Exercise and Glycemic Control in Individuals with Type 2 Diabetes

A Thesis Presented to the Faculty of the Graduate School at the University of Missouri

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The undersigned, appointed by the dean of the Graduate School, have examined the thesis entitled

EXERCISE AND GLYCEMIC CONTROL IN INDIVIDUALS WITH TYPE 2 DIABETES

presented by Douglas J. Oberlin II,

a candidate for the degree of master of science,

and hereby certify that, in their opinion, it is worthy of acceptance.

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I dedicate this thesis to:

My Family

It is thanks to the love and support of my family that I am able to be here. They have worked hard to provide me with opportunities to pursue my education and attempt to better myself. I am very thankful for all that they have sacrificed and all of their hard work to provide me the option to go to college and pursue higher education. Through all of my endeavors, they have supported and encouraged me. Thank you, to all of my family who has worked so hard to allow me to have the ability to accomplish this!

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Glycemic Control in Individuals with Type 2 Diabetes

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Abstract

Type 2 diabetes (T2D) and the associated impaired glycemic control greatly increases the risk of cardiovascular disease mortality. **PURPOSE:** Our lab previously has shown that five to seven consecutive days of aerobic exercise can effectively reduce the change in post-prandial glucose levels (ΔPPG; = post-meal glucose level – pre-meal glucose level) in previously sedentary individuals with T2D measured by continuous glucose monitors (CGMS). It is unknown if or for how long a single bout of exercise will reduce ΔPPG in individuals with T2D. **METHODS:** We recruited 9 individuals with T2D (BMI: 36 ± 1.9 kg/m^2; age 60 ± 1 years; HbA1c: 6.3 ± 0.2 %) who were not using exogenous insulin and sedentary (<30 minutes/week of exercise and less than 6,000 steps). The subjects consumed a eucaloric diet (51% carbohydrate, 31% fat, 18% protein) containing identical food components at each meal during two separate 3 day trials while wearing CGMS monitors to continually monitor blood glucose levels. During one 3 day trial the subjects performed one 60 minute, supervised exercise bout (exercised; 60% of heart rate reserve) prior to breakfast on the morning of the first day. During the second 3 day trial, the subjects maintained their sedentary lifestyle (sedentary). The order of the sedentary and exercised trials was randomly assigned. **RESULTS:** A comparison of the 2 trials revealed that one bout of exercise did significantly reduced ΔPPG at the 240 minute time point post-meal (averaged across all meals) in the exercised phase (p=0.003). A comparison of post-prandial glucose levels

(PPG) at the different post-prandial time points between phases showed lower PPG during the exercised phase at time points 120, 150, and 240 ($p=0.03$, $p=0.05$, and $p=0.01$ respectively). This was most likely driven by the first day's significant reduction in average PPG for meal 2 and 3 ($p=0.04$ and $p=0.03$ respectively). There was also a decrease in 24 hour average blood glucose level for the first day after the exercise bout ($p=0.003$). **CONCLUSION:** These results suggest that one moderate-intensity bout of aerobic exercise is effective in significantly improving glycemic control in subjects with T2D, however the improvement only seemed to last for a single day.

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Introduction:

Currently in America there is a health crisis stemming from the number of Americans who are overweight and obese (3, 56). Overweight and obese individuals are at a higher risk for many chronic diseases including type 2 diabetes mellitus (T2D), cardiovascular disease (CVD), and the metabolic syndrome (1, 6, 26, 56). Of these diseases, CVD in particular is the leading cause of death in America and 80% of individuals with T2D will die from CVD (8, 23, 63). Post challenge glucose levels, measured with an oral glucose tolerance test (OGTT) or following a mixed meal, positively correlates with risk of CVD death as well as all cause mortality in the general population and in those with T2D (49, 62). Post-prandial hyperglycemia, as well as large fluctuation in glucose levels, likely induce oxidative stress and inflammation which exacerbate the symptoms of CVD, T2D, and the metabolic syndrome (10, 15, 16, 23, 44, 49, 63).

Change in post-prandial glucose levels (PPG) following a mixed meal parallel the glycemic excursions seen with an OGTT, but have a slightly lower peak value (62). As the impairment of glucose tolerance progresses towards disease, peak glucose levels from OGTT, as well as PPG, increase (59, 62). Individuals with impaired glucose tolerance or T2D may experience PPGs which are higher than the OGTT peak glucose levels in healthy individuals, further deteriorating their condition (62). Individuals with T2D are also prone to having hyperglycemic excursions overnight while they are sleeping and in the early morning (22, 43). Increased hyperglycemic excursions can lead to increased glycated hemoglobin (HbA1c) which also correlates with risk of disease and mortality,

and is used as a marker of glycemic control in individuals with T2D (1, 5, 14, 25, 34, 40, 43, 47, 60). For all these reasons it is important to find more effective treatments for lowering PPG as well as fasting hyperglycemia.

There are multiple strategies for controlling blood glucose levels in individuals with T2D. People suffering from T2D usually take oral anti-hyperglycemic medications in the form of either a medication to enhance insulin secretion from the pancreas or a medication to improve insulin action in metabolically active tissues (muscle and liver) (50). While medications can acutely reduce average blood glucose levels and improve HbA1c, they do not stop metabolic dysfunction from progressing over time (50, 54). Another strategy for controlling blood glucose levels is through increasing physical activity (26, 51, 55, 56). Previous studies have shown improvements in fasting blood glucose levels, average 24 hour blood glucose level, as well as post-prandial glycemic response after moderate intensity exercise training (27, 35, 39, 53). The ADA's first line of defense is Metformin (a medication to improve insulin sensitivity) and lifestyle intervention.

Exercise's effect on Post-prandial Glucose Levels

Currently, the American Diabetes Association (ADA) recommends 150 minutes of moderate intensity exercise (40-60% VO_{2peak}) per week, or 60 minutes of vigorous intensity exercise (>60% VO_{2peak}) per week for individuals with T2D (1). The ADA also recommends that a person should not go more than two consecutive days without

exercise, and that a single bout last no less than 10 minutes (1). However, the frequency or volume of exercise to control PPG may need to be higher in this population (4, 37, 41).

Physical activity can reduce PPG by increasing insulin and non-insulin dependent glucose uptake into skeletal muscle (4, 11, 17, 18, 26, 31, 41, 48, 57, 58, 61). This effect seems to be specific to the muscles used during exercise, due to an exercise induced disruption in energy balance at the cellular level (7, 46). One proposed mechanism by which glucose uptake is improved following exercise is by increasing the AMP to ATP ratio which stimulates AMPK leading to downstream translocation of GLUT 4, a glucose transport protein (12, 27, 29, 48, 53). The amount of glycogen breakdown that occurs with exercise, and a subsequent drive to replenish glycogen stores, has also been tied to the degree to which post-exercise insulin sensitivity is improved. Importantly, the ability of exercise to improve glucose uptake has been observed in both diseased animal models as well as in individuals with T2D (31, 39, 56). There are both acute as well as chronic adaptations to exercise which can lead to reduced PPG (48). With chronic exercise there can be increases in concentrations of GLUT 4 in the muscle cells, increased mitochondrial enzymes, increased capillarization, as well as increases in the number of mitochondria in the cell (19, 30, 33, 48, 52).

There is also an improvement in insulin stimulated glucose transport for a certain amount of time after exercise, but the exact mechanism for this effect remains unknown (17, 29, 56). The improvements in glucose transport after exercise can be seen in Figure 1 which shows increased glucose transport in an acutely exercised leg versus an unexercised leg at various insulin doses, indicating improved glucose tolerance post-

exercise in working muscles. It has also been shown that acute exercise increases insulin stimulated glucose transport in insulin resistant muscle for humans and rodent models (17, 26, 56). Each exercise bout produces an acute response of improved insulin signaling and improved glucose uptake due to increased translocation of GLUT 4 proteins (29). Although the acute effect of exercise on improving skeletal muscle insulin sensitivity has been estimated to last as long as 48 hours to 5 days in healthy people or 20 to 24 hours in individuals with T2D, the exact duration of the effect of exercise on PPG has not been measured (27, 29, 41).

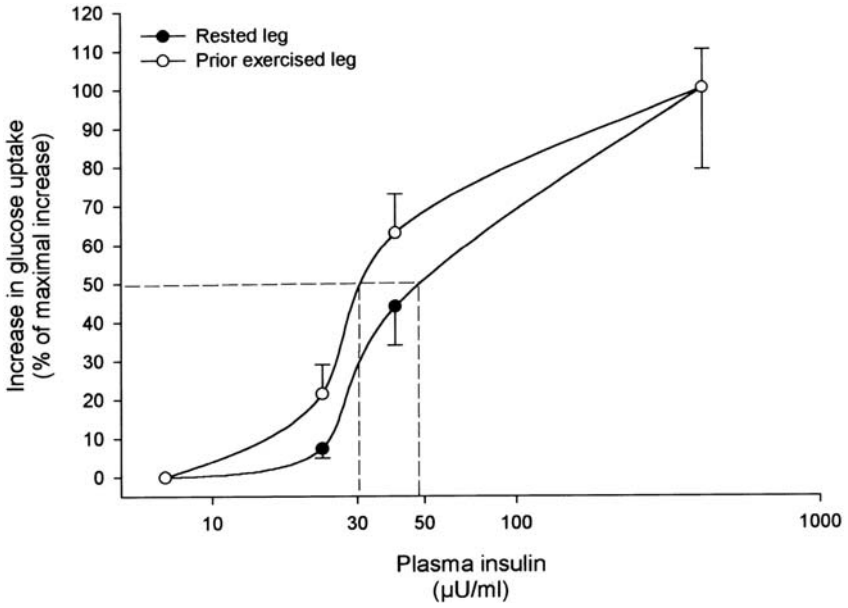


Figure 1. Glucose uptake in response to insulin in a rested vs. exercised leg. This figure by Wojtaszewski et al 2003 (61) with data from Richter et al 1989 (46) shows how a limb that recently underwent acute exercise training takes up glucose at a higher rate in response to the same amount of insulin relative to an unexercised limb.

The duration of exercise's acute effect specifically on PPG has not been measured due to limitations in available measurement methods. Most studies use either OGTT, an intravenous glucose tolerance test (IVGTT), or the hyperinsulinemic euglycemic clamp to measure glucose tolerance or insulin sensitivity (4, 9, 11, 21, 31, 41, 46, 53). Although these methods are useful for assessing insulin sensitivity and even estimating PPG, none of them measure PPG directly in a free living condition, because they do not assess mixed meals within the normal living environment. A better measurement tool for assessing PPG over several days is the continuous glucose monitoring system (CGMS) combined with controlled dietary intake (24).

CGMS is a small device with a probe inserted just beneath the skin that measures minute to minute glucose levels in the interstitial fluid (which equilibrates with circulating glucose levels) and can be worn in free living individuals for days at a time. Thus, the device can be used to measure glycemic control in free living individuals eating mixed meals (25, 39). These monitors have been used to successfully measure changes in PPG in response to exercise by other labs (24, 39). However, to our knowledge, no one has used CGMS to quantify how long one exercise bout improves PPG in individuals with T2D. Because CGMS is a better method of measuring PPG, and PPG correlates strongly with mortality, we will use the CGMS to determine the duration of exercise induced reduction in PPG in individuals with T2D consuming a study diet following a single exercise bout.

Our hypothesis is that PPG will be significantly lowered for only two meals, breakfast and lunch, following a morning exercise bout; and that glycemic responses to

subsequent meals will no longer be reduced compared to sedentary. Even though studies using the hyperinsulinemic-euglycemic clamp have shown improvements in insulin sensitivity for up to 48 hours, we expect PPG to be improved for only 2 meals. This is because carbohydrate consumption after exercise putatively replenishes depleted glycogen stores in muscle and thus blunts exercise induced improvement in insulin sensitivity. Furthermore, the CGMS is not able to measure very small changes in insulin sensitivity that can be detected using the hyperinsulinemic-euglycemic clamp because it only samples blood glucose levels under physiological conditions (7, 9, 21, 29, 41). We also hypothesize that the average overnight blood glucose level will be lower for the first night after following the exercise session compared to the sedentary phase. This may be due to increased insulin sensitivity in the liver (which is what is primarily being assessed with any fasting measure of glucose level); however hepatic insulin sensitivity will not be directly measured. Thus, the two aims for this study are:

1) To determine if a single bout of exercise reduces PPG in a subsequent meal, and to determine how long this effect persists in individuals with T2D.

2) To determine whether there is any significant change in overnight fasting blood glucose levels after the exercise bout.

Methods:

Subjects

Sedentary individuals with T2D were recruited from the city of Columbia, MO. Sedentary was defined as subjects who on average took less than 6000 steps per day and did not participate in any formal exercise program (>30 minutes of planned exercise 2 times a week). Two subjects with higher step counts were allowed; one due to participation in a previous study in which they had fewer steps, and the other was allowed because his steps were elevated due to work on the days the pedometer was worn. Both subjects had a lower number of steps through the study. The subjects were non smokers with a BMI between 30 and 42 kg/m^2 who were able to exercise safely on a treadmill and stationary bike. They were weight stable (±5%) and medication stable for at least 3 months before entering the study. In addition, the subjects had controlled diabetes with HbA1c< 7.5% with no insulin use, and no advanced retinopathy or neuropathy. Other exclusion criteria included pregnancy, sleep perturbations, night shift workers, or people who have recently traveled across more than two time zones, or individuals with irregular daily schedules. All subjects signed an informed consent which was approved by the University of Missouri Institutional Review Board.

After the consent meeting the subjects came to the exercise physiology lab during the morning for a baseline testing meeting where their height, weight, and blood pressure were measured. A fasting blood sample was also taken for measurement of glycated hemoglobin (HbA1c, a measure of long term average glucose levels), fasting blood glucose levels, blood lipids (total cholesterol, LDL, HDL, and Triglycerides), and a

complete metabolic panel. The HbA1c was run in the University of Missouri Exercise Physiology chemistry lab on a Siemens DCA Vantage analyzer using blood drawn in a heparin tube. The other blood tests were run by Boyce and Bynum pathology laboratory using blood drawn in an SST tube. The tests run were, a complete metabolic panel (Glucose, Bun, Creatine, Sodium, Potassium, Chloride, Carbon dioxide, Calcium, Total protein, Albumin, Alkaline phosphatase, Total bilirubin, AST, ALT, and eGFR) and a lipid panel (Cholesterol, Triglycerides, HDL, Total cholesterol:HDL ratio, LDL, LDL:HDL ratio, and Phenotype). After the baseline testing session, the subjects were given a diet log, and a pedometer to use over the next three days. This allowed us to measure the normal amount of physical activity (daily steps) the subjects performed as well as the typical caloric consumption and composition of the diet. Finally, on another visit the subjects had their body composition estimated using a duel energy x-ray absorptiometry (DEXA). The DEXA model used was a Hologic QDR 4500A Fan Beam X-Ray Bone Densitometer, and a whole body scan was used to measure body composition. The subjects then performed an exercise stress test to determine their maximal oxygen consumption (VO_{2peak}), their maximal heart rate, and to screen for any potential cardiac abnormalities with an EKG. The exercise stress test was performed on a treadmill using a Bruce protocol. During the test the subjects respiratory gases were measured by a metabolic cart (Parvo Medics True One 2400 Metabolic Measurement System), and cardiovascular function monitored by a 12 lead EKG (Quinton Qstress v3.5 Exercise Test Monitor). A physician was present to monitor EKG readouts during every exercise stress test. Criteria for a maximal test were two of the following: perceived

exertion of 17 or greater, respiratory exchange ratio of greater than 1.0, or a leveling off or slight decrease in oxygen consumption. The EKG data from each exercise stress test was reviewed by a cardiologist to ensure that the participants could safely participate in an exercise session. There was a five to fifteen day washout after the VO_{2peak} test before the subjects began the study protocol.

Study Design

The study design consisted of a sedentary measuring phase and an exercised measuring phase for all subjects. Therefore, the subjects served as their own control group. The subjects were randomized as to which phase they received first (the sedentary or exercised phase). During each phase the subjects consumed a study diet for five days. The first two days of the diet were to acclimate the subject to the new diet. The following 3 days of the standard diet coincided with the 3 day measurement period. The study design is shown with the two, five-day periods drawn in parallel in Figure 2 (shown below). During the sedentary phase the subjects continued their typical (sedentary) physical activity, which was verified using a Walk 4 Life Duo pedometer and an accelerometer (Body Media Sense Wear armband body monitoring system). A Medtronic iPro CGMS monitor was attached to the subject's abdomen with a probe inserted beneath the skin, and the monitor was attached and taped down with Smith & Nephew IV3000 adhesive pads the night before the first measurement day The CGMS was then worn for three consecutive days being removed on the fourth day. While the CGMS was worn, the subjects recorded four blood glucose levels with an Accu-Chek Compact Plus glucometer. The blood glucose data was later used to calibrate the CGMS

which measured blood glucose data each minute of the day (waking period) and night (sleeping period). After the first phase there was a five to fifteen day washout period during which the subjects continued their typical physical activity and consumed an ad libitum diet. Once the washout period ended, the subjects began the other phase (which ever they did first determined which would be second) of the study. The exercised phase was identical in all procedures to the sedentary phase of the study, except that the subjects performed one 60 minute bout of exercise prior to breakfast on the first CGMS measurement day.

