ventilatory Threshold 2 exercise data. Displaying peak power in watts, rate of perceived exertion (RPE), and peak heart rate during the exercise bout.

Measures	Participant 1	Participant 2	Participant 3	Participant 4
Peak power (watts)	160	140	100	160
RPE	7	8	7	7
Heart Rate	113	168	114	158

Table 4. Reduced high intensity interval training (REHIT). Displaying peak power in watts, rate of perceived exertion (RPE), and peak heart rate during the exercise bout.

Measures	Participant 1	Participant 2	Participant 3	Participant 4
Peak power (watts)	686	795	713	874
Peak power per lb	3.0	3.8	3.6	4.0
Total power (watts)	30,185	28,483	23,122	32,073
Heart Rate	137	153	142	157

Exercise Post Oxygen Consumption (EPOC)

Below in Figure 3, it is depicted that REHIT is intense enough to elicit effects of EPOC. The 30-min EPOC equated to 72 kcal.

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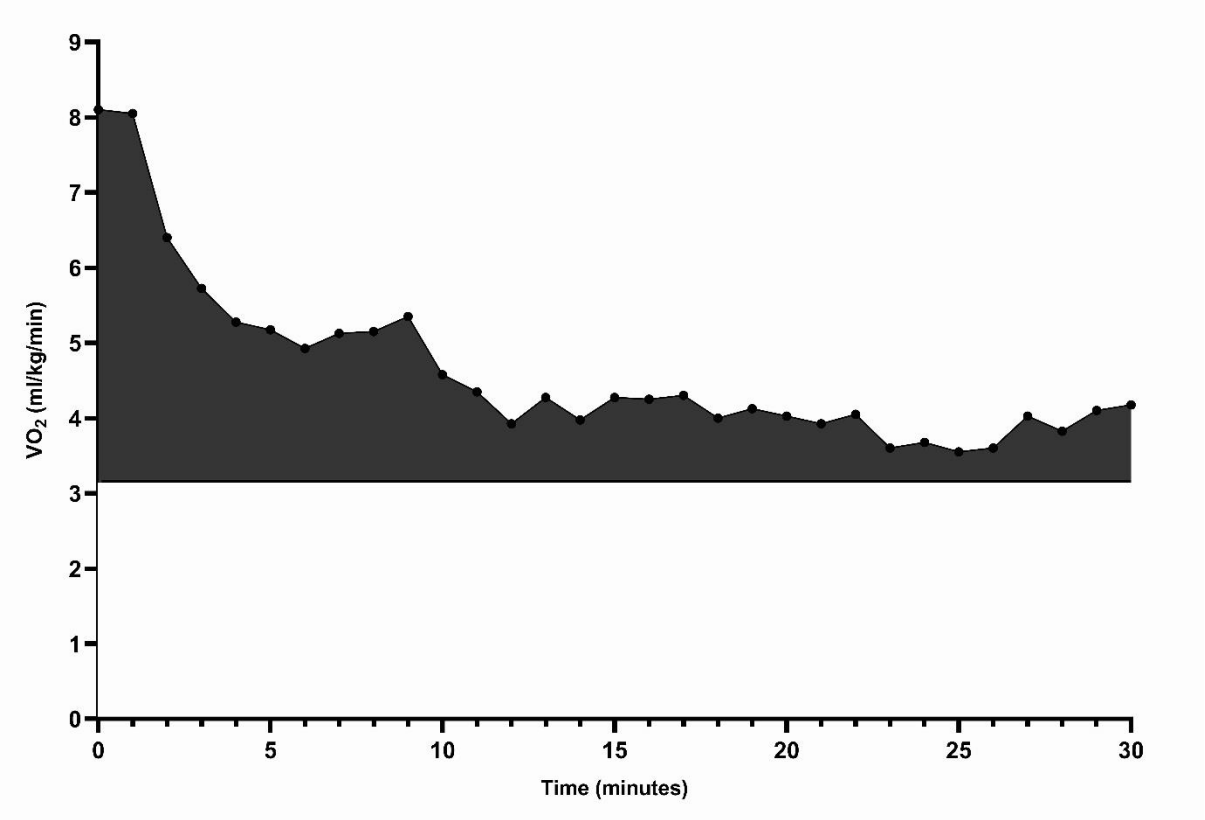

Figure 3. Area under the curve for post-exercise oxygen consumption. The line 3 on the y axis is the average RMR for the participants.

VT2 and REHIT Lipid Results

Graphs depicting group mean changes in HDLC, ApoB, LDLC, and TC are shown below (Figure 4). There were no significant changes between measures as seen in table 5. However, over all trends were seen and exercised was deemed beneficial. HDLC (F=1.128, p=.3826), was found to increase with exercise. ApoB (F=2.319, p=.1878) showed a decrease with exercise, tukey's multiple comparison baseline to REHIT (p=0.1612) was more effective than VT2 (p=0.67). As assumed since ApoB decreased with exercise so did LDLC (F=1.508, p=0.304), according to Tukey's multiple comparison, baseline to REHIT (p=0.14) was more effective than VT2 (p=0.72). Also shown below (Figure 4), a slight decrease in overall means were seen in TC (F=0.80, p=0.46).

Table 5. Mean changes from baseline to VT2 and REHIT, followed by their respective effect size (Cohens d). Significance was set at p<0.05.