Exercise Session

The exercise bout consisted of 60 minutes of aerobic exercise broken into three 20-minute sections starting at approximately 6:30 AM. This included 20 minutes on a treadmill, 20 minutes on a stationary cycle, and another 20 minutes on a treadmill. The exercise intensity was within five beats per minute of 60% of HRR for the duration of the exercise bout (as determined from a previous graded exercise stress test). After the exercise bout, the speed and grade (or RPMs and Watts for the cycle) were used to calculate Mets for determining the percent of aerobic capacity at which the subjects had been working. Intensity was adjusted during the exercise session by adjusting speed or grade on treadmill or adjusting resistance on the stationary cycle, to maintain the target heart rate throughout the entire exercise bout. This intensity and duration of exercise falls within the recommendations of the ADA and ACSM which recommend 150 minutes per week at an intensity of 40-60% VO_{2peak} (1). In addition, this exercise prescription was used in a previous study from our laboratory which measured a decrease in PPG after

seven days of exercise. As shown in Figure 3, our previous study using seven days of exercise at this prescription reduced post prandial PPG after meals as measured by CGMS. Thus, in this study we wanted to determine if and how long one bout of exercise prescribed at the same intensity and duration would have upon postprandial glycemic responses.

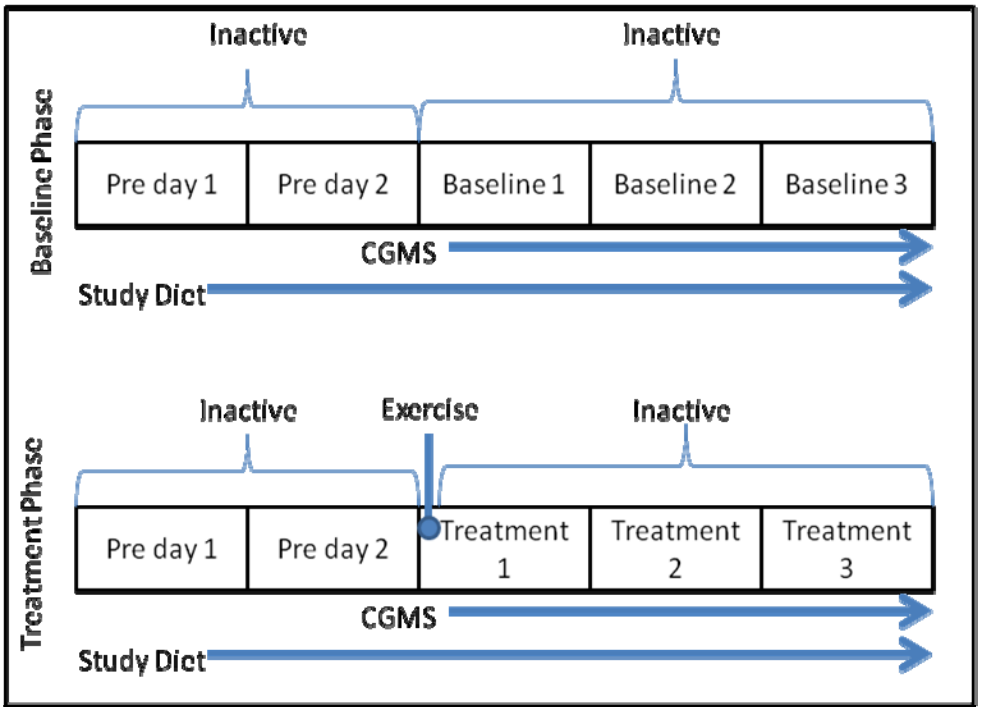


Figure 2. Study design for the sedentary vs. exercised study phases. This figure shows the baseline period (sedentary) of inactivity with the CGMS monitor being attached at the end of the second day and worn through the next three days. During the treatment period (Exercised), the CGMS is attached the night before a 60 minute exercise session and then worn through the next three days. The control study diet is eaten through both phases.

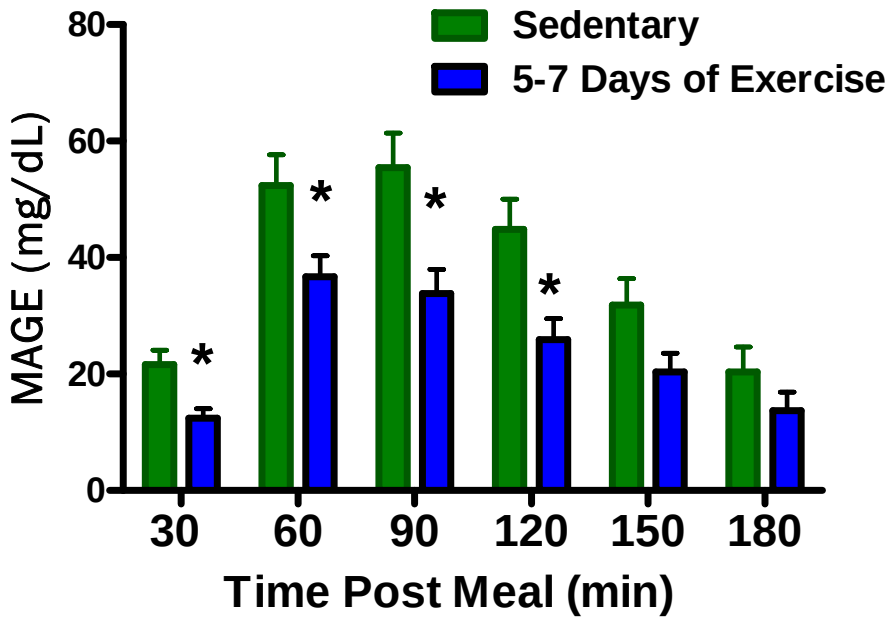


Figure 3. Change in post-prandial glucose levels in individuals with type 2 diabetes during either a sedentary condition, or after 5 to 7 days of exercise. This figure shows the 30, 60, 90, 120, 150, and 180 minute PPG (listed as MAGE) for subjects averaged across 3 days of sedentary acivity, and averaged across the 5th, 6st, and 7th day of a 7 day exercise program, measured with CGMS. The 5 to 7 days of exercise was effective at reducing the post-prandial glycemic response in individuals with T2D.

Study Diet

During the study, the subjects consumed a control study diet which can be seen in Table 1. The diet was prepared by study staff and was packed out for the subjects to eat during both the sedentary and exercised phases. The subjects were instructed to eat the meals at the same times each day and allow 5 hours between meals. Every meal had the exact same nutrient composition, caloric content, and contained the exact same food items prepared as breakfast, lunch, or dinner. Breakfast was a potato hash with seasoned ground beef topped with salsa and cheese served with buttered toast, applesauce and a juice drink. Lunch was mini cheeseburgers with salsa mixed into the patties and baked french fries served with a side of apple sauce and a juice drink. Dinner was a mini-meatloaf with salsa and cheese baked in and mashed potatoes served with garlic toast, applesauce, and a juice drink.

The macronutrient distribution was 51.4% carbohydrate, 30.9% fat, and 17.8% protein for the total energy content of the meal. The study diet met the DRI for all micronutrients except: Vitamins A, B1, B2, D, E, K, Biotin, Folate, Pantothenic Acid, Calcium, Copper, Fluorine, Iodide, Chromium, Magnesium, Manganese, Potassium, and Selenium (see Tables 1 and 2 in Appendix B). The glycemic load of each meal was approximately 46. The daily energy requirement was estimated for each subject using the Harris-Benedict equation and verified using a three-day dietary record filled out by the subject. The three day average was then averaged with the Harris-Benedict estimate to determine their individual energy requirements. From this information the subjects were provided a diet containing 1600, 1800, 2000, 2200, or 2400 kcals per day, whichever kcal

level was within100 kcals of their predicted requirements. For example, if a person was estimated at 2063 kcals, they would receive the 2000 kcal diet. However, if they were estimated at 2115 they would receive the 2200 kcal diet. Overall, the diet was designed to simulate a typical American diet, and provided consistent diet composition between meals and between subjects, and not meant to serve as a treatment or alteration from their individual normal dietary routine.

The subjects were given a log sheet to track when they ate their meals. They were instructed to eat all of the food provided for each meal. The meals were to be consumed at least 5 hours apart, and all meals were to be consumed at the same time for each day of the study. The subjects were also instructed to consume the meal within the timeframe of 15 to 20 minutes. In addition, the subjects noted when they went to bed at night and when they got up in the morning.

Glycemic Control

Post-prandial glucose levels (PPG) as well as peak glucose levels were measured from the CGMS output. The peak glucose level is simply the highest level of blood glucose which is achieved after each meal. The PPG was calculated at 15 minute intervals for four hours post-prandially at every meal. Delta PPG (ΔPPG) is the glucose level value at the start of the meal (time point 0) subtracted from the glucose level at 15 min increments after the start of the meal (measures the change from pre-meal glucose level). We also measured the area under the curve (AUC) for each post-prandial period, as well as the 2-hour post-prandial glucose level because it has been shown to be

predictive of cardiovascular events (13). Overnight (sleeping period) fasting glucose levels were measured by peak glucose level, minimum glucose level, and average glucose level measured between the times of going to bed and getting up in the morning. We also examined glucose levels by time spent within, above or below the range of 3.9 to 10.0 mmol/l

Food item	Amount in meal (g)	Kcals	CHO (g)	Fat (g)	Protein (g)
Great Value white sandwich bread	52.00	137.00	28.00	1.00	4.00
Idaho potatoes	140.00	105.95	24.59	0.00	1.89
salsa, mild, Great Value	33.00	8.00	2.00	0.00	0.00
ground beef 93/7	101.00	147.89	0.00	7.21	20.74
salted butter, light, Land o Lakes	14.00	45.00	0.00	5.00	0.00
olive oil, extra light, Great Value	4.75	42.00	0.00	4.67	0.00
Applesauce, Great Value, original	98.00	68.44	17.11	0.00	0.00
Cheese, Kraft medium cheddar	14.00	57.00	0.00	5.00	3.00
Juicy Juice, punch	125.00	56.00	14.00	0.00	0.00
Totals	**581.75**	**667.28**	**85.70**	**22.88**	**29.63**

Table 1. Foods and quantities in the study diet for a 2000 kcal/per day diet. This table shows the amount of each food item in one meal for the study diet at the 2000 kcal level. The amount of food was adjusted for each calorie level to achieve 200 kcal differences.

Statistical analysis

We used SPSS and Sigmastat software to perform the statistical analysis of our data. For statistical analysis of the 2 h glucose level at each meal, the glucose AUC response to each meal, and the day to day average PPG, a two way repeated measures ANOVA was run using Sigmastat software. The level of statistical significance was set at a P value of 0.05 with the main effects to be tested being: meal and phase for the 2 hour glucose level and AUC, and phase and day for day-to-day average PPG. Phase compared sedentary and exercised phases, and meal compared breakfast, lunch and dinner across the three day period (breakfast day 1, lunch day 1, dinner day 1, breakfast day 2, etc.). Day compared between days 1, 2, and 3, where all meals PPG in each day were averaged together. The two way repeated measures ANOVA also tested for interaction of meal x phase or phase x day. For overnight glucose measures, meal-to-meal PPG, post-prandial time points for PPG and delta PPG, and average glucose levels, paired T-tests were used to compare means, because subjects served as their own controls. Statistical significance was set at $P<0.05$ and all data are expressed as means ± standard error.

Results:

Fourteen subjects were recruited for the study and ten completed the protocol. Subjects who did not complete the study did not have their data included in analysis. Reasons for not completing the study were: not adhering to the study diet, too much baseline activity to be matched with the rest of the study group, loss of interest in the study, and health problems which made participation in the study ill-advised. One of the ten subjects who completed the study was later excluded from data analysis because their HbA1c was greater than 7.5 and greater than 2 standard deviations above the other subjects. The baseline anthropometric characteristics of the nine subjects who completed the study are shown in Table 2. The pre-study blood chemistry measures as well as the aerobic capacity of the subjects are shown in Table 3.

The exercise sessions were supervised by a lab member at all times, and the intensity of the exercise was adjusted according to both the subjects' heart rates as well as what the subjects were able to tolerate. The subjects, as a whole, found it difficult to perform exercise for 60 minutes at the intensity of 60% HRR. In the event of a subject feeling too fatigued to continue, the intensity was reduced and the subjects were encouraged to continue until 60 minutes of exercise had been completed. All subjects performed 60 minutes of exercise, and average percent of HRR for all subjects was 58% which was higher than our lab's previous study which showed improvement in glycemic control after 5 days (42). The average exercise data for all subjects is shown in Table 4.

The subjects had a statistically significant increase in both steps and energy expenditure on the day of the exercise session ($p=0.007$ and $p=0.005$ respectively),

however, there was no statistically significant difference between phases on the following days. This shows that the exercise session was effective at increasing the physical activity and energy expenditure of the subjects for one day allowing for a comparison of the two phases after an acute exercise bout.

Glycemic Conrol

The post-prandial glucose level was examined as an area under the curve (AUC) for each meal as well as 2 hour post-meal glucose level, shown in figure 5. There were no statistically significantly lower 2 hour glucose levels by meal in the exercised phase, however there was a statistically significantly lower 2 hour glucose level for the entire exercised phase compared to the sedentary phase ($p=0.03$). When run as a repeated measure ANOVA, there was a statistically significant ($p<0.1$) interaction between the phase and meals for glucose AUC. There was a statistically significant difference seen in AUC for meal 2 and for the entire exercised phase compared to the sedentary phase ($p=0.04$ and $p=0.01$ respectively).

Post-prandial glucose level was measured both as an absolute glucose level measure (PPG) as well as a relative measure of change from pre-meal glucose level (ΔPPG). The PPG between phases was significantly lower during the exercised versus the sedentary phase only at 120, 150, and 240 minute time points averaged across all meals for each phase ($p=0.03$, $p=0.05$, and $p=0.01$ respectively). The ΔPPG was also statistically significantly lower during the exercised phase, but only at the 240 minute time point averaged across all meals for each phase ($p=0.004$). The PPG was also compared as means by either meal to meal between phases or day to day. The second

and third meal had statistically significantly lower average PPG during the exercised phase compared to the sedentary phase as seen in Figure 6. The day-to-day comparisons, however, had no statistical significance.

There was a statistically significant improvement in average glucose level the first 24 hour period in which the exercise session occurred ($p=0.003$), but not for the following days. There was a statistically significant reduction in daytime average glucose level for the first waking period ($p=0.04$) during which the exercise session occurred. However there was no statistically significant difference in average blood glucose levels for the following overnight period, or any of the subsequent days or nights as shown in table 6.

The Medtronic CGMS Software allows us to analyze the percent of recorded time spent within pre-set glucose level ranges. We compared the days and phases for the percent of time spent below 3.9 mmol/l, above 10.0 mmol/l or within this range (2). There were no statistically significant differences in the time spent within these limits across days or between phases as seen in Table 7.

In addition to the average post-prandial glucose level data, the post-prandial peak glucose level was compared for each meal between the two phases and across all meals between the two phases. There was no statistically significant difference between the post-prandial peak glucose levels between phases. There was only a statistically significant difference in post-prandial peak glucose levels at meal 2 ($P=0.03$) within the meals between the two phases.

The overnight glucose levels were also assessed during this study, and were measured over time as well as average, peak and nadir during sleeping hours. There were no statistically significant differences found between phases or individual nights for average glucose levels, peak glucose level or nadir.

Subject Anthropometric Characteristics	
Age (years)	60.3 ± 1.0
Sex	5 Females / 4 Males
BMI	36.0 ± 1.1
Body Fat Percentage	39.6 ± 1.9
Average Activity (steps)	5139 ± 951

Table 2. Anthropometric characteristics of the subjects. This table shows the baseline characteristics for the study participants. Data is shown as means ± SE.

Subject Metabolic Characteristics	
Total Cholesterol (mg/dl)	172.9 ± 15.8
LDL (mg/dl)	88.8 ± 12.6
HDL (mg/dl)	48.1 ± 2.7
Triglycerides (mg/dl)	179.7 ± 23.6
Fasting glucose (mmol/l)	6.5 ± 0.6
Hemoglobin A1c (%)	6.3 ± 0.2
Average Glucose (mmol/l) (calculated from CGMS)	8.3 ± 0.3
Relative VO_{2peak} (ml/kg/min)	20.4 ± 0.7
Average daily caloric intake (self reported, kcals)	2,114.0 ± 106.9

Table 3. Metabolic measurements. This table shows the results of the lipid panel and complete metabolic panel, both run at a third party laboratory, as well as the hemoglobin A1c, metabolic cart readouts from an exercise stress test, and the average daily caloric intake of the subjects. Data is shown as means ± SE.