Measures	Baseline	VT2	Effect Size (VT2)	REHIT	Effect Size (REHIT)	F	p
HDLC (mg/dL)	38.62±8.9	41.25±7.76	0.31	42.5±11.3	0.38	1.128	0.38
ApoB (mg/dL)	126.87±23.89	121.5±26.0	0.22	115.5±5	0.54	2.319	0.19
LDLC (mg/dL)	151.75±29.8	144.5±40.4	0.20	138.25±27.15	0.47	1.508	0.30
TC (mg/dL)	222.51±31.04	213.5±38.16	0.25	209.5±25.77	0.44	0.81	0.47
Insulin (mcIU/mL)	11.5±5.27	10.57±4.9	0.18	15.45±5.88	0.70	3.446	0.13

Note: Data are presented as mean ± SD. Abbreviations: high density lipoprotein (HDLC), apolipoprotein B (apoB), low density lipoprotein (LDLC), total cholesterol (TC).

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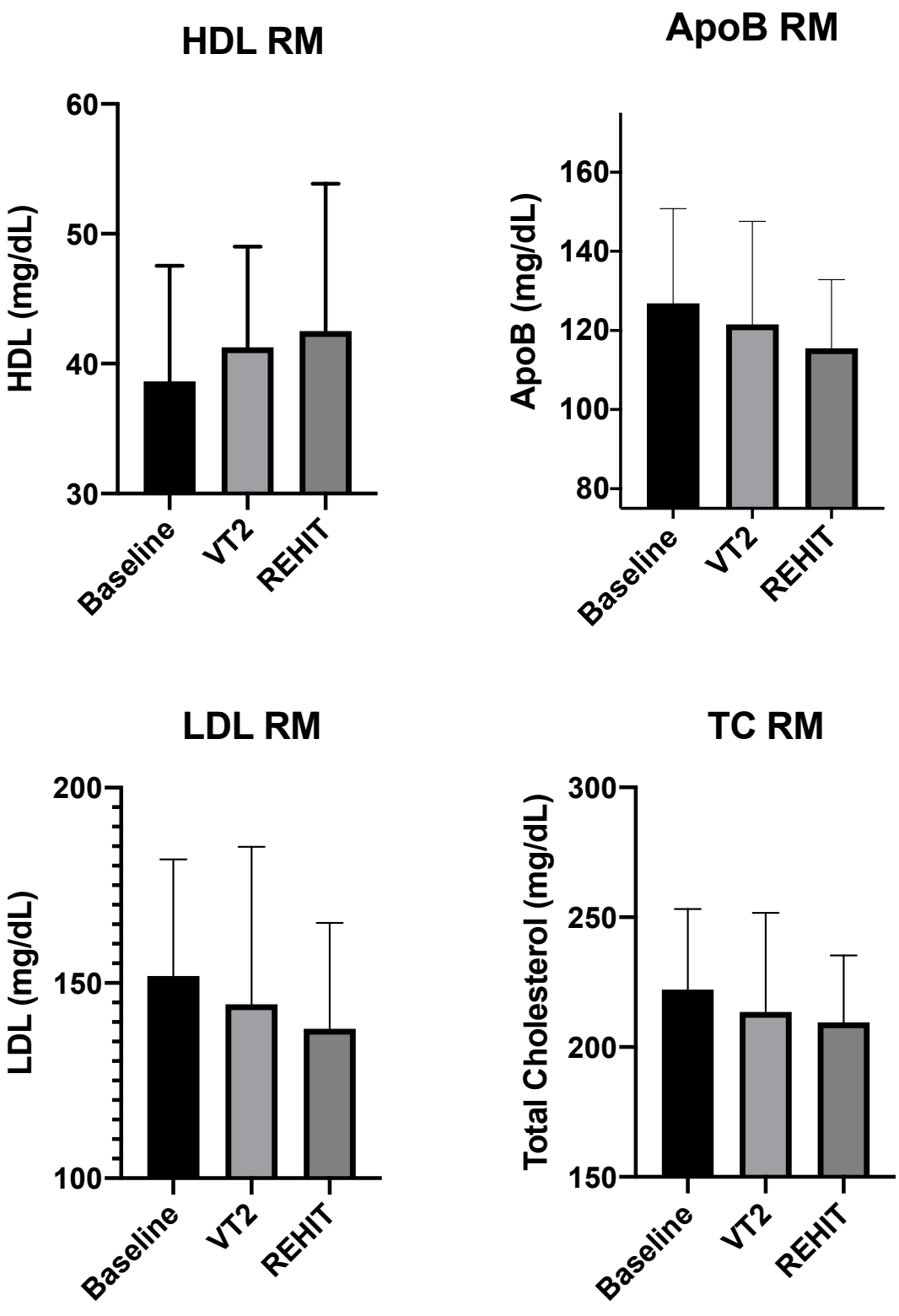

Figure 4. HDLC, ApoB, LDLC, and TC average repeated measures data.

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Individual Responses

Out of 5 of the variables (TC, ApoB, HDL, LDL, and insulin) looking at VT2 we had 8 responders with this intervention. With REHIT and the same variables we had 14 responders. This implies that REHIT was a more effective intervention by almost doubling responsiveness when analyzing the ADLP although it was not statistically significant (p = 0.055). See figure 5 to see responders, non-responders of HDLC, Insulin, LDLC, and ApoB.

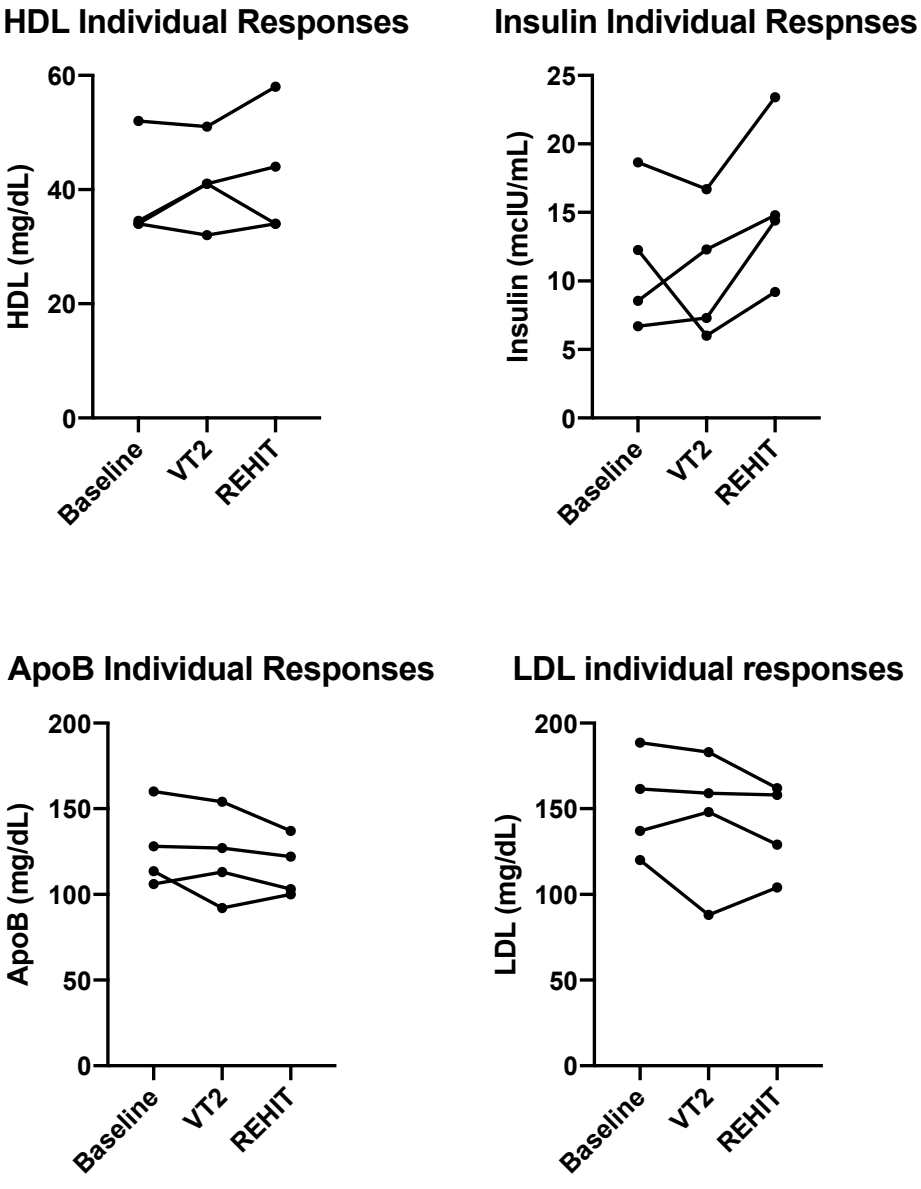

Figure 5. HDLC, Insulin, ApoB, and LDLC individual responses.

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Insulin Measures VT2 and REHIT

Seen in figure 6, REHIT was higher but no statistical significance was found. VT2 decreased 9% from baseline, while REHIT increased insulin 25.5% from baseline.