Exercise Session Data	
Percent of Heart Rate Reserve	58.1 ± 5.5%
Metabolic Equivalents	4.1 ± 0.2
Percent of Maximal Aerobic Capacity	72.1 ± 4.4%

Table 4. Exercise session data. This table shows the average heart rate reserve as well as the relative proportion of aerobic capacity achieved during the exercise session for all subjects. The aerobic capacity was calculated from formulas in the ACSM's guidelines for exercise testing and prescription 7th edition book. Data is shown as means ± SE.

Activity Data		
Variable	**Sedentary**	**Exercised**
Accelerometer Energy Expenditure	2353.7 ± 105.6	2642.3 ± 41.4*
Accelerometer Steps	3736 ± 163	4980 ± 1141*
Pedometer Steps	3719 ± 302	5348 ± 622*

Table 5. Physical activity levels. This table shows the average steps and energy expenditure between the two phases of the study. These measure were obtained from the Body Media Sense Wear armband monitoring system. (*) indicates a statistically significant difference ($p<0.05$). Data is shown as means ± SE

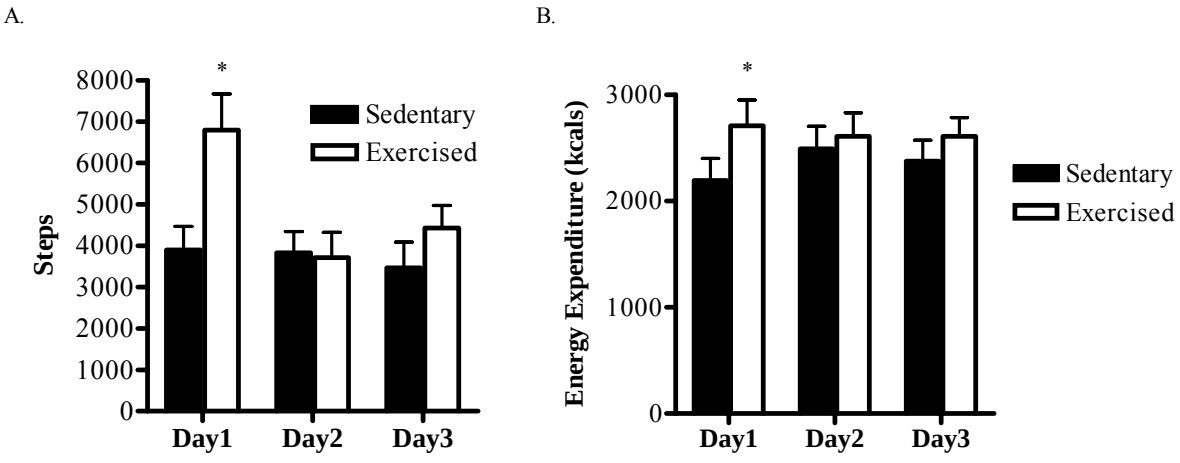


Figure 4. Steps per day and energy expenditure per day across both sedentary and exercised phases of the study. This figure shows the steps per day (A) as well as the estimated daily energy expenditure (B) across the three day period of both phases. Both steps as well as energy expenditure were recorded with the Body Media Sense Wear armband monitoring system. (*) indicates statistical significance ($p<0.05$).

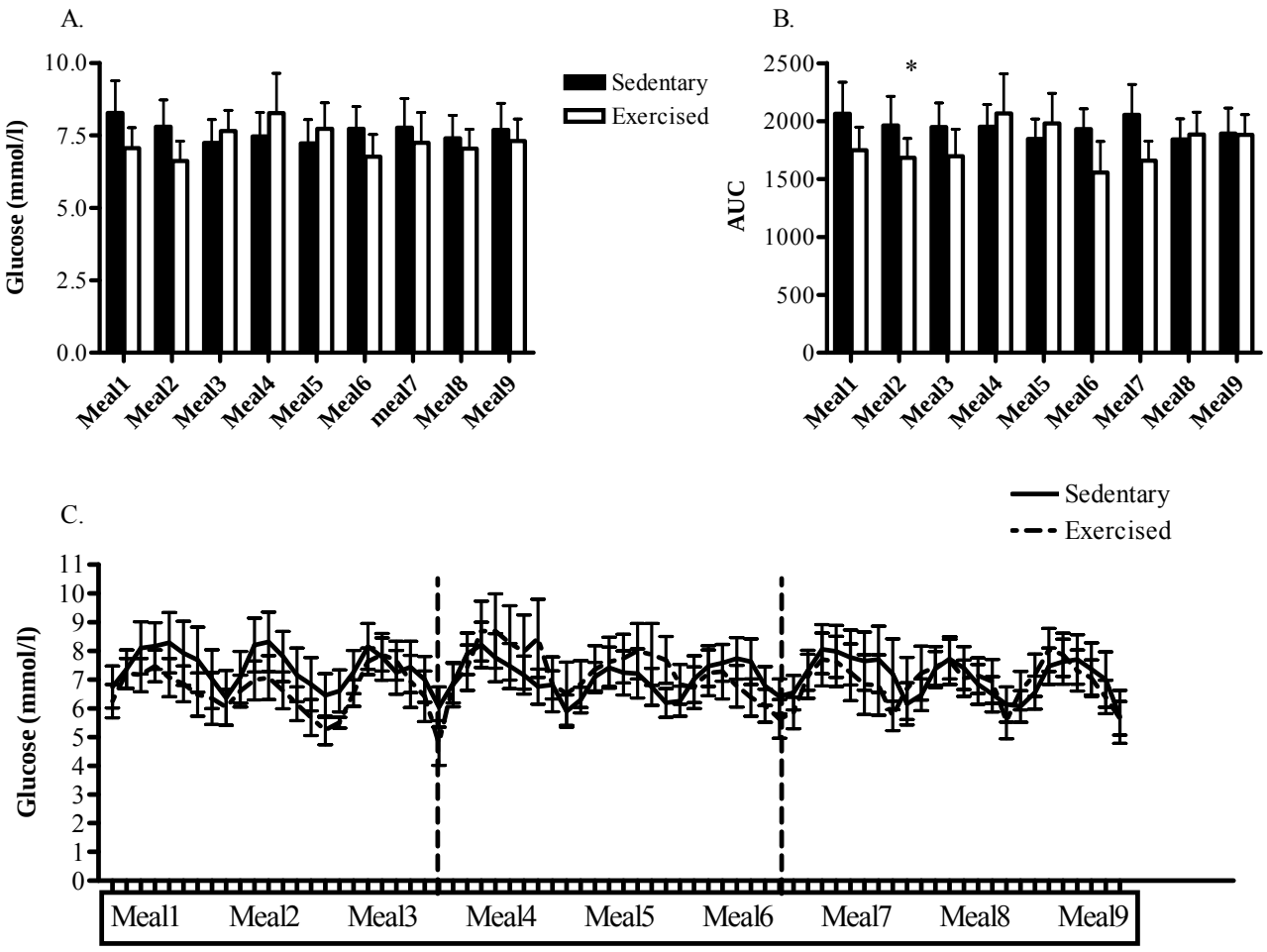


Figure 5. 2 Hour post-prandial glucose level and glucose area under the curve as well as all pos-prandial glucose levels for all three days. This figure shows the 2 hour post-prandial glucose compared at each meal between the two phases (A). Also shown is the area under the glucose curve for each meal between the two phases (B). The blood glucose levels after all the meals are shown although statistics were not run (C). The data is not continuous, and the overnight periods are excluded (indicated by the dashed line). (*) indicates statistical significance ($p<0.05$).

Glucose Averages (mmol/l)	Sedentary			Exercised		
	Day1	Day2	Day3	Day1	Day2	Day3
Daytime	7.2 ± 0.8	7.0 ± 0.6	7.0 ± 0.7	6.4 ± 0.5*	7.0 ± 0.8	6.7 ± 0.7
Nighttime	5.8 ± 0.7	6.0 ± 0.5	5.4 ± 0.6	5.2 ± 0.4	5.4 ± 0.5	5.8 ± 0.4
Total 24 hour	6.9 ± 0.6	6.5 ± 0.6	6.5 ± 0.6	6.3 ± 0.5*	6.3 ± 0.6	6.3 ± 0.6

Table 6. Daytime average glucose level, nighttime average glucose level, and 24 hour average glucose level This table shows the average daytime and nighttime blood glucose levels across the three days of both phases, as well as the 24 hour average glucose levels. The averages were taken from the CGMS readings during the waking and sleeping periods which the subjects noted on a log sheet during the study. The total 24 hour glucose levels were calculated between 12:00am and 11:59pm of each day. Values were compared using a paired T-test. (*) indicates statistical significance ($p<0.05$). Data is shown as means ± SE.

Percent of time in glucose ranges	Sedentary			Exercised		
	Day1	Day2	Day3	Day1	Day2	Day3
Below 3.9 mmol/l	2.4 ± 1.4%	6.7 ± 4.1%	2.0 ± 0.9%	7.2 ± 4.4%	5.1 ± 4.5%	3.1 ± 1.5%
Within 3.9 and 10.0 mmol/l	90.11 ± 4.7%	86.4 ± 5.0%	88.9 ± 5.3%	87.4 ± 5.6%	86.9 ± 5.8%	91.6 ± 3.8%
Above 10.0 mmol/l	7.4 ± 4.9%	6.9 ± 3.6%	9.1 ± 5.4%	5.3 ± 4.5%	8.0 ± 4.6%	5.3 ± 3.9%

Table 7. Percent of time spent within preset glucose level limits. This table shows the percent of time spent within the different glucose level limits across days and between phases. These values were calculated using the Medtronic CGMS software. The percentages were divided by day at 12:00am and the percentage comes from all time during which the monitor was recording during a given day. No statistically significant differences were found. Data is shown as means ± SE.

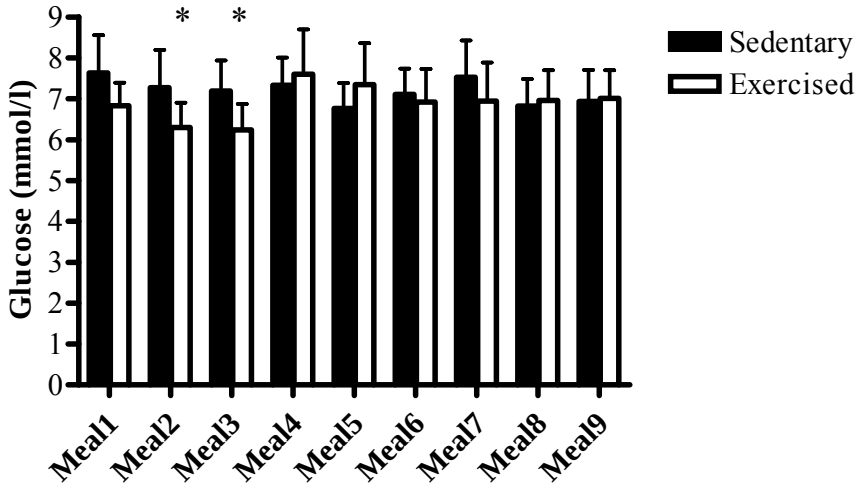


Figure 6. Average post-prandial glucose level per meal across phases. This figure shows the averaged post-prandial 2 hour period for all of the time points for each meal across both phases. (*) indicates statistical significance ($p<0.05$).

Discussion:

The current study found that there was a statistically significant reduction in the glucose AUC in the second meal following a 60 minute exercise bout in previously sedentary individuals with T2D. There was, however no improvement in any of the meals 2 hour post-prandial glucose level. Although the exercise bout was a relatively high volume for this population, the improvement seems to only last several hours beyond the exercise bout. This may be due to the duration of disease development versus the short term intervention. That is to say, several years of inactivity and dietary overconsumption may not be entirely corrected by 60 minutes of exercise on one day. Because of the metabolic inflexibility of this population, early exercise interventions may need to be fairly frequent at the onset of an exercise intervention. This frequency may not have to be maintained after more chronic effects of exercise begin to occur.

The single exercise bout was effective at lowering PPG compared to the sedentary phase for the later time points of the post-prandial period (120, 150, and 240 minutes) when averaged for all 9 meals. This change was only detected in the delta PPG (post-prandial glucose level – pre meal glucose level) at time point 240 minutes, also averaged for all 9 meals. Our lab has previously shown that the exercise prescription used in the current study is effective at improving glycemic control after 5-7 consecutive days of exercise in individuals with T2D (42). Some of these improvements in glucose control seem to change with a single exercise bout; however, the reductions in blood glucose levels after a single exercise bout were not as pronounced as they had been after 5 to 7 days. The time course of changes from completely sedentary to habitual exercise has not

been fully examined in the T2D population, however it seems that there are changes occurring as early as after a single bout of exercise.

The PPG data was observed, but not analyzed for each subject individually because we cannot run statistics on single individual's data. It seemed that only about half of the subjects had a response to the exercise at the given intensity and duration. There was no clear pattern to why these individuals responded more robustly than the other subjects. Neither medications, exercise intensity nor pre-invervention glycemic control seemed to be correlated with the response to the current exercise intensity and duration.

There was no improvement in the overnight glucose in our subjects after the exercise bout. This may have been due to the long duration between the exercise bout and the next sleeping period. If the effect of the exercise bout only lasts several hours, the effect would be gone by the next sleeping period when exercise is performed in the morning (as it was in this study). If the exercise session had occurred in the evening after dinner, there may have been a much better response in the sleeping period.

Average blood glucose level, often estimated using HbA1c, is considered a tool to assess control of disease in individuals with T2D, and HbA1c itself is a combination of post-prandial blood glucose levels and fasting blood glucose level (5). Many medications are used to control either fasting glucose levels, post-prandial glucose levels, or both. It has been shown, however, that exercise, even relatively acute exercise, can be effective at reducing blood glucose levels in both fasting as well as post-prandial periods (39, 42).

Exercise is inexpensive and has relatively few negative side effects when performed properly. If exercise were able to be effectively implemented among the T2D population, there would most likely be improvements in health as well as reductions in health care costs associated with the disease (45, 60).

The current study found that the average blood glucose level over the first 24 hour period was significantly lower after a single exercise bout; however the average blood glucose level was not significantly lower for any days beyond the first. This suggests that individuals with T2D need to exercise on a daily basis to maintain glycemic control. This is in line with other studies suggesting that a single bout of exercise is capable of improving glycemic control in an acute manner. However, it more accurately defines the timeframe in which the effect seems to last (39). When broken down into waking and sleeping periods, there was only a statistically significant difference between the average glucose level for the first waking period, but no statistically significant difference between the overnight periods. Therefore, it seems that the reduction in 24 hour glucose level after a single exercise bout in individuals with T2D was accounted for by a reduction in that day's waking period. Most of the waking period is spent in the post-prandial condition for individuals with T2D, which suggests that the reduction in PPG was driving the reduction of average glucose levels during the first day. Although studies have shown improvements in hepatic insulin sensitivity after a single exercise bout, the failure to see reductions in overnight glucose levels after the exercise bout suggests that this improvement was likely not translated to measureable reductions in hepatic glucose output in this population sample (20).

One bout of exercise did not significantly affect the percent of time spent within pre-set glucose level limits set by the ADA (2) compared to the sedentary condition. This may have been due to the variability in glucose control in the subjects as evidenced by the average glucose level standard deviations. The average standard deviation in the current study was 1.8 mmol/l in the sedentary phase and 1.7 mmol/l in the exercised phase. Some subjects' glucose levels remained relatively low throughout both phases and in both day and night, while other subjects' glucose levels, even while fasting, were relatively high. If we had adjusted the pre-set glucose level limits for each subjects baseline glucose control, we may have been able to better detect changes in time spent within the ranges relative to each subject. Although this method was ineffective at detecting changes in glucose control in the present study, this does not necessarily mean that the method would be ineffective with a larger sample size, with less variability between subjects, or by using a different set of values to determine high and low blood glucose levels.

Some study limitations were due to the measurement technique of using CGMS. A drawback of using CGMS as a primary measure is a lack of focus on any single tissue function. The CGMS assesses whole body changes in blood glucose level, which is a significant outcome when considering that the body must function as a whole organism rather than the sum of its systems. Unfortunately, without focusing on one of the many tissues which are responsive to exercise, glucose, and insulin, we cannot determine where within the physiologic system the improvement occurred. The most likely candidate would be skeletal muscle improvement in glucose uptake. It has been shown in many

studies which used hyperinsulinemic euglycemic clamps that exercise improves skeletal muscle insulin sensitivity in individuals with T2D (31, 51). Another organ which is responsive to insulin and responsible for maintaining fasting blood glucose levels is the liver. Animal models have shown improvements in hepatic insulin sensitivity after only a single bout of exercise (20). Without observing an improvement in overnight (fasting) blood glucose levels, we conclude that either the liver was not significantly affected by the exercise bout, or that the effect did not last beyond the daytime period. These tissues may be speculated on; however there is no direct evidence to conclude the contribution of either on the changes in glycemic control which were observed after only a single exercise bout. Tissues such as adipose or endothelium were not assessed, and we have no good means to estimate any changes by using the CGMS. No measures of blood lipids were taken before or after the exercise bout to assess any changes in circulating NEFAs. In addition, no measures of resting respiratory quotient were taken to estimate changes in macronutrient utilization. No measures were taken to examine any changes in vascular endothelial responsiveness to insulin, although it has been speculated that this may contribute to degradations in glycemic control in individuals with T2D (52). Additional measurement techniques would need to be employed to determine the earliest changes in these tissues in response to a single exercise bout in individuals with T2D.