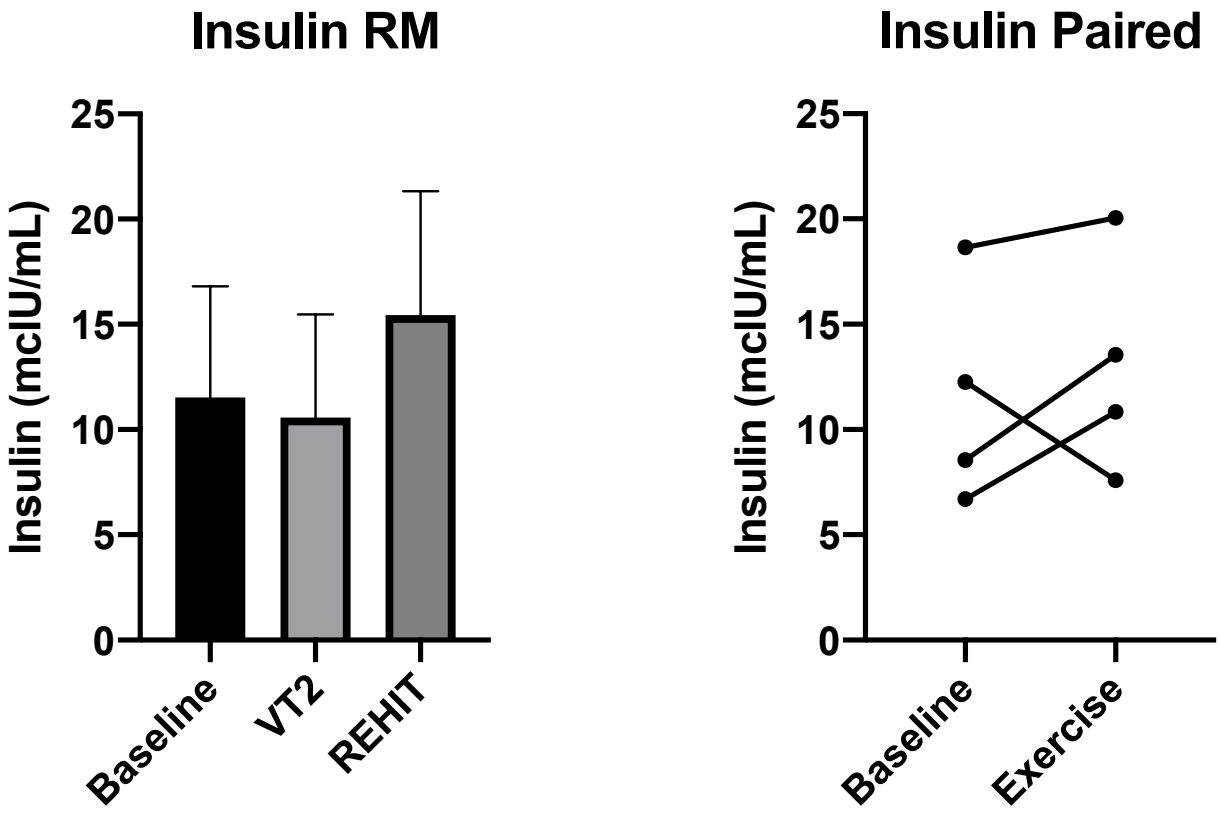

Figure 6. Insulin repeated measures and paired t-test showing overall effect of exercise on insulin.

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Discussion

This study found that in men with metabolic syndrome (MetS), acute VT2 and REHIT exercise bouts were both effective at lowering various lipid measures. Indeed, although no significance was found, our most notable findings were exercise interventions VT2 and REHIT were effective, REHIT was more effective as evidenced by the higher incidence of responders and larger effect size (see table 5). Importantly, REHIT was time-efficient with a total time of 8 minutes and 40 seconds comparing to VT2, which was 25 minutes including warm up, cool down, and the vigorous exercise routine itself. REHIT takes approximately a third of the time while showing more effectiveness. Lastly, this novel study was the first to our knowledge to demonstrate the acute benefits of REHIT on the lipid profile. These preliminary findings are encouraging and show the potential for REHIT to be used as an exercise strategy for those with dyslipidemia, which is 1 out of every 3 adults in the United States[19].

There were more responders with REHIT(n=14) than VT2 (n=8) when looking at the five main variables (TC, HDLC, ApoB, LDLC, and Insulin). This could be due to a higher intensity or more personalized exercise bout with REHIT. REHIT intensity is greater than VT2, attributing this to be why we saw a greater response number with REHIT. We can say with certainty if the individual responded or not based on the repeated baseline measures (see table 2). However, if an individual changed their diet this could've affected baseline measures. See figure 5, to visualize individual responses of HDL, LDL, insulin, and ApoB.

The lack of accounting for biological variability often serves as the main contributor to lack of significance in other lipid studies. Marcovina et al.[20] estimated the biological variability for 20 healthy adults for cholesterol, triglyceride, HDL, LDL, apo A, apo B, and Lp(a). Apo A's biological variability was about 6.2% and when its HDL counterpart's variability was 7.4%. We found HDL's biological variability to be 1.8% of an n=4 males. Marcovina Apo B's variability calculated to 7.4%, while LDL's is 9.5%. Our ApoB BV was calculated to be 3.7%, while LDL was respectively 5.5%. This information strongly suggests that apolipoproteins are less sensitive to environmental or physiological changes than their lipoprotein counter parts. Our biological variability was found to be lower than Marcovina et al. This could be attributed to closer attention to pre-testing procedures or lower measurement error. Since our CV was lower than Marcovina et al., this also indicates an effective matching on our part, finding homogenous sample.

EPOC

The average EPOC in terms of caloric expenditure for this study was 72 kcal. Townsend et al.[21] performed sprint intervals (SI) by doing three 30 second Wingate tests separated by 4 minutes passive recovery and compared this to a moderate aerobic

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exercise (MA) session of 30 minutes at 60% HRR. VO_2 during recovery for the SI was 7.5 L, while during MA reported 1.8 L. SI produced a recovery caloric expenditure of 37.5 kcal, and MA producing approximately 9.8 kcals. Variation among this previous literature compared to the present study was likely due to the difference of passive and active rest between sprint intervals. Energy expenditure during an active recovery is greater than passive recovery resulting in the greater observed energy expenditure.

Increases in VO_2 are greater with REHIT meaning a variety of physiological occurrences. We have an increase in the cost of glycogen resynthesis, dissipation of lactate, catecholamine induced increases in metabolism and lipolysis, a thermic effect of exercise and food, and increased muscle-protein turnover[22, 23, 24, 25, 26]. With increased intensity we see specifically a greater production of lactate and catecholamines, while decreases occur in pH, ATP, and phosphocreatine[22]. In EPOC, ATP regeneration is fueled by glucose and fatty acid catabolism. Most of glucose is being stored as glycogen meaning fatty acids are needed to fuel our other metabolic needs. The utilization of theses fatty acids is driving the changes we see in our lipids. Free fatty acids are hydrolyzed meaning LPL is increased and LPL hydrolyzes TG out of VLDL. LCAT as well increases, and this increase in energy utilization is contributing to changes in lipid count and size. This also demonstrates what we see with heightened EPOC, meaning an elevation in metabolism due to increased oxygen consumption to again help the body restore to pre-exercise levels. This elevation occurs because of a larger utilization of free fatty acids, increases in sympathetic activity, and an overall greater metabolic stress. Foureaux et al.[27] suggests that the oxidation rate of lipids post high intensity exercise is higher than moderate intensity exercise. Lipid oxidation is also associated with the increased turnover of free fatty acids contributing to a higher EPOC[28].

Advanced Lipids

HDLC is increased primarily via LCAT. Although traces of LCAT were not measured, previous research has suggested that RCT is a result of LCAT increasing during exercise and CTEP decreasing[2, 29, 30]. This is why we see HDL primarily being affected from exercise. HDLC baseline measured 38.6 mg/dL. After VT2 bout HDLC increased 41.25 mg/dL, and to 42.5 mg/dL post REHIT bout. This demonstrates immediate effects of exercise on HDLC. Although size or ApoA was not measured it is assumed that both increased with exercise, especially with REHIT. This can be assumed from the STRRIDE studies, because both interventions surpassed the intensity deemed beneficial to elicit positive changes. Kraus and colleagues demonstrated chronic effects of exercise on lipid size and count[10]. STRRIDE[10] reported HDL size increased (pre: 8.77, post: 8.89), as well as the concentration of HDL particles (pre: 44.3±2.9, post: 48.6±3.3). Although their study was chronic not acute, this

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suggests that it is possible for HDL size to increase with exercise. HDL is the primary player in reverse cholesterol transport, meaning cholesterol is being eliminated from our system.

Another study done by Varady et al.[7] showed that exercise not only significantly increased HDL cholesterol, but also revealed that HDL particle size changed from smaller (pre: 21 mg/dL, post: 14 mg/dL) and large (pre: 28 mg/dL, post: 34 mg/dL) particles. Although this study was a training study and not an acute study, it implies the importance of the need of acute studies to identify exact acute mechanisms and exercise bouts to ensure response chronically.

ApoB baseline was 126.8 mg/dL, while post VT2 ApoB was 121.5 mg/dL, and post REHIT measured at 115.5 mg/dL. ApoB is important because it is a measure of the total LDL particles, rather than the amount of cholesterol per all the particles. LDLC baseline measured 151.75 mg/dL, post VT2 was 144.5 mg/dL, and post REHIT 138.25 mg/dL. Again, these measures demonstrate acute changes from exercise. Looking at apoB versus LDLC the numbers are very different, and it is important for people and health care providers to know the differences between these measures. Triglycerides (TG) are extremely variable and a hard measure to control. However, to begin drawing conclusions on size we needed to measure LDLTG. Baseline LDLTG was 39 mg/dL and post VT2 bout was 35.5 mg/dL. Post REHIT was 41 mg/dL, actually increasing from baseline. This increase post REHIT was perhaps attributable to one of the subject's previous history of training, allowing his body to be more efficient in utilizing free fatty acids and storing glycogen, suggesting the increase in TG. Overall clinically significant changes were seen with the REHIT intervention when looking at LDL and ApoB.

Weise et al.[2] reported that following an exercise bout with an energy expenditure of 400 kcals, TG significantly decreased and stayed decreased up to 48 hours. HDLC significantly increased up to 24 hours post exercise intervention. Weise et al. [2] performed an acute study on females which differed from our male cohort. Weise and colleagues also reported a not statically significant decrease but rather clinically significant decrease in LDL (pre (108.5 mg/dL) to 48h post (105.2 mg/dL) exercise), TC (pre: 198.8 mg/dL, 48h: 193.8 mg/dL), and free cholesterol (FC) (pre: 61.2 mg/dL, immediately post exercise: 58.2 mg/dL). Weise also found that LCAT shows relatively resistant to acute changes with exercise. This could explain the lack of change seen in our cholesterol esters pre (74.87 mg/dL) to post VT2 (74.5 mg/dL) and finally to REHIT (74.75 mg/dL). LCAT again is the enzyme responsible for causing esters to the HDL particle. Thus, meaning we were hypothesizing to see a decrease in CE due to an increase in LCAT. A reason why it is thought we didn't find support with this hypothesis could be due to LCAT activity only being increased during exercise causing CE

to go to HDL explaining the increasing HDLC seen. Further research should explore immediately pre/post measures of CE and LCAT to help support this hypothesis.

Weise et al.[2] had an effect size from pre to post LDLC of 0.35, while their change in TC effect size calculates to 0.67. For our REHIT intervention our effect size for LDLC was 0.47 (greater than Weise and colleagues whose n=40) and TC was 0.44. Although TC's effect was less than Weise, we still had a positive medium correlation. Weise et al. total change for LDL was -2.5 mg/dL and change in TC was -5.2 mg/dL. Our VT2 intervention found the change of LDLC to be -7.3 mg/dL, and TC -8.6 mg/dL. To further reiterate the power of REHIT we saw LDLC change -13.6 mg/dL and TC change -12.63 mg/dL.

According to Silverman et al.[31], who conducted a meta-regression analysis, reported that a 38.7 mg/dL reduction in LDLC is associated with a decreased relative risk of major cardiovascular events. In a single bout of REHIT we report an average decrease of 13.6 mg/dL in LDLC, which is one third of a decrease in relative risk reduction from a single REHIT bout. "The Framingham study and others that followed showed HDLC as an independent cardiovascular risk factor and that an increase of only 10 mg/dL of HDLC leads to a risk reduction of 2-3%"[32]. We found that a single REHIT intervention increased HDLC 3.8 mg/dL, and a the single VT2 bout increased HDLC 2.6 mg/dL. This comparison demonstrates the ability of REHIT to sufficiently reduce risk of a cardiovascular event. It is possible that this 8 minute and 40 sec exercise bout could be key in combatting dyslipidemia and the progression of atherosclerosis.