The high variability in glucose levels among this population (the average standard error was 0.6 mmol/l in both phases compard to 0.1 mmol/l in both phases in a previous study in our lab) makes it difficult to detect small changes in glycemic control which may be occurring acutely upon the beginning of an exercise program. The trend towards

lower glucose levels followed by a rebound and higher glucose levels in the few meals following a single exercise bout suggest that the homeostatic environment was disrupted by the exercise bout, leading to an overcompensation. It is unknown how many bouts of exercise and with what frequency would elicit improvements in glycemic control without disruption and overcompensation of the homeostatic system. The subjects were also taking different types and dosages of drugs (Alloperinol, lisinopril, Metformin, Simvastatin, Hydrochlorothazid, Pravastatin, Lovatadine, Meloxicam, Primadone, Liraglutide, Venlafaxine, Exenatide, Bupropion Hydrochloride, Pioglitazone, Losartan Potassium, Omeprazole, Sertraline, Fenofibrate, Rosuvastatin, Alendronate, Levothyroxine, Albuterol, Gengraf, Prednisone, Furosemide, Lovastatin, Glipizide, Vicodin, and Benicar) and it is unknown how these may have modified the response to exercise. A recent study on the effect of combining Metformin and exercise to improve insulin sensitivity showed that Metformin may reduce the effectiveness of exercise to increase insulin sensitivity in individuals with pre-diabetes (38). Of our 9 subjects, 6 were taking Metformin during the current study.

Another potential confounding variable was the average age of our subjects. This study was meant to look at how exercise affects glycemic control in individuals with T2D, however the aging effect was not accounted for due to the lack of younger individuals with T2D in our study. It has been suggested that a single exercise bout may not be enough to elicit an improvement in glycemic control in middle aged or older adults, but could in younger adults (28). Young sedentary controls have shown improvements in glycemic control on an OGTT 2 to 3 days after a single exercise bout

(3). However, currently, it is not typical to find as many individuals with T2D who are very young to compare with the aging population.

The current study suggests that individuals with T2D are able to improve glycemic control after a single bout of exercise, both measured by daytime average glucose level as well as reductions in PPG, while previous studies have shown that this population can significantly improve their health and control of their disease through habitual exercise (36, 42, 60). This implies that exercise can truly be used as medicine, at least within the confines of this specific disease population. Lifestyle interventions have shown more effectiveness at reducing incidence of T2D than Metformin, a common medication for controlling blood glucose levels (32). The current, and other studies, suggest that the amount of exercise required to begin receiving benefits from exercise is very small (39). Therefore, regular exercise, as well as other lifestyle changes, should be used as the first line of treatment for individuals with T2D or at risk of developing T2D.

Future studies may use CGMS to monitor individuals with T2D during the first 5 days of exercise training to determine the earliest changes in glycemic control with exercise. The possible increase in PPG that occurs after an initial reduction in PPG post-exercise may or may not be eliminated, or it may be an unintended consequence of beginning an exercise regimen in this population. Other studies to further clarify findings in our current study would be to repeat the current study design in non-diseased sedentary adults to determine whether there is any difference in the acute effects of exercise in the presence or absence of T2D. Also, a study using younger subjects could help determine

what amount of loss of glycemic control in the current sample is due to aging versus the disease condition.

In conclusion, a single bout of moderate intensity aerobic exercise was effective at lowering 24 hour average blood glucose level and daytime (waking period) glucose level. This effect only lasted the first day following the exercise bout. Therefore, it appears that sedentary individuals with T2D require exercise on a daily basis to maintain lower blood glucose levels when first starting an exercise program. This frequency may change once more chronic adaptations to exercise have occurred, however, acutely, there seems to be an improvement in glycemic control which only lasts several hours post-exercise. The recommendations may need to be adjusted to reflect that exercise should be performed daily rather than "no more than two consecutive days between bouts" at least when beginning an exercise program (1).

References

1) Exercise and type 2 diabetes: American College of Sports Medicine and the American Diabetes Association: joint position statement. Exercise and type 2 diabetes. Med Sci Sports Exerc. 2010;42:2282-303.

2) Standards of medical care in diabetes--2010. Diabetes Care. 2010;33 Suppl 1:S11-61.

3) Babraj, J. A., Vollaard, N. B., Keast, C., Guppy, F. M., Cottrell, G. and Timmons, J. A.Extremely short duration high intensity interval training substantially improves insulin action in young healthy males. BMC Endocr Disord. 2009;9:3.

4) Bajpeyi, S., Tanner, C. J., Slentz, C. A., Duscha, B. D., McCartney, J. S., Hickner, R. C., Kraus, W. E. and Houmard, J. A.Effect of exercise intensity and volume on persistence of insulin sensitivity during training cessation. J Appl Physiol. 2009;106:1079-85.

5) Barr, R. G., Nathan, D. M., Meigs, J. B. and Singer, D. E.Tests of glycemia for the diagnosis of type 2 diabetes mellitus. Ann Intern Med. 2002;137:263-72.

6) Black, P. H.The inflammatory consequences of psychologic stress: relationship to insulin resistance, obesity, atherosclerosis and diabetes mellitus, type II. Med Hypotheses. 2006;67:879-91.

7) Black, S. E., Mitchell, E., Freedson, P. S., Chipkin, S. R. and Braun, B.Improved insulin action following short-term exercise training: role of energy and carbohydrate balance. J Appl Physiol. 2005;99:2285-93.

8) Blumenthal, J. A., Sherwood, A., Babyak, M. A., Watkins, L. L., Waugh, R., Georgiades, A., Bacon, S. L., Hayano, J., Coleman, R. E. and Hinderliter, A.Effects of exercise and stress management training on markers of cardiovascular risk in patients with ischemic heart disease: a randomized controlled trial. JAMA. 2005;293:1626-34.

9) Bogardus, C., Thuillez, P., Ravussin, E., Vasquez, B., Narimiga, M. and Azhar, S.Effect of muscle glycogen depletion on in vivo insulin action in man. J Clin Invest. 1983;72:1605-10.

10) Bonora, E. and Muggeo, M.Postprandial blood glucose as a risk factor for cardiovascular disease in Type II diabetes: the epidemiological evidence. Diabetologia. 2001;44:2107-14.

11) Brestoff, J. R., Clippinger, B., Spinella, T., von Duvillard, S. P., Nindl, B. C. and Arciero, P. J.An acute bout of endurance exercise but not sprint interval exercise enhances insulin sensitivity. Appl Physiol Nutr Metab. 2009;34:25-32.

12) Cartee, G. D. and Funai, K.Exercise and insulin: Convergence or divergence at AS160 and TBC1D1? Exerc Sport Sci Rev. 2009;37:188-95.

13) Cavalot, F., Pagliarino, A., Valle, M., Di Martino, L., Bonomo, K., Massucco, P., Anfossi, G. and Trovati, M.Postprandial blood glucose predicts cardiovascular events and all-cause mortality in type 2 diabetes in a 14-year follow-up: lessons from the san luigi gonzaga diabetes study. Diabetes Care. 2011;34:2237-43.

14) Cederberg, H., Saukkonen, T., Laakso, M., Jokelainen, J., Harkonen, P., Timonen, M., Keinanen-Kiukaanniemi, S. and Rajala, U.Postchallenge glucose, A1C, and fasting glucose as predictors of type 2 diabetes and cardiovascular disease: a 10-year prospective cohort study. Diabetes Care. 2010;33:2077-83.

15) Ceriello, A.Postprandial hyperglycemia and diabetes complications: is it time to treat? Diabetes. 2005;54:1-7.

16) Ceriello, A. and Motz, E.Is oxidative stress the pathogenic mechanism underlying insulin resistance, diabetes, and cardiovascular disease? The common soil hypothesis revisited. Arterioscler Thromb Vasc Biol. 2004;24:816-23.

17) Christ-Roberts, C. Y., Pratipanawatr, T., Pratipanawatr, W., Berria, R., Belfort, R. and Mandarino, L. J.Increased insulin receptor signaling and glycogen synthase activity contribute to the synergistic effect of exercise on insulin action. J Appl Physiol. 2003;95:2519-29.

18) Colberg, S. R., Zarrabi, L., Bennington, L., Nakave, A., Thomas Somma, C., Swain, D. P. and Sechrist, S. R.Postprandial walking is better for lowering the glycemic effect of dinner than pre-dinner exercise in type 2 diabetic individuals. J Am Med Dir Assoc. 2009;10:394-7.

19) Cox, J. H., Cortright, R. N., Dohm, G. L. and Houmard, J. A.Effect of aging on response to exercise training in humans: skeletal muscle GLUT-4 and insulin sensitivity. J Appl Physiol. 1999;86:2019-25.

20) De Souza, C. T., Frederico, M. J., da Luz, G., Cintra, D. E., Ropelle, E. R., Pauli, J. R. and Velloso, L. A.Acute exercise reduces hepatic glucose production through inhibition of the Foxo1/HNF-4alpha pathway in insulin resistant mice. J Physiol. 2010;588:2239-53.

21) Devlin, J. T. and Horton, E. S.Effects of prior high-intensity exercise on glucose metabolism in normal and insulin-resistant men. Diabetes. 1985;34:973-9.

22) Dyer-Parziale, M.The effect of extend bar containing uncooked cornstarch on night-time glycemic excursion in subjects with type 2 diabetes. Diabetes Res Clin Pract. 2001;53:137-9.

23) Esposito, K., Nappo, F., Marfella, R., Giugliano, G., Giugliano, F., Ciotola, M., Quagliaro, L., Ceriello, A. and Giugliano, D.Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans: role of oxidative stress. Circulation. 2002;106:2067-72.

24) Fiallo-Scharer, R.Eight-point glucose testing versus the continuous glucose monitoring system in evaluation of glycemic control in type 1 diabetes. J Clin Endocrinol Metab. 2005;90:3387-91.

25) Franc, S., Dardari, D., Peschard, C., Riveline, J. P., Biedzinski, M., Boucherie, B., Petit, C., Requeda, E., Mistretta, F., Varroud-Vial, M. and Charpentier, G.Can postprandial blood glucose excursion be predicted in type 2 diabetes? Diabetes Care. 2010;33:1913-8.

26) Hawley, J. A.Exercise as a therapeutic intervention for the prevention and treatment of insulin resistance. Diabetes Metab Res Rev. 2004;20:383-93.

27) Henriksen, E. J.Invited review: Effects of acute exercise and exercise training on insulin resistance. J Appl Physiol. 2002;93:788-96.

28) Henriksson, J.Influence of exercise on insulin sensitivity. J Cardiovasc Risk. 1995;2:303-9.

29) Holloszy, J. O.A forty-year memoir of research on the regulation of glucose transport into muscle. Am J Physiol Endocrinol Metab. 2003;284:E453-67.

30) Houmard, J. A., Hickey, M. S., Tyndall, G. L., Gavigan, K. E. and Dohm, G. L.Seven days of exercise increase GLUT-4 protein content in human skeletal muscle. J Appl Physiol. 1995;79:1936-8.

31) Kirwan, J. P., Solomon, T. P., Wojta, D. M., Staten, M. A. and Holloszy, J. O.Effects of 7 days of exercise training on insulin sensitivity and responsiveness in type 2 diabetes mellitus. Am J Physiol Endocrinol Metab. 2009;297:E151-6.

32) Knowler, W. C., Barrett-Connor, E., Fowler, S. E., Hamman, R. F., Lachin, J. M., Walker, E. A. and Nathan, D. M.Reduction in the incidence of type 2 diabetes with lifestyle intervention or metformin. N Engl J Med. 2002;346:393-403.

33) Kraniou, G. N., Cameron-Smith, D. and Hargreaves, M.Acute exercise and GLUT4 expression in human skeletal muscle: influence of exercise intensity. J Appl Physiol. 2006;101:934-7.

34) Kuenen, J. C., Borg, R., Kuik, D. J., Zheng, H., Schoenfeld, D., Diamant, M., Nathan, D. M. and Heine, R. J.Does glucose variability influence the relationship between mean plasma glucose and HbA1c levels in type 1 and type 2 diabetic patients? Diabetes Care. 2011;34:1843-7.

35) Lima, A. F., Ropelle, E. R., Pauli, J. R., Cintra, D. E., Frederico, M. J., Pinho, R. A., Velloso, L. A. and De Souza, C. T.Acute exercise reduces insulin resistance-induced TRB3 expression and amelioration of the hepatic production of glucose in the liver of diabetic mice. J Cell Physiol. 2009;221:92-7.

36) Little, J. P., Gillen, J. B., Percival, M., Safdar, A., Tarnopolsky, M. A., Punthakee, Z., Jung, M. E. and Gibala, M. J.Low-volume high-intensity interval training reduces hyperglycemia and increases muscle mitochondrial capacity in patients with type 2 diabetes. J Appl Physiol. 2011;

37) Magkos, F., Tsekouras, Y., Kavouras, S. A., Mittendorfer, B. and Sidossis, L. S.Improved insulin sensitivity after a single bout of exercise is curvilinearly related to exercise energy expenditure. Clin Sci (Lond). 2008;114:59-64.

38) Malin, S. K., Gerber, Robert, Chipkin, Stuart R., Braun, Barry.Independent and Combined Effects of Exercise Training and Metformin on Insulin Sensitivity in Individuals With Prediabetes. Diabetes Care. 2011;10:

39) Manders, R. J., Van Dijk, J. W. and van Loon, L. J.Low-intensity exercise reduces the prevalence of hyperglycemia in type 2 diabetes. Med Sci Sports Exerc. 2010;42:219-25.

40) Meigs, J. B., Nathan, D. M., D'Agostino, R. B., Sr. and Wilson, P. W.Fasting and postchallenge glycemia and cardiovascular disease risk: the Framingham Offspring Study. Diabetes Care. 2002;25:1845-50.

41) Mikines, K. J., Sonne, B., Farrell, P. A., Tronier, B. and Galbo, H.Effect of physical exercise on sensitivity and responsiveness to insulin in humans. Am J Physiol. 1988;254:E248-59.

42) Mikus, C. R., Fairfax, S. T., Libla, J. L., Boyle, L. J., Vianna, L. C., Oberlin, D. J., Uptergrove, G. M., Deo, S. H., Kim, A., Kanaley, J. A., Fadel, P. J. and Thyfault, J. P.Seven days of aerobic exercise training improves conduit artery blood flow following glucose ingestion in patients with type 2 diabetes. J Appl Physiol. 2011;

43) Monnier, L., Colette, C., Dunseath, G. J. and Owens, D. R.The loss of postprandial glycemic control precedes stepwise deterioration of fasting with worsening diabetes. Diabetes Care. 2007;30:263-9.

44) Monnier, L., Mas, E., Ginet, C., Michel, F., Villon, L., Cristol, J. P. and Colette, C.Activation of oxidative stress by acute glucose fluctuations compared with sustained chronic hyperglycemia in patients with type 2 diabetes. JAMA. 2006;295:1681-7.

45) Pahor, M. M.Consideration of Insurance Reimbursement for Physical Activity and Exercise Programs for Patients With Diabetes. Journal of the American Medical Association. 2011;305:1808 -1809.

46) Richter, E. A., Mikines, K. J., Galbo, H. and Kiens, B.Effect of exercise on insulin action in human skeletal muscle. J Appl Physiol. 1989;66:876-85.

47) Riddle, M. C., Rosenstock, J. and Gerich, J.The treat-to-target trial: randomized addition of glargine or human NPH insulin to oral therapy of type 2 diabetic patients. Diabetes Care. 2003;26:3080-6.

48) Rockl, K. S., Witczak, C. A. and Goodyear, L. J.Signaling mechanisms in skeletal muscle: acute responses and chronic adaptations to exercise. IUBMB Life. 2008;60:145-53.

49) Saydah, S. H., Miret, M., Sung, J., Varas, C., Gause, D. and Brancati, F. L.Postchallenge hyperglycemia and mortality in a national sample of U.S. adults. Diabetes Care. 2001;24:1397-402.

50) Scheen, A. J.Treatment of type 2 diabetes. Acta Clin Belg. 2003;58:318-24.