Insulin

Insulin is peptide hormone that helps facilitate movement of glucose to maintain blood sugar. Insulin baseline measures were 11.5 mcIU/L, and post VT2 recorded to be 10.57 mcIU/L. Post REHIT an increase in insulin was seen 15.4 mcIU/L. A study done by Lithgow and Leggate[33]. reported that less than 24 hours after an acute bout of HIIT, insulin secretion increased, with highest marks seen 60 minutes post exercise. Following a single bout of HIIT or REHIT in this case, demonstrates that more insulin is secreted to increase the influx of glucose into the muscle to fuel ATP or to be converted to glycogen. Acutely the metabolic response following REHIT and HIIT is a secretion of insulin, implying elevated beta cell action[34]. Thus, the aim of acute intense exercise is to improve glucose load via insulin because of the glycogen depletion that occurred[33].

Berryman-Maciel[35] reports seeing an increase in insulin values in REHIT vs control post 3-week crossover training study. This study helps support the idea of long-term REHIT or HIIT exposure may induce beta cell function, increasing their capacity to respond when glucose loading. This increase in beta cell action, regardless of the presence of insulin resistance, could be

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beneficial in a clinical setting with regards to people obtaining MetS and type 2 diabetes.

Insulin's relationship to lipids

It is thought that insulin increased post REHIT due to a release of catecholamines which stimulate muscle glycogenolysis to fuel ATP, and hepatic glycogenolysis, increasing blood glucose. This glycogenolysis from catecholamines induces a prolonged lipolytic affect via growth hormone (perhaps explanation of improved apoB [36]. Since REHIT is short, the Ca+ stimulated glucose uptake doesn't last very long, and insulin is needed to bring glucose to homeostasis post exercise (hence insulin increases seen). Additionally, catecholamines stimulate HSL in adipose tissues and raise blood fatty acid levels. In trained individuals, the catecholamine dump is greater, so FA and glucose would be greater as well[37,25,38,39].

Tooth et al. [38] explains the role insulin resistance has on HDLC, apoB and LDLC very well. Individuals with dyslipidemia have an increase in triglycerides, leading to a decrease in VLDL triglyceride hydrolysis. This inhibits lipoprotein lipase (LPL). The ultimate consequence to the inhibition of LPL is that it results in an increase in VLDL's and TG. LPL eventually forms a dimer, angiopoietin-like protein 4, further decreasing its own activity. This causes less VLDL into LDL. People with this present with normal to low LDL levels. At this point metabolic changes transpire. CTEP exchanges TG out of a VLDL with a CE from HDL or LDL, this occurs at a 1:1 ratio[38]. This can be misleading if a normal lipid panel is used because you are measuring cholesterol per all particles, not necessarily the content of each particle. The increase in TG from the exchange lead to lipolysis via hepatic lipase (HL), causing HDL catabolism and the creation of sdLDL. SdLDL's have a decreased affinity to bind to LDL-R on the liver, dwindling systemic clearance. When the body experiences an insulin resistance, there is lower expression of ATP binding cassette proteins. This reduces the production of HDL, leading to liver insulin resistance, ultimately downregulating the apoA1 gene (the gene in charge of making apoA's, which eventually become HDL's).

The previous scenario is in the presence of insulin resistance. Exercise increases insulin sensitivity; so, what does this mean for individuals with dyslipidemia, like the one in this study? Insulin sensitivity is increased due to the depletion of glycogen stores, and it is known insulin sensitivity is positive for decreasing apoB and LDLC, as this study showed.

Future research should focus on the changes associated with lipid size and exercise, discussing the importance of lipid particle size. This study and others imply REHIT is a very effective time-efficient strategy for favorably altering the lipid profile[14,35]. Further research should also investigate the safety of the REHIT exercise bout in other disease-burdened populations (e.g., CVD). It is important to note that this is a preliminary study with an n of 4, suggesting that if this study was adequately powered differences

would be significantly different. The previous statement can be supported by the power of the effect sizes shown in table 5. Also, further research on this topic should account for biological variability, for it is a powerful way to confirm true changes are among lipids. Lastly, identifying the acute window of response regarding lipid changes and insulin would be advantageous to future researchers.

Limitations

Limitations to this study merit discussion. First, the sample size was small and therefore the findings should be interpreted as preliminary. Another limitation is the absence of a second blood measure further away from the intervention time to help identify true acute changes. An additional limitation is the lack of diet control. Participants were advised to keep their dietary patterns consistent based on initial screening, however this could've altered the data. Participants were advised to also fast at least 10-12 hours before screening and all other blood measures

however, the longer or shorter fasts could have also affected the outcomes.

Conclusions

This study concludes that REHIT is an effective exercise in altering lipids and increasing insulin. Increases in insulin help alter lipid metabolism, resulting in increases in HDLC, and decreases in LDLC, ApoB, and TC. Biological variability permitted increased confidence that changes in the key outcomes were attributable to the acute bouts of exercise rather than day-to- day variability and/or measurement error. RREHIT sufficiently reduces risk of a CVD, and it is possible that this 8 minute and 40 sec exercise bout could be key in combatting dyslipidemia and the progression of atherosclerosis.

Disclosures

This study was funded by Gunnison Valley Health (GVH). However, GVH was not involved in the design of this study, collection or analysis of the data, or the preparation of this manuscript.