51) Segerstrom, A. B., Glans, F., Eriksson, K. F., Holmback, A. M., Groop, L., Thorsson, O. and Wollmer, P.Impact of exercise intensity and duration on insulin sensitivity in women with T2D. Eur J Intern Med. 2010;21:404-8.

52) Solomon, T. P., Haus, J. M., Li, Y. and Kirwan, J. P.Progressive Hyperglycemia across the Glucose Tolerance Continuum in Older Obese Adults Is Related to Skeletal Muscle Capillarization and Nitric Oxide Bioavailability. J Clin Endocrinol Metab. 2011;

53) Sriwijitkamol, A., Coletta, D. K., Wajcberg, E., Balbontin, G. B., Reyna, S. M., Barrientes, J., Eagan, P. A., Jenkinson, C. P., Cersosimo, E., DeFronzo, R. A., Sakamoto, K. and Musi, N.Effect of acute exercise on AMPK signaling in skeletal muscle of subjects with type 2 diabetes: a time-course and dose-response study. Diabetes. 2007;56:836-48.

54) Tahrani, A. A., Bailey, C. J., Del Prato, S. and Barnett, A. H.Management of type 2 diabetes: new and future developments in treatment. Lancet. 2011;378:182-97.

55) Thompson, P. D., Buchner, D., Pina, I. L., Balady, G. J., Williams, M. A., Marcus, B. H., Berra, K., Blair, S. N., Costa, F., Franklin, B., Fletcher, G. F., Gordon, N. F., Pate, R. R., Rodriguez, B. L., Yancey, A. K. and Wenger, N. K.Exercise and physical activity in the prevention and treatment of atherosclerotic cardiovascular disease: a statement from the Council on Clinical Cardiology (Subcommittee on Exercise, Rehabilitation, and Prevention) and the Council on Nutrition, Physical Activity, and Metabolism (Subcommittee on Physical Activity). Circulation. 2003;107:3109-16.

56) Thyfault, J. P.Setting the stage: possible mechanisms by which acute contraction restores insulin sensitivity in muscle. Am J Physiol Regul Integr Comp Physiol. 2008;294:R1103-10.

57) Thyfault, J. P., Cree, M. G., Zheng, D., Zwetsloot, J. J., Tapscott, E. B., Koves, T. R., Ilkayeva, O., Wolfe, R. R., Muoio, D. M. and Dohm, G. L.Contraction of insulin-resistant muscle normalizes insulin action in association with increased mitochondrial activity and fatty acid catabolism. Am J Physiol Cell Physiol. 2007;292:C729-39.

58) Thyfault, J. P., Rector, R. S. and Noland, R. C.Metabolic inflexibility in skeletal muscle: a prelude to the cardiometabolic syndrome? J Cardiometab Syndr. 2006;1:184-9.

59) Translation, C. s. D. o. D.Long-term trends in diabetes. 2010;

60) Umpierre, D., Ribeiro, P. A., Kramer, C. K., Leitao, C. B., Zucatti, A. T., Azevedo, M. J., Gross, J. L., Ribeiro, J. P. and Schaan, B. D.Physical activity advice only or structured exercise training and association with HbA1c levels in type 2 diabetes: a systematic review and meta-analysis. JAMA. 2011;305:1790-9.

61) Wojtaszewski, J. F., Jorgensen, S. B., Frosig, C., MacDonald, C., Birk, J. B. and Richter, E. A.Insulin signalling: effects of prior exercise. Acta Physiol Scand. 2003;178:321-8.

62) Wolever, T. M., Chiasson, J. L., Csima, A., Hunt, J. A., Palmason, C., Ross, S. A. and Ryan, E. A.Variation of postprandial plasma glucose, palatability, and symptoms

associated with a standardized mixed test meal versus 75 g oral glucose. Diabetes Care. 1998;21:336-40.

63) Wright, E., Jr., Scism-Bacon, J. L. and Glass, L. C.Oxidative stress in type 2 diabetes: the role of fasting and postprandial glycaemia. Int J Clin Pract. 2006;60:308-14.

APPENDIX A

EXTENDED LITERATURE REVIEW

Review of Literature:

History of Diabetes

Diabetes is a disease which has been known in humans for over 2000 years (58). The word diabetes comes from the Greek word which means "to siphon", because even long ago individuals with diabetes were known for excessive urination and wasting (58). Type 1 diabetes was historically more common, however, the most common form of diabetes in America today is type 2 diabetes (T2D); formerly known as adult onset diabetes. This is different from type 1 diabetes which occurs in children, which is an auto-immune disease in most cases. T2D stems from obesity, excessive caloric consumption, and too little physical activity leading to insulin resistance, hyperinsulinemia, and β cell failure (16, 53). The two distinct variations were not known until as late as 1935 (58).

Until fairly recently there was no effective treatment for diabetes. It was not until as late as the 17th century that it was discovered by Dr. Thomas Willis that the diabetic urine had a sweet taste indicating glucose in the urine (58). Also, not until 1921 was insulin discovered from the pancreas of a dog, followed by insulin injections which were the first effective treatment for diabetes. By the 1950s oral hypoglycemic medications (sulfonylureas, which increase β cell production of insulin) were developed to treat diabetes (58). Between 1983 and 1993 the diabetes control and complications trial (DCCT) was conducted which showed that controlling blood glucose levels in diabetic patients had clear benefits in reducing complications such as retinopathy and neuropathy

(1, 58). It has not been until recent times that more effective treatments for diabetes have been discovered, however despite better treatments, the prevalence of T2D has steadily increased (70).

Type 2 diabetes has increased in prevalence at an alarming rate in recent years (70). In the late 1950s less than 1% of the United States population had T2D (70). This was about 1.5 million people (70). However, by 2009, 6.8% of the United States population had T2D, which translates to greater than 20 million people (70). This trend seems to still be increasing; therefore it is critical to develop new strategies for prevention and treatment of T2D.

In addition to diabetes, cardiovascular disease and metabolic syndrome are also associated with insulin resistance and uncontrolled blood glucose levels. Elevated post-challenge glucose levels can lead to the development of cardiovascular disease, as well as increased death from cardiac events and increased all cause mortality (14, 15, 31, 59). These diseases are associated with obesity which is also increasing in the United States today. This has led to the common soil hypothesis that these conditions all have similar root causes in poor diet and inactivity which can lead to insulin resistance and impaired glycemic control (16, 53).

Insulin Resistance

Development of insulin resistance

Insulin resistance is a condition that develops slowly over many years (7, 52, 55, 60). The very earliest physiologic changes are only beginning to be understood. There seems to be an early stage of oxidative damage, inflammation, and accumulation of fat which have all been linked to the malfunctioning of insulin stimulated glucose transport pathways in muscle (6, 7, 16, 30, 34, 72, 75-77). The pancreatic β cells can compensate for insulin resistance by producing greater amounts of insulin which leads to a state called hyperinsulinemia (28, 69). The hyperinsulinemic state can persist as long as the pancreatic β cells can continue to produce insulin in large enough quantities. Higher concentrations of insulin are still effective at controlling blood glucose levels, however, the pancreatic β cells, through mechanisms that are still incompletely understood, eventually start to have an impaired ability to produce insulin (Figure 1)(28, 38, 39, 74). At this point, a patient's blood glucose levels begins to be elevated, a condition called impaired glucose tolerance, which if left unmitigated, can lead to type 2 diabetes (28, 38, 69, 74). After two fasting blood glucose level readings above 7.0 mmol/L (126 mg/dl) a person is classified as having T2D (5). Initially blood glucose levels cannot be controlled post-prandially. However, as the disease progresses, blood glucose levels will be uncontrolled during the fasting state, usually in the morning hours. Finally when the disease is further progressed the individual may have elevated fasting glucose levels even during overnight sleep periods (51). Although this is considered the typical disease

progression, there are cases in which individuals have impaired fasting glucose levels without impaired post-prandial glucose control and vice-versa.

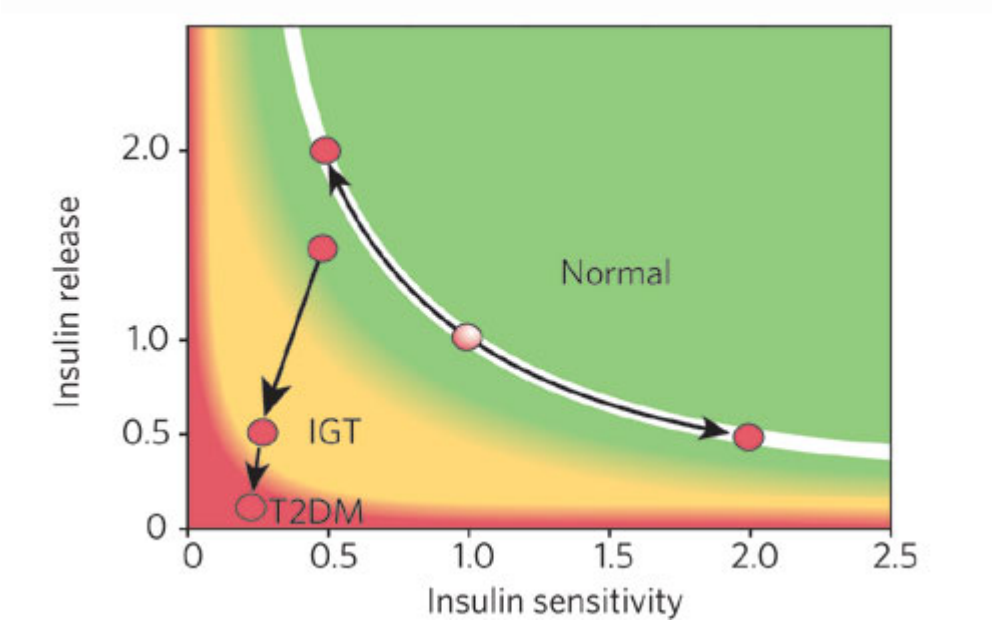


Figure 1. Shows data from Kahn et al 2006 which shows how increased insulin production compensates for decreased insulin sensitivity until β cell failure occurs and impaired glucose tolerance and T2D develop (39).

Oxidative stress is one possible factor which leads to the development of insulin resistance (15, 16, 21, 75). The oxidative stress is caused by over consumption of food, too little physical activity, and obesity (16). One possible consequence of excess glucose levels in the blood is glucose being forced into skeletal muscle cells by insulin to maintain blood glucose levels within normal ranges (16). The excess glucose can move through the tricarboxylic acid cycle and produce NADH which can generate reactive oxygen species (ROS) in the electron transport chain of the mitochondria, which in turn can lead to the activation of NFκB and TNF-α (15, 16, 75). NFκB can also lead to the production of IL-6 which stimulates hepatocytes to produce CRP (6, 77). These can lead to inflammation and the production of other inflammatory cytokines. This process is set in motion during periods of hyperglycemia (27). The production of ROS can occur in different tissues and lead to different outcomes depending upon the tissue's function.

Obesity, inactivity, and over eating can all also produce an inflammatory state in the body, which has its own effects on the development of insulin resistance. Inflammatory proteins can directly reduce insulin sensitivity (6, 7, 30, 34, 76, 77). For example, increased circulating TNF-α can cause serine phosphorylation of IRS-1 and inhibit insulin signaling in endothelial cells (6, 30). TNF-α may also have deleterious effects on pancreatic β cells reducing their ability to produce insulin in sufficient amounts (30). Beyond direct insulin signaling, this inflammation can lead to other chronic disease symptoms such as atherosclerosis (7, 16).

Accumulation of ectopic fat stores in skeletal muscle and liver may also contribute to the development of insulin resistance (2, 6, 7, 30, 34, 67, 72). In particular,

the over-accumulation of intramuscular diacylglycerol and ceramides in particular are thought to cause a decrease insulin signaling in skeletal muscle and liver cells (67, 72). In addition, increased adiposity can lead to an increase in inflammatory cytokine production which can reduce insulin signaling in muscle and liver (6, 7, 30, 34). An accumulation of fat in the liver may precede reduced insulin sensitivity, or be caused by the reduced insulin sensitivity. This results in excessive hepatic glucose output.

Effected Tissues

Many tissues in the body are dependent upon insulin and glucose for normal functioning, therefore, insulin resistance has many effects on various tissues in the body. The main tissues which show large changes in insulin sensitivity and functionality are the liver, vascular endothelial cells, adipose, and skeletal muscles (9, 46, 54, 65-67). Each of these responds to insulin in a different way. When they become insulin resistant, they display different signs and symptoms in the body. Also, each tissue's dysfunction leads to symptoms associated with T2D as well as cardiovascular disease and metabolic syndrome.

The liver is responsible for maintaining blood glucose levels within normal ranges, particularly during a fasting state (26, 36). The liver is used as a store for glycogen, which can be broken down into glucose, and can produce glucose from non-carbohydrate sources (gluconeogenesis) to be moved into circulation to maintain blood glucose levels (26, 36, 46, 73). The two main hormones responsible for control of hepatic glucose output are glucagon and insulin (26, 36). Glucagon stimulates

glycogenolysis to break glycogen into glucose and gluconeogenesis to create glucose from non-carbohydrate sources. Insulin, however, stimulates the liver to cease glycogenolysis and gluconeogenesis and instead take up glucose for glycogenesis (26, 36). These two hormones balance one another, and keep blood glucose levels within a tight range in healthy individuals.

When the hepatocytes become insulin resistant, they no longer receive the signal to cease glycogenolysis and gluconeogenesis after a meal or during a fasting condition (26, 46). This can lead to increases in fasting blood glucose levels (26). Besides the increase in fasting blood glucose levels, the inability of the liver to cease glycogenolysis and gluconeogenesis in response to insulin leads to an amplification of post-prandial hyperglycemic excursions. Other cells have to take up the excess glucose to keep blood levels down, which increases stress on the β cells to produce increasing amounts of insulin.

The liver becomes insulin resistant and also begins to accumulate stores of lipids, however it is unknown which occurs first (73). Insulin resistance can lead to decreased Foxa1 activation in the liver which leads to decreased lipid oxidation (73). At the same time the insulin resistance and associated hyperinsulinemia stimulates sterol receptor binding protein 1-c which increases lipogenesis (73). This, over time, can lead to an accumulation of lipids in the liver and eventual non-alcoholic fatty liver disease (73).

Endothelial cells of the vasculature are also affected by insulin. Insulin, in a healthy person, stimulates vasodilatation of arterioles to facilitate the delivery of glucose

and other nutrients to tissues, such as skeletal muscle (4, 65). Insulin's vasodilating effect is mediated through the stimulation of endothelium-derived nitric oxide in the vessels (65). This affects the delivery of glucose to body tissues, such as skeletal muscle (64, 65). ROS, generated by hyperglycemia and inflammation, can also lead to eNOS uncoupling, or the scavenging of NO, thus reducing the ability of insulin to activate vasodilation. The decreased production of NO, increased degradation of NO, and increased production of endothelin-1 (a potent vasoconstrictor) can lead to changes which reduce the responsiveness to NO and insulin stimulated vasodilation (4). The change in endothelial insulin induced vasodilation is one symptom of insulin resistance and T2D as it reduces the ability of insulin to increase blood flow to capillaries and thus decrease deliver of glucose and insulin to all skeletal muscle beds (4, 64).

Adipocytes also can become insulin resistant over time in a hyperinsulinemic state. Adipocytes respond to insulin by ceasing lipolysis and storing non-esterified fatty acids (NEFAs) as TAG (29). However when they become insulin resistant, they do not stop lipolysis after a meal (3). This leads to increased circulating NEFAs which may result in storage of fat in other tissues and or compete for glucose as a predominant source of energy, both of which may have pathologic consequences (3, 29). Thus insulin resistance in adipocytes has also been linked to insulin resistance in muscle and liver (29, 73).

Skeletal muscle plays a key role in maintaining homeostasis by alternating its fuel source according to different metabolic conditions, and is the primary disposal site for glucose post-prandially (40, 69). During a post-prandial state, blood glucose levels and

insulin levels rise and muscle oxidizes carbohydrate for energy, or stores it as glycogen (25, 40, 69). Conversely, during a fast muscle will oxidize lipids from NEFAs for energy (40, 69). As muscles become resistant to insulin, they also seem to become inflexible to changing their usage of energy substrates during fed and fasting conditions (40, 69). This can exacerbate the condition by increasing the storage of lipids within the skeletal muscles or not being able to reduce the increases in blood glucose levels (40, 69).

Glycogen content of the muscles may play a role in the sensitivity to insulin and flexibility of muscles to oxidize different substrates (8, 33). The increased blood glucose disposal post-exercise is largely due to an increase in non-oxidative glucose disposal, i.e. storage as muscle glycogen. Muscle takes up glucose to store as glycogen while fat oxidation is still predominant in the muscle (25). This is called glycogen super-compensation, when stored muscle glycogen is repleated to a higher level than before exercise. The super-compensation of muscle glycogen after being depleted appears to be driven by the increased glucose transport rather than an increase in glycogen synthase activity (33). When subjects are fed carbohydrate post-exercise they show a smaller improvement in glucose tolerance later (8). Therefore, when the muscle has full stores of energy it is less sensitive to insulin and may start to become inflexible to switching substrate utilization.