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REFERENCES

1. Wenger, N. K. Preventative coronary interventions for women. Med Science Sports and Exercise: 28: 3-6 1996. DOI:10.1097/00005768-199601000-00002
2. Weise, S. D., Grandjean, P. W., Rohack, J. J., Womack, J. W., & Crouse, S. F. (2005). Acute changes in blood lipids and enzymes in postmenopausal women after exercise. Journal of Applied Physiology, 99,605-615. doi:10.1152/japplphysiol.01354.2004
3. Shaheen, H. A., Alpert, P. T., Navalta, J., Tandy, R. D., Young, J. C., & Santo, A. S. (2014). The effect of acute endurance exercise on lipoproteins: A comparison of the nuclear magnetic resonance technique with the conventional lipid profile in healthy men. Applied Physiology, Nutrition, and Metabolism, 39(2), 233–237. https://doi.org/10.1139/apnm-2013-0139
4. Freedman, D. S, Itvis, J. D., Jeyarajah, E. J., Barboriak, J. J., Anderson, A. J., and Walker, J. A. (1998). Relation of lipoprotein subclasses as measured by proton nuclear magnetic resonance spectroscopy to coronary heart

disease. Arteriosclerosis, Thrombosis, Vascular Biology. 18: 1046-1053. DOI:10.1161/01.atv.18.7.1046

5. Gerber, P. A, Nikoloc, D., and Rizzo, M. (2017). Small Dense LDL; an update. Current Opinion in Cardiology: 32(4): 454-459. doi: 10.1097/HCO.0000000000000410
6. The straight dope on cholesterol – Part VII - Peter Attia. (2019). Retrieved November 12, 2019, from https://peterattiamd.com/the-straight-dope-on-cholesterol-part-vii/
7. Varady, K. A., Bhutani, S., Klempel, M. C., & Kroeger, C. M. (2011). Comparison of effects of diet versus exercise weight loss regimens on LDL and HDL particle size in obese adults. Lipids in Health and Disease, 10(1), 119. https://doi.org/10.1186/1476-511X-10-119
8. The straight dope on cholesterol – Part V - Peter Attia. (n.d.). Retrieved November 12, 2019, from https://peterattiamd.com/the-straight-dope-on-cholesterol-part-v/
9. Wang, Y. and Xu, D. (2017). Effects of aerobic exercise on lipids and lipoproteins. *Lipids in Health and Disease,16(*132): 1–8. https://doi.org/10.1186/s12944-017-0515-5
10. Kraus W. E. et al. (2002). Effects of the amount and intensity if exercise on plasma lipoproteins. New England Journal of Medicine, 347(19): 1483-1492. DOI: 10.1056/NEJMoa020194
11. Bellou, E. et al. (2013). Effect of high-intensity interval exercise on basal triglyceride metabolism in non-obese men. Applied Physiology, Nutrition, and Metabolism, 38: 823-829. https://doi.org/10.1139/apnm-2012-0468
12. Fergison M.A., Alderson N. L., Trost S. G., et al. Effects of four different single exercise sessions on lipids, lipoproteins and lipoprotein lipase. J applied physiology 1998; 85 (3): 1169-74. DOI:10.1152/jappl.1998.85.3.1169
13. Weatherwax, R. M., Harris, N. K., Kilding, A.E., and Dalleck, L. C. (2018). Incidence of VO2max responders to personalized versus standardized exercise prescription. American College of Sports Medicine, 681-691. https://doi.org/10.1249/MSS.0000000000001842
14. Cuddy, T., Ramos, J., & Dalleck, L. (2019). Reduced Exertion High-Intensity Interval Training is More Effective at Improving Cardiorespiratory Fitness and Cardiometabolic Health than Traditional Moderate-Intensity Continuous Training. International Journal of Environmental Research and Public Health, 16(3), 483. https://doi.org/10.3390/ijerph16030483
15. Vollaard, N.B.J & Metcalfe, R.S. (2017). Research into the health benefits of sprint interval training should focus on protocols with fewer and shorter sprints. Sports Medicine, 47, 2443-2451. DOI:10.1007/s40279-017-0727-x
16. American College of Sports Medicine. (2013). *ACSM's guidelines for exercise testing and prescription*. Lippincott Williams & Wilkins.
17. Meltcalfe, R. S., Baraj, J. A., Fawkner, S.G., and Vallaard, N. B. (2012). Towards the minimal amount of exercise for improving metabolic health: Beneficial effects of reduced-exertion high-intensity interval training. European Journal of Applied Physiology, 112: 2676-2775. DOI:10.1007/s00421-011-2254-z
18. Meltcalfe, R. S., Tardif, N., Thompson, D., Vollaard, N. B. (2016). Changes in aerobic capacity and glycemic control in response to reduced-exertion high-intensity interval training (REHIT) are not different between sedentary men and women. Applied Physiology of Nutrition and Metabolism, 41: 1117-1123. DOI:10.1007/s00421-015-3217-6
19. Frank, A. T., Zhao, B., Jose, P. O., Azar, K. M., Fortmann, S. P., & Palaniappan, L. P. (2014). Racial/Ethnic Differences in Dyslipidemia Patterns. Circulation, 129(5), 570–579. https://doi.org/10.1161/CIRCULATIONAHA.113.005757
20. Marcovina, S. M., Guar, V. P., and Albers, J. J. (1994). Biological Variability of Cholesterol, Triglyceride, Low- and High-Density Lipoprotein Cholesterol, Lipoprotein(a), and Apolipoproteins A-1 and B. Clinical Chemistry, 40(4): 574-578.
21. Townsend, J. R., Stout, J. R., Morton, A. B., Jajtner, A. R., Gonzalez, A. M., Wells, A. J., Mangine, G. T., McCormack, W. P., Emerson, N. S., Iv, E. H. R., Hoffman, J. R., Fragala, M. S., & Cosio-Lima, L. (2013). Excess Post-Exercise Oxygen Consumption (Epoc) Following Multiple Effort Sprint and Moderate Aerobic Exercise. 8. https://stars.library.ucf.edu/facultybib2010/4767
22. Hazell, T. J., Olver, D., Hamilton, C. D., & Lemon, P. W.R. (2012). Two Minutes of Sprint-Interval Exercise Elicits 24-hr Oxygen Consumption Similar to That of 30 min of Continuous Endurance Exercise. International Journal of Sport Nutrition and Exercise Metabolism 22, 276-283.
23. Moniz, S. C., Islam, H., & Hazell, T. J. (2020). Mechanistic and methodological perspectives on the impact of intense interval training on post-exercise metabolism. Scandinavian Journal of Medicine & Science in Sports, 30(4), 638–651. https://doi.org/10.1111/sms.13610
24. Malatesta, D., Werlen, C., Bulfaro, S., Chenevière, X., & Borrani, F. (2009). Effect of High-Intensity Interval Exercise on Lipid Oxidation during Postexercise Recovery: Medicine & Science in Sports & Exercise, 41(2), 364–374. https://doi.org/10.1249/MSS.0b013e3181857edo