Glucose transport

The main sink in the body for peripheral glucose disposal is skeletal muscle. Skeletal muscle is believed to account for up to 90% of post-prandial glucose uptake

(32). Glucose transport in skeletal muscle is handled mostly by GLUT 1 and GLUT 4 glucose transport proteins (33). GLUT 1 is always at the cell membrane, where as GLUT 4 translocates from within the cell to the cell membrane when stimulated by contraction or insulin (20, 33, 44),(33, 56). Glucose tolerance seems to track with higher or lower GLUT 4 protein content rather than changes in GLUT 1 (33). In light of this, the effects of insulin and contraction of skeletal muscle on GLUT 4 translocation are important for understanding the development of glucose intolerance in skeletal muscle.

Glucose uptake by skeletal muscle is primarily stimulated via the insulin signaling pathway (56). Insulin is released by the pancreatic β cells when blood glucose levels rise. Insulin then travels in circulation where it can bind to insulin receptors on skeletal muscle and stimulate tyrosine phosphorylation on IRS-1 (32, 56). The IRS-1 then activates Phosphatidylinositol 3-kinase (PI3K) leading to a series of events which causes activation of protein kinase B (Akt) (32, 56). Akt phosphorylates Akt substrate of 160 kilodaltons (AS 160) which prevents the hydrolysis of GTP on Rab proteins which bind GLUT 4 (13, 56). The Rab protein GTPase inhibition allows the release and translocation of GLUT 4 from intracellular docking stations to the cell membrane, where it can then facilitate the movement of glucose into the muscle cell (13, 56).

Muscle contraction also causes GLUT 4 translocation as well as stimulating an up-regulation of GLUT 4 expression (33, 56). The pathway for muscular contraction stimulated GLUT 4 translocation is, however, less well understood compared with insulin stimulated GLUT 4 translocation. During muscular contractions, an increase in the AMP to ATP ratio in the cell leads to activation of AMP activated protein kinase (AMPK) (56).

AMPK can then phosphorylate AS 160 to cause GLUT 4 translocation (56). Calcium (Ca) from the sarcoplasmic reticulum triggers calmodulin which seems to affect GLUT 4 translocation, most likely through AS160 (13, 56). Furthermore, calmodulin dependent protein kinases (CAMK) and AMPK may also increase GLUT 4 expression in the cell (33), thus chronic muscle contraction should lead to a larger depot of GLUT 4 protein. Also, nNOS activation and production of some ROS can lead to contraction stimulated glucose uptake (17, 37, 49). These processes, stimulated by contraction, allow glucose to enter the cell independent of insulin (33, 67).

Insulin stimulated and contraction stimulated GLUT 4 translocation can operate separately or together (33, 67, 68). By either stimulating a muscle with insulin or inducing contraction, GLUT 4 will translocate and allow glucose uptake (33, 67). However, when insulin stimulation is preceded by contraction, an even greater GLUT 4 translocation and glucose uptake occurs (67, 68). In healthy lean individuals, there is an additive effect between insulin and contraction, however in obese insulin resistant individuals, there is a synergistic effect which seems to restore responsiveness to insulin (67, 68) It has been suggested that muscular contraction also enhances insulin stimulated glucose uptake although it has not yet been conclusively determined (18). For people with diabetes or pre-diabetes, exercise can improve glucose tolerance and potentially lower post-prandial glucose levels, lower insulin level requirements to drive glucose uptake, and prevent development of diabetes (61).

Treatment of Insulin Resistance

Exercise and Blood Glucose Levels

Inactivity is one of the causes of insulin resistance which can lead to T2D. Exercise can be used as a treatment because it affects GLUT 4 translocation, as well as potentially improving insulin signaling (11, 18, 20, 41, 44). Using regular planned physical activity can have an effect similar to a hypoglycemic medication (42, 50). After engaging in an exercise bout, individuals with T2D have decreased blood glucose levels (19, 45, 48, 63). In addition, after exercise training, there can be an improvement in glucose tolerance which can have a positive effect on post-prandial glycemic excursions in individuals with T2D (45, 48, 50). A study in healthy men showed an improvement in glucose uptake by the skeletal muscle of an exercised leg versus an unexercised leg in the same individual (54). Studies in animals have shown improved insulin sensitivity in the liver after exercise and reduced hepatic glucose output (46, 57). It has also been shown that a single bout of exercise can improve the insulin signaling pathway in the liver in diseased animal models (22). Exercise has also been shown to improve pancreatic β cell function in obese animals (24). Besides treating the symptoms of T2D, namely insulin resistance and uncontrolled blood glucose levels, exercise can treat the root causes of the illness (53).

Exercise and Glycemic Control

Using exercise as a treatment can be an effective way to improve clinical markers of diabetes such as HbA1c and fasting glucose levels, and more importantly exercise can

help control post-prandial glycemic excursions (11, 19, 71). These three parameters are used to diagnose and monitor T2D. These parameters can be affected by exercise by reducing the amplitude of a post-prandial glycemic excursion (5, 45).

Studies have shown that exercise or lifestyle modification can be as effective if not more so than pharmacological interventions at preventing and treating T2D. A study by the Diabetes Prevention Program Research Group examined the effects of exercise or Metformin versus a placebo (43). The study recruited 3,234 participants who were randomly assigned to either a Metformin group, a placebo group, or a lifestyle intervention group. The Metformin group initially took 850 mg of Metformin a day and a placebo once a day. After a month the dose was increased to 850 mg twice daily while the placebo group took a placebo twice daily throughout. The lifestyle intervention group was told to lose approximately 7% of their body mass through a healthy low fat, low calorie, diet and to engage in physical activity for at least 150 minutes per week. Both the Metformin and placebo groups were recommended to follow the food guide pyramid, reduce weight, and increase physical activity. Both HbA1c as well as diagnosis of T2D were tracked over four years at six month intervals.

The results of the study showed, as expected, that the placebo group had the highest incidence of diagnosis with T2D. By one year into the study, there was statistically significantly higher incidence of T2D in the placebo group compared with both the lifestyle and Metformin group, and this significance would persist until the end of the study. The lifestyle group had statistically significantly lower incidence of T2D than the Metformin group, which would also persist until the end of the study. The

Metformin group had 17 – 43% lower risk of developing T2D compared to the placebo group while the lifestyle group had 48 – 66% lower risk compared to the placebo group. Thus, lifestyle was the most effective at preventing T2D in a at risk population.

There were also differences seen in HbA1c and fasting plasma glucose levels between the groups. As expected, the placebo group had statistically significantly higher fasting plasma glucose levels as well as HbA1c throughout the study. The Metformin group and lifestyle group both had similar fasting plasma glucose levels throughout the study, however their HbA1c differed. The lifestyle group had statistically significantly lower HbA1c compared with the Metformin group between 6 months and 3 years into the study. This seems to suggest that the lifestyle intervention was better at controlling PPG than was Metformin.

A recent meta-analysis examined different training modes and their effectiveness at lowering HbA1c (71). The study looked at 47 randomized controlled clinical trials which tested resistance training, aerobic training, a combination of resistance and aerobic training, physical activity and diet recommendations, or only physical activity or diet recommendations. The meta-analysis found that the aerobic exercise was associated with an HbA1c reduction of 0.73% compared to control. Resistance training was associated with an HbA1c reduction of 0.57% compared to control. The combination of aerobic and resistance training was associated with an HbA1c reduction of 0.51% compared to control. Physical activity advice was associated with a decline in HbA1c of 0.43% compared to control, but only when paired with dietary advice. An additional finding of the meta-analysis was that there was a significant difference between exercise for 150

minutes or more per week versus less than 150 minutes of exercise per week. Exercising 150 minutes or more per week reduced HbA1c by 0.89% compared to control; versus a 0.36% reduction in HbA1c compared to control from exercising less than 150 minutes per week. Therefore it seems that both type and duration of exercise have differing effects on glycemic control in subjects with T2D.

A clinical trial examined whether there was a difference between aerobic exercise and resistance exercise in their ability to improve glycemic control measured by HbA1c in individuals with T2D (62). The researchers studied 251 adults with T2D over six months. The subjects were divided into four groups: aerobic exercise, resistance exercise, aerobic and resistance exercise, or control group. Before the training period began, all the subjects were exercised moderately for four weeks to ensure compliance during the six month training period. The subjects were given memberships to local training facilities and were supervised by personal trainers. The primary variable measured was HbA1c pre and post intervention. Both the resistance and aerobic exercise training groups had statistically significant reductions in HbA1c compared to the control group. The combined resistance and aerobic exercise group had statistically significant reductions in HbA1c compared to either aerobic or resistance training alone. This study seems to indicate that either the total amount of muscle mass being used or the total amount of exercise being performed, or both, is important for improving glycemic control. The exercise groups were not isocaloric, therefore whether the effect is due to differences in caloric expenditure during the different forms of exercise is unknown. Also, because the resistance and aerobic exercises might recruit different muscle groups,

there is a chance that activating a larger mass of skeletal muscle improves the amount of tissue which has improved glucose tolerance. Because the intensity of the exercise was not strictly controlled during the training period, there is no way of knowing whether or not there was an effect of the intensity of exercise on the improvements in glycemic control.

Exercise training seems to be an effective strategy for improving HbA1c over time; however, that measure can take a long time to change because as already stated, it is believed to reflect the average blood glucose levels over a 3 month period. Another study examined the effects of an acute bout of exercise on glycemic control. Brestoff et al. compared the effects of an acute bout of either sprint interval training or endurance exercise on glycemic control in healthy individuals (12). The subjects were given an OGTT at baseline and then over the next few weeks performed, in a random order, both an acute bout of sprint interval training and endurance exercise. The day after the training, another OGTT was given for comparison to baseline. There was no statistically significant difference in plasma glucose levels at any time point or for total glucose area under the curve (AUC) for any of the testing periods. However, the plasma insulin levels were statistically significantly lower than baseline in the endurance exercise at the zero and 60 minute time points which lead to a statistically significantly lower insulin AUC. In addition, the endurance exercise showed a statistically significantly higher insulin sensitivity index (ISI) compared to both baseline and sprint interval training. This seems to indicate, at least in healthy individuals, that the acute effect of exercise on glycemic control is related to the duration of the exercise more than the intensity.

While the previous studies have used techniques to estimate glycemic control, none measured it directly over time in free living conditions. To measure this, some studies have used continuous glucose monitoring systems (CGMS). One study examined high intensity interval training specifically to determine whether shorter duration, higher intensity exercise could be used in place of long duration endurance exercise. This study by Little et al. recruited eight individuals with T2D to perform two weeks of high intensity interval training (6 exercise sessions) (47). The training sessions were conducted on cycle ergometers and supervised by lab personnel. The intervals were ten 60 second sprints on the cycle ergometers at 90% of max heart rate. Also, glucose levels were continuously monitored for two days before the training period and two days after the training period.

The subjects had statistically significantly lower 24 hour average blood glucose levels after the two week training intervention as compared to before the intervention. The area under the 24 hour glucose curve was also statistically significantly lower after the training intervention. Total amount of GLUT 4 protein present in the muscle biopsy was statistically significantly higher post-exercise intervention, indicating that the low volume of high intensity exercise was enough to elicit an increase in GLUT 4 mRNA. It is still unknown whether the intensity of the exercise is as important as the persistence of exercise over time.

The previous studies have shown how improving activity over time can help reduce risk of T2D as well as improve glycemic control, however they do not examine how long a person has to exercise to start to see improvements. A study by our lab

examined the changes in post-challenge glycemic control after only seven days of aerobic exercise training in subjects with T2D (50). The study recruited 11 men and women with T2D to perform seven consecutive days of aerobic exercise training for 60 minutes each day. The exercise sessions were all supervised by lab members and subjects were required to maintain a heart rate of 60 to 75% of their maximal heart rate achieved during a graded exercise test. The subjects were given an oral glucose tolerance test (OGTT) before the study and at the end of the seven day training period. The subjects' insulin sensitivity was assessed with post-challenge glucose levels, insulin levels, c-peptide levels, and the Matsuda insulin sensitivity index (ISI). Also, continuous glucose monitors were used to assess blood glucose levels for three days at baseline and for the final three days of the 7 day exercise training protocol.

CGMS monitors worn during the 5-7 days of exercise showed a statistically significant reduction in blood glucose levels in response to the standardized breakfast meal. Although the blood glucose level changes were not statistically significantly different during an OGTT, the blood glucose level changes due to mixed meals over the study may have been lower as indicated by the continuous glucose monitors. Because the monitors were not worn throughout the study, it is not known how each individual exercise session affected changes in PPG.

A recent study by Manders et al studied the effect of a single bout of either high or low intensity aerobic exercise on glycemic control (48). The ten subjects from this study were all male with long standing T2D. During the study the subjects participated in 3 different conditions: no exercise, low intensity exercise, and high intensity exercise.

The two exercise bouts were isocaloric, and the kcals were not replenished post-exercise. During all three periods the diet was provided by the lab and was calculated to meet each subject's needs using the Harris-Benedict equation. During the next 24 hours, blood glucose levels were monitored using a CGMS. The three conditions were then compared over the 24 hour period to determine how the conditions differed in glycemic control. Neither lunch, dinner, nor breakfast showed any statistically significant difference in ΔPPG. However, the 24 hour average glucose level was statistically significantly lower for the low intensity exercise versus no exercise; but the high intensity exercise was not. Also, the low intensity exercise showed a decrease in the prevalence of hyperglycemia versus no exercise while the high intensity exercise did not.

Overall the study seemed to show that the duration of the exercise may have been more important than the caloric expenditure of the exercise session. However, there may have been some stimulation of counter regulatory hormonal responses during the high intensity exercise which reduced the effectiveness of the exercise. Insulin sensitivity was not measured directly during this study, so whether the skeletal muscle was affected similarly by both exercise intensities is not known. Using the measure of blood glucose level does show that whether or not the skeletal muscle became more glucose tolerant, the overall control of blood glucose levels were not improved as much with high intensity exercise as they were with the low intensity exercise.

Activity to control blood glucose levels is part of normal human physiology (10, 53). People are evolved to be active, and it is only when we are getting normal amounts of physical activity that we display a normal healthy physiology (10). There are many

aspects of physiology which are affected by activity which can, during inactivity, lead to the development of chronic diseases (10, 16, 53). These include changes in insulin sensitivity, glucose tolerance, use of carbohydrate or lipid metabolism, and many other chronic risk factors (10, 53, 69). These factors can change the normal physiological health in an individual who is at risk of developing T2D (10, 53).

Technology and Methods

There have been dramatic increases in the technologies used to assess glucose tolerance, and insulin sensitivity. Previously, insulin sensitivity had to be assessed through blood samples. Fasting blood glucose levels or glucose challenges were used to assess insulin sensitivity and possible diabetes risk. Glucose challenges could be given either orally or intravenously. These required several blood draws to detect changes in blood glucose levels over time. Another option is the hyperinsulinemic euglycemic clamp. This is considered the best measure of insulin sensitivity, however it is more invasive, and is not without risk of hypoglycemia if not performed properly (23). Also, this method only measures a part of the total body components (skeletal muscle and hepatic insulin sensitivity) which account for glycemic control, while eliminating the role of endogenous insulin production. Unfortunately, none of these methods can accurately represent a free living mixed meal situation. While they are useful clinical measurements, they cannot show us the day to day affect of mixed meals on a diabetic individual's post-prandial glycemic excursions.

The fasting blood glucose level measure is the simplest measure; however it gives the least information about insulin sensitivity in the whole body. Because it is a fasted measure, the test does not show the person's ability to deal with a glucose challenge or mixed meal. Using either HOMAIR or QUICKI, insulin resistance or insulin sensitivity can be estimated (35). This test at best can measure the control of endogenous glucose output which is useful in assessing the sensitivity of hepatocytes to insulin, and give information about how far the disease has progressed. This is because in most cases, endogenous glucose output does not increase until a normal glucose tolerant person is already progressing to becoming impaired glucose tolerant (74). Although these are useful clinical measures, fasting blood glucose levels and insulin levels do not yield much information about the systemic insulin sensitivity and glucose tolerance.

Both oral glucose tolerance test (OGTT) and an intravenous glucose tolerance test (IVGTT) can assess glucose tolerance, and then insulin sensitivity can be calculated. During an OGTT a patient is given 75 grams of glucose in a beverage. Blood samples are collected repeatedly for at least 2 hours after the beverage is consumed. The glucose curve can then be plotted and the time course to bring glucose levels back within the pre-OGTT range can be assessed. An IVGTT works in the same way, but the patients' mass is taken into consideration, and the glucose is delivered intravenously. A set amount of glucose per kilogram of body mass is injected into the patient, and again blood samples are taken at several time points for hours after the injection. These methods can both show the changes in glucose levels over time. They are limited however, because they only see the time points when blood samples were taken. Also, to understand insulin

sensitivity, an insulin level curve must also be plotted to assess the tissue response to insulin. The OGTT is also not easily reproducible which requires more than one OGTT to estimate a good average (9). These two methods are a good way to estimate post-prandial responses, however because they only use glucose the effect of a mixed meal is not measured.

The hyperinsulinemic euglycemic clamp is the best measure of tissue sensitivity to insulin. During this test, insulin is infused at a constant rate into the patient. This would usually result in a hypoglycemic state; however, during the test glucose is also infused at a variable rate to maintain blood glucose levels at a constant value. By monitoring the amount of glucose being infused, the sensitivity of the tissue to the insulin can be assessed. If a large amount of glucose has to be infused to maintain euglycemia, the subject is very responsive to the insulin. On the other hand, if only a small amount of glucose has to be infused, the subject is not responding to the insulin. This is the best measure of insulin sensitivity; however it is more invasive than the others. Unfortunately it does not truly measure glycemic control because the pancreas as well as gut absorption and potential effects of incretins and other gut derived hormones are taken out of the equation. This test takes a long time to perform and requires multiple blood draws; it also carries the additional risk of hypoglycemia if performed incorrectly.

A more modern technology which allows researchers to measure glucose levels frequently throughout the day is the CGMS. CGMS measures blood glucose levels by sampling the interstitial fluid. A small probe can be implanted just beneath the skin on the torso and worn for several days. This allows researchers to measure changes from

meal to meal in a person's normal daily life. This new technology can be applied to real life questions, such as "does exercise reduce glycemic excursions throughout the day?" Although it does not give information about insulin, or insulin sensitivity directly, as the euglycemic clamp does, it gives useful practical information about how blood glucose levels are actually being controlled. Because people can wear the monitor for several days, it allows a look into changes over time in free living conditions, something which is almost impossible to measure with the other methods.

References

1) The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. The Diabetes Control and Complications Trial Research Group. N Engl J Med. 1993;329:977-86.

2) A. Hussain, M. Z. I. H., B. Claussen, S. Asghar.Type 2 Diabetes and Obesity: A Review. Journal of Diabetology. 2010;2:

3) Avramoglu, R. K., Qiu, W. and Adeli, K.Mechanisms of metabolic dyslipidemia in insulin resistant states: deregulation of hepatic and intestinal lipoprotein secretion. Front Biosci. 2003;8:d464-76.

4) Bakker, W., Eringa, E. C., Sipkema, P. and van Hinsbergh, V. W.Endothelial dysfunction and diabetes: roles of hyperglycemia, impaired insulin signaling and obesity. Cell Tissue Res. 2009;335:165-89.

5) Barr, R. G., Nathan, D. M., Meigs, J. B. and Singer, D. E.Tests of glycemia for the diagnosis of type 2 diabetes mellitus. Ann Intern Med. 2002;137:263-72.

6) Bastard, J. P., Maachi, M., Lagathu, C., Kim, M. J., Caron, M., Vidal, H., Capeau, J. and Feve, B.Recent advances in the relationship between obesity, inflammation, and insulin resistance. Eur Cytokine Netw. 2006;17:4-12.

7) Black, P. H.The inflammatory consequences of psychologic stress: relationship to insulin resistance, obesity, atherosclerosis and diabetes mellitus, type II. Med Hypotheses. 2006;67:879-91.

8) Bogardus, C., Thuillez, P., Ravussin, E., Vasquez, B., Narimiga, M. and Azhar, S.Effect of muscle glycogen depletion on in vivo insulin action in man. J Clin Invest. 1983;72:1605-10.

9) Bonora, E. and Muggeo, M.Postprandial blood glucose as a risk factor for cardiovascular disease in Type II diabetes: the epidemiological evidence. Diabetologia. 2001;44:2107-14.

10) Booth, F. W., Laye, M. J., Lees, S. J., Rector, R. S. and Thyfault, J. P.Reduced physical activity and risk of chronic disease: the biology behind the consequences. Eur J Appl Physiol. 2008;102:381-90.

11) Boule, N. G., Haddad, E., Kenny, G. P., Wells, G. A. and Sigal, R. J.Effects of exercise on glycemic control and body mass in type 2 diabetes mellitus: a meta-analysis of controlled clinical trials. JAMA. 2001;286:1218-27.

12) Brestoff, J. R., Clippinger, B., Spinella, T., von Duvillard, S. P., Nindl, B. C. and Arciero, P. J.An acute bout of endurance exercise but not sprint interval exercise enhances insulin sensitivity. Appl Physiol Nutr Metab. 2009;34:25-32.

13) Cartee, G. D. and Funai, K.Exercise and insulin: Convergence or divergence at AS160 and TBC1D1? Exerc Sport Sci Rev. 2009;37:188-95.

14) Cavalot, F., Pagliarino, A., Valle, M., Di Martino, L., Bonomo, K., Massucco, P., Anfossi, G. and Trovati, M.Postprandial blood glucose predicts cardiovascular events and

all-cause mortality in type 2 diabetes in a 14-year follow-up: lessons from the san luigi gonzaga diabetes study. Diabetes Care. 2011;34:2237-43.

15) Ceriello, A.Postprandial hyperglycemia and diabetes complications: is it time to treat? Diabetes. 2005;54:1-7.

16) Ceriello, A. and Motz, E.Is oxidative stress the pathogenic mechanism underlying insulin resistance, diabetes, and cardiovascular disease? The common soil hypothesis revisited. Arterioscler Thromb Vasc Biol. 2004;24:816-23.

17) Chen, Z. P., Stephens, T. J., Murthy, S., Canny, B. J., Hargreaves, M., Witters, L. A., Kemp, B. E. and McConell, G. K.Effect of exercise intensity on skeletal muscle AMPK signaling in humans. Diabetes. 2003;52:2205-12.

18) Christ-Roberts, C. Y., Pratipanawatr, T., Pratipanawatr, W., Berria, R., Belfort, R. and Mandarino, L. J.Increased insulin receptor signaling and glycogen synthase activity contribute to the synergistic effect of exercise on insulin action. J Appl Physiol. 2003;95:2519-29.

19) Colberg, S. R., Zarrabi, L., Bennington, L., Nakave, A., Thomas Somma, C., Swain, D. P. and Sechrist, S. R.Postprandial walking is better for lowering the glycemic effect of dinner than pre-dinner exercise in type 2 diabetic individuals. J Am Med Dir Assoc. 2009;10:394-7.

20) Cox, J. H., Cortright, R. N., Dohm, G. L. and Houmard, J. A.Effect of aging on response to exercise training in humans: skeletal muscle GLUT-4 and insulin sensitivity. J Appl Physiol. 1999;86:2019-25.

21) Da Ros, R., Assaloni, R. and Ceriello, A.The preventive anti-oxidant action of thiazolidinediones: a new therapeutic prospect in diabetes and insulin resistance. Diabet Med. 2004;21:1249-52.

22) De Souza, C. T., Frederico, M. J., da Luz, G., Cintra, D. E., Ropelle, E. R., Pauli, J. R. and Velloso, L. A.Acute exercise reduces hepatic glucose production through inhibition of the Foxo1/HNF-4alpha pathway in insulin resistant mice. J Physiol. 2010;588:2239-53.

23) DeFronzo, R. A., Tobin, J. D. and Andres, R.Glucose clamp technique: a method for quantifying insulin secretion and resistance. Am J Physiol. 1979;237:E214-23.

24) Delghingaro-Augusto, V., Decary, S., Peyot, M. L., Latour, M. G., Lamontagne, J., Paradis-Isler, N., Lacharite-Lemieux, M., Akakpo, H., Birot, O., Nolan, C. J., Prentki, M. and Bergeron, R.Voluntary running exercise prevents beta cell failure in susceptible islets of the Zucker diabetic fatty rat. Am J Physiol Endocrinol Metab. 2011;

25) Devlin, J. T. and Horton, E. S.Effects of prior high-intensity exercise on glucose metabolism in normal and insulin-resistant men. Diabetes. 1985;34:973-9.

26) Edgerton, D. S., Lautz, M., Scott, M., Everett, C. A., Stettler, K. M., Neal, D. W., Chu, C. A. and Cherrington, A. D.Insulin's direct effects on the liver dominate the control of hepatic glucose production. J Clin Invest. 2006;116:521-7.

27) Esposito, K., Nappo, F., Marfella, R., Giugliano, G., Giugliano, F., Ciotola, M., Quagliaro, L., Ceriello, A. and Giugliano, D.Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans: role of oxidative stress. Circulation. 2002;106:2067-72.

28) Faerch, K., Vaag, A., Holst, J. J., Hansen, T., Jorgensen, T. and Borch-Johnsen, K.Natural history of insulin sensitivity and insulin secretion in the progression from normal glucose tolerance to impaired fasting glycemia and impaired glucose tolerance: the Inter99 study. Diabetes Care. 2009;32:439-44.

29) Gastaldelli, A., Harrison, S. A., Belfort-Aguilar, R., Hardies, L. J., Balas, B., Schenker, S. and Cusi, K.Importance of changes in adipose tissue insulin resistance to histological response during thiazolidinedione treatment of patients with nonalcoholic steatohepatitis. Hepatology. 2009;50:1087-93.

30) Greenberg, A. S. and McDaniel, M. L.Identifying the links between obesity, insulin resistance and beta-cell function: potential role of adipocyte-derived cytokines in the pathogenesis of type 2 diabetes. Eur J Clin Invest. 2002;32 Suppl 3:24-34.

31) Haffner, S. M.The importance of hyperglycemia in the nonfasting state to the development of cardiovascular disease. Endocr Rev. 1998;19:583-92.

32) Henriksen, E. J.Invited review: Effects of acute exercise and exercise training on insulin resistance. J Appl Physiol. 2002;93:788-96.

33) Holloszy, J. O.A forty-year memoir of research on the regulation of glucose transport into muscle. Am J Physiol Endocrinol Metab. 2003;284:E453-67.

34) Hotamisligil, G. S., Arner, P., Caro, J. F., Atkinson, R. L. and Spiegelman, B. M.Increased adipose tissue expression of tumor necrosis factor-alpha in human obesity and insulin resistance. J Clin Invest. 1995;95:2409-15.

35) Hrebicek, J., Janout, V., Malincikova, J., Horakova, D. and Cizek, L.Detection of insulin resistance by simple quantitative insulin sensitivity check index QUICKI for epidemiological assessment and prevention. J Clin Endocrinol Metab. 2002;87:144-7.

36) Jacobson, P. B., von Geldern, T. W., Ohman, L., Osterland, M., Wang, J., Zinker, B., Wilcox, D., Nguyen, P. T., Mika, A., Fung, S., Fey, T., Goos-Nilsson, A., Grynfarb, M., Barkhem, T., Marsh, K., Beno, D. W., Nga-Nguyen, B., Kym, P. R., Link, J. T., Tu, N., Edgerton, D. S., Cherrington, A., Efendic, S., Lane, B. C. and Opgenorth, T. J.Hepatic glucocorticoid receptor antagonism is sufficient to reduce elevated hepatic glucose output and improve glucose control in animal models of type 2 diabetes. J Pharmacol Exp Ther. 2005;314:191-200.

37) Jensen, T. E., Schjerling, P., Viollet, B., Wojtaszewski, J. F. and Richter, E. A.AMPK alpha1 activation is required for stimulation of glucose uptake by twitch contraction, but not by H2O2, in mouse skeletal muscle. PLoS One. 2008;3:e2102.

38) Kahn, S. E.Clinical review 135: The importance of beta-cell failure in the development and progression of type 2 diabetes. J Clin Endocrinol Metab. 2001;86:4047-58.

39) Kahn, S. E., Hull, R. L. and Utzschneider, K. M.Mechanisms linking obesity to insulin resistance and type 2 diabetes. Nature. 2006;444:840-6.

40) Kelley, D. E., Goodpaster, B., Wing, R. R. and Simoneau, J. A.Skeletal muscle fatty acid metabolism in association with insulin resistance, obesity, and weight loss. Am J Physiol. 1999;277:E1130-41.

41) King, D. S., Dalsky, G. P., Clutter, W. E., Young, D. A., Staten, M. A., Cryer, P. E. and Holloszy, J. O.Effects of exercise and lack of exercise on insulin sensitivity and responsiveness. J Appl Physiol. 1988;64:1942-6.

42) Kirwan, J. P., Solomon, T. P., Wojta, D. M., Staten, M. A. and Holloszy, J. O.Effects of 7 days of exercise training on insulin sensitivity and responsiveness in type 2 diabetes mellitus. . Am J Physiol Endocrinol Metab. 2009;297:E151-6.

43) Knowler, W. C., Barrett-Connor, E., Fowler, S. E., Hamman, R. F., Lachin, J. M., Walker, E. A. and Nathan, D. M.Reduction in the incidence of type 2 diabetes with lifestyle intervention or metformin. N Engl J Med. 2002;346:393-403.

44) Kraniou, G. N., Cameron-Smith, D. and Hargreaves, M.Acute exercise and GLUT4 expression in human skeletal muscle: influence of exercise intensity. J Appl Physiol. 2006;101:934-7.

45) Larsen, J. J., Dela, F., Kjaer, M. and Galbo, H.The effect of moderate exercise on postprandial glucose homeostasis in NIDDM patients. Diabetologia. 1997;40:447-53.

46) Lima, A. F., Ropelle, E. R., Pauli, J. R., Cintra, D. E., Frederico, M. J., Pinho, R. A., Velloso, L. A. and De Souza, C. T.Acute exercise reduces insulin resistance-induced TRB3 expression and amelioration of the hepatic production of glucose in the liver of diabetic mice. J Cell Physiol. 2009;221:92-7.

47) Little, J. P., Gillen, J. B., Percival, M., Safdar, A., Tarnopolsky, M. A., Punthakee, Z., Jung, M. E. and Gibala, M. J.Low-volume high-intensity interval training reduces hyperglycemia and increases muscle mitochondrial capacity in patients with type 2 diabetes. J Appl Physiol. 2011;

48) Manders, R. J., Van Dijk, J. W. and van Loon, L. J.Low-intensity exercise reduces the prevalence of hyperglycemia in type 2 diabetes. Med Sci Sports Exerc. 2010;42:219-25.

49) Merry, T. L., Steinberg, G. R., Lynch, G. S. and McConell, G. K.Skeletal muscle glucose uptake during contraction is regulated by nitric oxide and ROS independently of AMPK. Am J Physiol Endocrinol Metab. 2010;298:E577-85.

50) Mikus, C. R., Fairfax, S. T., Libla, J. L., Boyle, L. J., Vianna, L. C., Oberlin, D. J., Uptergrove, G. M., Deo, S. H., Kim, A., Kanaley, J. A., Fadel, P. J. and Thyfault, J. P.Seven days of aerobic exercise training improves conduit artery blood flow following glucose ingestion in patients with type 2 diabetes. J Appl Physiol. 2011;

51) Monnier, L., Colette, C., Dunseath, G. J. and Owens, D. R.The loss of postprandial glycemic control precedes stepwise deterioration of fasting with worsening diabetes. Diabetes Care. 2007;30:263-9.

52) Nichols, G. A., Hillier, T. A. and Brown, J. B.Progression from newly acquired impaired fasting glusose to type 2 diabetes. Diabetes Care. 2007;30:228-33.

53) Pedersen, B. K.The diseasome of physical inactivity--and the role of myokines in muscle--fat cross talk. J Physiol. 2009;587:5559-68.

54) Richter, E. A., Mikines, K. J., Galbo, H. and Kiens, B.Effect of exercise on insulin action in human skeletal muscle. J Appl Physiol. 1989;66:876-85.

55) Riddle, M. C., Rosenstock, J. and Gerich, J.The treat-to-target trial: randomized addition of glargine or human NPH insulin to oral therapy of type 2 diabetic patients. Diabetes Care. 2003;26:3080-6.

56) Rockl, K. S., Witczak, C. A. and Goodyear, L. J.Signaling mechanisms in skeletal muscle: acute responses and chronic adaptations to exercise. IUBMB Life. 2008;60:145-53.

57) Ropelle, E. R., Pauli, J. R., Cintra, D. E., Frederico, M. J., de Pinho, R. A., Velloso, L. A. and De Souza, C. T.Acute exercise modulates the Foxo1/PGC-1alpha pathway in the liver of diet-induced obesity rats. J Physiol. 2009;587:2069-76.

58) Sattley, M.The History of Diabetes. Diabetes Health. 2009;

59) Saydah, S. H., Miret, M., Sung, J., Varas, C., Gause, D. and Brancati, F. L.Postchallenge hyperglycemia and mortality in a national sample of U.S. adults. Diabetes Care. 2001;24:1397-402.

60) Scheen, A. J.Treatment of type 2 diabetes. Acta Clin Belg. 2003;58:318-24.

61) Segerstrom, A. B., Glans, F., Eriksson, K. F., Holmback, A. M., Groop, L., Thorsson, O. and Wollmer, P.Impact of exercise intensity and duration on insulin sensitivity in women with T2D. Eur J Intern Med. 2010;21:404-8.