25. Bahr, R. & Sejersted, O. M (1991). Effect of intensity of exercise on excess post exercise O2 consumption. Metabolism 40(8), 836-841. DOI:https://doi.org/10.1016/0026- 0495(91)90012-L
26. de Souza, J. F. T., Dáttilo, M., de Mello, M. T., Tufik, S., & Antunes, H. K. M. (2017). High-Intensity Interval Training Attenuates Insulin Resistance Induced by Sleep Deprivation in Healthy Males. Frontiers in Physiology, 8, 992. https://doi.org/10.3389/fphys.2017.00992
27. Foureaux, G. (2006). Effects of excess post-exercise oxygen consumption and resting metabolic rate in energetic cost. Rev Bras Med Esporte, 12, 5. https://doi.org/10.1590/S1517-86922006000600018
28. Hunter, G., Weinsier, R., Bamman, M., & Larson, D. (1998). A role for high intensity exercise on energy balance and weight control. International Journal of Obesity, 22(6), 489–493. https://doi.org/10.1038/sj.ijo.0800629
29. Grandjean, P. W., Crouse, S. F., & Rohack, J. J. (2000). Influence of cholesterol status on blood lipid and lipoprotein enzyme responses to aerobic exercise. Journal of Applied Physiology, 89:472-480. DOI:10.1152/jappl.2000.89.2.472
30. Mann, S., Beedie, C., and Jimenez, A. (2014). Differential effect of aerobic, resistance training, and combined exercise modalities on cholesterol and the lipid profile: Review, synthesis and recommendations. Sports Medicine, 44: 211-221. DOI:0.1007/s40279-013-0110-5
31. Silverman, M. G., Ference, B. A., Im, K., Wiviott, S. D., Giugliano, R. P., Grundy, S. M., Braunwald, E., & Sabatine, M. S. (2016). Association Between Lowering LDL-C and Cardiovascular Risk Reduction Among Different Therapeutic Interventions: A Systematic Review and Meta-analysis. JAMA, 316(12), 1289. https://doi.org/10.1001/jama.2016.13985
32. Ali, K. M., Wonnerth, A., Huber, K., & Wojta, J. (2012). Cardiovascular disease risk reduction by raising HDL cholesterol – current therapies and future opportunities. British Journal of Pharmacology, 167(6), 1177–1194. https://doi.org/10.1111/j.1476-5381.2012.02081.x
33. LITHGOW, H. M., & LEGGATE, M. (2018). The Effect of a Single Bout of High Intensity Intermittent Exercise on Glucose Tolerance in Non-diabetic Older Adults. International Journal of Exercise Science, 11(3), 95–105. PMID: 29795727
34. Madsen, S. M., Thorup, A. C., Overgaard, K., & Jeppesen, P. B. (2015). High Intensity Interval Training Improves Glycaemic Control and Pancreatic β Cell Function of Type 2 Diabetes Patients. PLoS ONE, 10(8). https://doi.org/10.1371/journal.pone.0133286
35. Berryman-Maciel, M. J., Yeung, L. L., Negley, L., Buchanan, C. A., & Dalleck, L. C. (2019). Can reduced-exertion, high-intensity interval training combats the deleterious cardiometabolic effects of a sedentary lifestyle? 20.
36. A Sjöberg, J Oscarsson, K Boström, T L Innerarity, S Edén, S O Olofsson, Effects of growth hormone on apolipoprotein-B (apoB) messenger ribonucleic acid editing, and apoB 48 and apoB 100 synthesis and secretion in the rat liver., Endocrinology 130(6), 3356-3364, https://doi.org/10.1210/endo.130.6.1597147
37. Cartee, G. D. (2015). Mechanisms for greater insulin-stimulated glucose uptake in normal and insulin-resistant skeletal muscle after acute exercise. American Journal of Physiology-Endocrinology and Metabolism, 309(12), E949–E959. https://doi.org/10.1152/ajpendo.00416.2015
38. Toth, P. P. (2014). Insulin Resistance, Small LDL Particles, and Risk for Atherosclerotic Disease. Current Vascular Pharmacology, 12, 653-657. PMID:23627975
39. Arner, P. (1999). Catecholamine-induced lipolysis in obesity. International Journal of Obesity, 23(S1), S10–S13. https://doi.org/10.1038/sj.ijo.0800789