62) Sigal, R. J., Kenny, G. P., Boule, N. G., Wells, G. A., Prud'homme, D., Fortier, M., Reid, R. D., Tulloch, H., Coyle, D., Phillips, P., Jennings, A. and Jaffey, J.Effects of aerobic training, resistance training, or both on glycemic control in type 2 diabetes: a randomized trial. Ann Intern Med. 2007;147:357-69.

63) Sigal, R. J., Kenny, G. P., Wasserman, D. H. and Castaneda-Sceppa, C.Physical activity/exercise and type 2 diabetes. Diabetes Care. 2004;27:2518-39.

64) Solomon, T. P., Haus, J. M., Li, Y. and Kirwan, J. P.Progressive hyperglycemia across the glucose tolerance continuum in older obese adults is related to skeletal muscle

capillarization and nitric oxide bioavailability. J Clin Endocrinol Metab. 2011;96:1377-84.

65) Steinberg, H. O., Chaker, H., Leaming, R., Johnson, A., Brechtel, G. and Baron, A. D.Obesity/insulin resistance is associated with endothelial dysfunction. Implications for the syndrome of insulin resistance. J Clin Invest. 1996;97:2601-10.

66) Thomson, M. J., Williams, M. G. and Frost, S. C.Development of insulin resistance in 3T3-L1 adipocytes. J Biol Chem. 1997;272:7759-64.

67) Thyfault, J. P.Setting the stage: possible mechanisms by which acute contraction restores insulin sensitivity in muscle. Am J Physiol Regul Integr Comp Physiol. 2008;294:R1103-10.

68) Thyfault, J. P., Cree, M. G., Zheng, D., Zwetsloot, J. J., Tapscott, E. B., Koves, T. R., Ilkayeva, O., Wolfe, R. R., Muoio, D. M. and Dohm, G. L.Contraction of insulin-resistant muscle normalizes insulin action in association with increased mitochondrial activity and fatty acid catabolism. Am J Physiol Cell Physiol. 2007;292:C729-39.

69) Thyfault, J. P., Rector, R. S. and Noland, R. C.Metabolic inflexibility in skeletal muscle: a prelude to the cardiometabolic syndrome? J Cardiometab Syndr. 2006;1:184-9.

70) Translation, C. s. D. o. D.Long-term trends in diabetes. 2010;

71) Umpierre, D., Ribeiro, P. A., Kramer, C. K., Leitao, C. B., Zucatti, A. T., Azevedo, M. J., Gross, J. L., Ribeiro, J. P. and Schaan, B. D.Physical activity advice only or

structured exercise training and association with HbA1c levels in type 2 diabetes: a systematic review and meta-analysis. JAMA. 2011;305:1790-9.

72) Ussher, J. R., Koves, T. R., Cadete, V. J., Zhang, L., Jaswal, J. S., Swyrd, S. J., Lopaschuk, D. G., Proctor, S. D., Keung, W., Muoio, D. M. and Lopaschuk, G. D.Inhibition of de novo ceramide synthesis reverses diet-induced insulin resistance and enhances whole-body oxygen consumption. Diabetes. 2010;59:2453-64.

73) Utzschneider, K. M. and Kahn, S. E.Review: The role of insulin resistance in nonalcoholic fatty liver disease. J Clin Endocrinol Metab. 2006;91:4753-61.

74) Weyer, C., Bogardus, C., Mott, D. M. and Pratley, R. E.The natural history of insulin secretory dysfunction and insulin resistance in the pathogenesis of type 2 diabetes mellitus. J Clin Invest. 1999;104:787-94.

75) Wright, E., Jr., Scism-Bacon, J. L. and Glass, L. C.Oxidative stress in type 2 diabetes: the role of fasting and postprandial glycaemia. Int J Clin Pract. 2006;60:308-14.

76) Xu, H., Barnes, G. T., Yang, Q., Tan, G., Yang, D., Chou, C. J., Sole, J., Nichols, A., Ross, J. S., Tartaglia, L. A. and Chen, H.Chronic inflammation in fat plays a crucial role in the development of obesity-related insulin resistance. J Clin Invest. 2003;112:1821-30.

77) Yudkin, J. S., Stehouwer, C. D., Emeis, J. J. and Coppack, S. W.C-reactive protein in healthy subjects: associations with obesity, insulin resistance, and endothelial dysfunction: a potential role for cytokines originating from adipose tissue? Arterioscler Thromb Vasc Biol. 1999;19:972-8.

APPENDIX B

SUPPLEMENTAL MATERIALS

Table 1 Vitamin content of meal

Item Name	Water (g)	Vit A (RAE)	Vit B1 (mg)	Vit B2 (mg)	Vit B3 (mg)	Vit B6 (mg)
Bread, white, enriched, sandwich	16.92	0.00	0.24	0.14	1.60	--
Potatoes, baked, salted	104.85	0.70	0.09	0.07	1.97	0.44
Juice, punch, indv box	--	0.00	--	--	--	--
Beef, ground, hamburger, raw, 10% fat	70.19	0.00	0.04	0.16	5.12	0.37
Butter, whipped, sweet cream, light, salted	--	--	--	--	--	--
Salsa, mild	29.14	5.32	--	--	--	--
Oil, olive, light	0.00	0.00	--	--	--	--
Applesauce, swtnd, cnd	80.34	0.29	0.02	0.02	0.07	0.03
Cheese, cheddar, medium	4.95	0.00	--	--	--	--
Total	306.39	6.32	0.39	0.38	8.77	0.83
% Recommendation	24.84	2.11	97.17	88.23	164.39	192.59

Item Name	Vit B12 (mcg)	Biot (mcg)	Vit C (mg)	Vit D (mcg)	Vit E-a-Toco (mg)	Folate (mcg)	Vit K (mcg)	Panto (mg)
Bread, white, enriched, sandwich	--	--	0.00	--	--	48.00	--	--
Potatoes, baked, salted	0.00	--	13.44	0.00	0.06	39.20	2.80	0.53
Juice, punch, indv box	--	--	60.00	--	--	--	--	--
Beef, ground, hamburger, raw, 10% fat	2.23	--	0.00	0.10	0.32	6.06	0.81	0.60
Butter, whipped, sweet cream, light, salted	--	--	0.00	--	--	--	--	--
Salsa, mild	--	--	5.11	--	--	--	--	--
Oil, olive, light	--	--	0.00	--	--	--	--	--
Applesauce, swtnd, cnd	0.00	0.78	1.67	0.00	0.18	0.98	0.59	0.04
Cheese, cheddar, medium	--	--	0.00	--	--	--	--	--
Total	2.23	0.78	80.22	0.10	0.56	94.24	4.20	1.17
% Recommendation	279.01	7.84	267.39	2.02	11.11	70.68	10.49	70.23

Table 1 Shows the vitamin content of the meal.

Table 2 Mineral content of meal

Item Name	Ca (mg)	Ch (mcg)	Cu (mg)	Fl (mg)	I (mcg)	Fe (mg)
Bread, white, enriched, sandwich	80	--	--	--	--	1.44
Potatoes, baked, salted	21	--	0.17	--	--	1.51
Juice, punch, indv box	0	--	--	--	--	0.36
Beef, ground, hamburger, raw, 10% fat	12.12	--	0.07	0.02	--	2.26
Butter, whipped, sweet cream, light, salted	0	--	--	--	--	0
Salsa, mild	0	--	--	--	--	0
Oil, olive, light	0	--	--	--	--	0
Applesauce, swtnd, cnd	2.94	--	0.03	--	--	0.12
Cheese, cheddar, medium	0	--	--	--	--	0
Total	116.06	--	0.27	0.02	--	5.69
% Recommendation	34.82		89.76	1.7		213.45

Item Name	Mg (mg)	Mn (mg)	P (mg)	K (mg)	Se (mcg)	Na (mg)	Zn (mg)
Bread, white, enriched, sandwich	--	--	--	--	--	270	--
Potatoes, baked, salted	39.2	0.31	98	749	0.56	14	0.5
Juice, punch, indv box	--	--	--	250	--	10	--
Beef, ground, hamburger, raw, 10% fat	20.2	0.01	185.84	324.21	16.77	66.66	4.84
Butter, whipped, sweet cream, light, salted	--	--	--	--	--	85	--
Salsa, mild	--	--	--	--	--	138.39	--
Oil, olive, light	--	--	--	--	--	0	--
Applesauce, swtnd, cnd	2.94	0.03	5.88	73.5	0.29	1.96	0.03
Cheese, cheddar, medium	--	--	--	--	--	90	--
Total	62.34	0.35	289.72	1396.71	17.62	676.01	5.37
% Recommendation	44.53	45.14	124.17	89.15	96.11	135.2	146.49

Table 2 shows the mineral content of the meal.

Table 3. Subject medications

	blood pressure	blood pressure	Blood pressure and dieabetic neropathy	Blood pressure	blood pressure	Insulin Sensitivity	sulfonylurea	GLP-1 analog	GLP-1 analog	Muscle IS	Blood lipids	cholesterol	cholesterol	cholesterol	cholesterol
subject	lisinopril	Hydrochlorothiazide	Losartan Potassium	Benicar	Furosemide	Pioglitazone	Glipizide	Liraglutide	Exenatide	Metformin	Fenofibrate	Simvastatin	Pravastatin	Rosuvastatin	Lovastatin
106	5mg									500mg		40mg			
109	40mg	12.5mg								1000mg			40mg		
118	10mg							1.8mg		1000mg		20mg			
119	10mg								10mcg x2	500mg x2		80mg			
121		25mg	50mg			45mg								10mg	
137	20mg	60mg								500mg x2	145mg			40mg	
145	20mg											20mg			
151				20mg								20mg			
154			50mg		40mg		5mg			500mg 2x					20mg

Table 3. This table shows the subject medications that are associated with their chronic disease conditions. Other medications for problems unrelated to diseases of obesity and glycemic control have been omitted from this table.

CONSENT FORM TO PARTICIPATE IN A RESEARCH STUDY

INVESTIGATOR'S NAME: JOHN P. THYFAULT, PHD
PROJECT #:1187377
STUDY TITLE: GLYCEMIC CONTROL IN TYPE II DIABETICS.

INTRODUCTION

This consent may contain words that you do not understand. Please ask the investigator or the study staff to explain any words or information that you do not clearly understand.

This is a research study. Research studies include only people who choose to participate. As a study participant you have the right to know about the procedures that will be used in this research study so that you can make the decision whether or not to participate. The information presented here is simply an effort to make you better informed so that you may give or withhold your consent to participate in this research study.

Please take your time to make your decision and discuss it with your family and friends.

You are being asked to take part in this study because you have type 2 diabetes or pre-diabetes.

This study is being sponsored by the MU Institute for Clinical and Translational Sciences.

In order to participate in this study, it will be necessary to give your written consent.

WHY IS THIS STUDY BEING DONE?

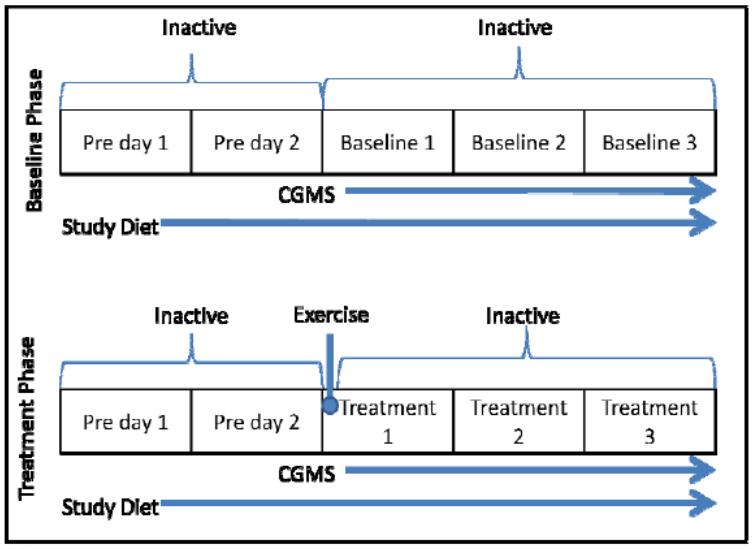
The purpose of this study is to determine how long a single bout of exercise improves the blood glucose (sugar) concentrations of people with type 2 diabetes.

This research is being done because exercise has been shown to lower blood sugar by increasing the ability of the muscles to remove glucose from the blood. This study is being done because it is unclear how long the glucose lowering effects of one session of exercise last.

HOW MANY PEOPLE WILL TAKE PART IN THE STUDY?

Approximately 100 people will be screened to identify 30 people who are eligible to take part in this study.

WHAT IS INVOLVED IN THE STUDY?



Example Schedule:

- *Visit 1: basic measures (height, weight, blood pressure, etc.) and given food log*
- *Visit 2: exercise stress test followed by a 5 to 15 day washout.*
- *Visit 3: pick up food and start study diet*
- *Visit 4: on the second evening attach CGMS and continue study diet for up to 3 days*
- *Visit 5: remove CGMS and stop study diet. Consume regular diet with regular activity for 5 to 15 days*
- *Visit 6: pick up food and begin study diet a second time*
- *Visit 7: on the second evening attach CGMS a second time*
- *Visit 8: on the next day come to McKee for one 60 minute exercise bout. Continue to eat the study diet for up to 3 more days*
- *Visit 9: remove CGMS and stop study diet*
- *The order of the CGMS monitoring periods (active versus inactive) will be randomized. In other words, some participants will complete the CGMS monitoring with exercise prior to the monitoring period without exercise.*

If you take part in this study, you will have the following tests and procedures:

You will initially be screened to see if you meet the criteria of the study.

Visit 1 will take place at McKee Gymnasium. This visit will last approximately 60 minutes and will include the following:

- ✓ Basic Measures: You will answer questionnaires on the following: medical history, any medication allergies, diet, physical activity, and menstrual cycle. You will have your height, body weight, waist circumference, and blood pressure measured.

- ✓ Blood draw: You will have 100 ml (<1/2 cup) of blood withdrawn from a vein in your arm. This is considerably less than the 450 mls of blood collected during blood donations. We will measure blood fats, glucose, inflammation, and hormone levels in these samples. Urine will be collected to test for markers of liver and kidney function.

- ✓ You will be given a food log to fill out over 3 days of normal diet and activity to assess caloric requirements, and a step log to record number of steps per day.

Visit 2 will take place at McKee Gymnasium. This visit will last approximately 60 minutes and will include the following:

- ✓ Medical Exam: A physician will perform a brief medical exam to ensure that it is safe for you to participate in this study. The physician may check your heart and breathing rates, look at your eyes and feet, and ask you questions about your medication use and medical history. *This is not a comprehensive medical exam and is not intended to replace a visit with your personal physician. This study is neither designed nor staffed to provide comprehensive medical care to participants. All participants are required to receive their diabetes and general health care from their personal physicians.*

- ✓ We will also perform a scan, called a DXA scan, to determine how much of your body is composed of fat, bone and muscle. This scan will expose you to a small amount of radiation, similar to what you receive if you stood in the sun all day. For this reason, women of child-bearing age will be administered a pregnancy test to ensure they are not pregnant prior to the scan.

- ✓ Exercise Test: The exercise test will be performed on a treadmill (or stationary bicycle). Prior to starting the test, electrodes will be placed on your chest and abdomen so that we may monitor your heart's activity during the test. You will be asked to wear a nose-clip during the test and to breathe through a mouthpiece attached to a machine that measures the amount of oxygen you consume and carbon dioxide you expel. The test begins with 3 minutes of very slow walking (or cycling). Next, the speed and incline of the treadmill (or resistance on the bicycle) will be increased every 3 minutes until you reach exhaustion. The test is typically over within 9-15 minutes. You may be asked to check your blood glucose levels before and/or after this test. If your blood glucose levels are too low, you may be asked to consume a small snack, and if they are too high, your test may be rescheduled.

Visit 3 will take place at McKee Gymnasium. This will last approximately 30 minutes and will include the following:

- ✓ You will receive your study diet food and a diet history sheet to record what you eat. We will remind you of how to record your diet including what you eat or do not eat.

Visit 4 will take place at McKee Gymnasium. This visit will last approximately 45 minutes and will include the following:

- ✓ Continuous Glucose Monitoring: You will be asked to come to the Exercise Physiology lab for placement and removal of the continuous glucose monitor. The monitor will be placed by trained study staff under sterile conditions. After cleaning a small area on your abdomen or back, a small tube (catheter) approximately ½ the size of a sewing needle will be placed under your skin. The tube is attached to a wire connected to a small device approximately the size of a small cellular phone that will be worn on your belt during the 3 days of glucose monitoring. This device measures and records your blood glucose values throughout the day. In addition, it may be necessary for you to perform finger sticks (4-5 each day) to monitor your blood glucose. This will be done once during days in which you maintain normal levels of daily physical activity and again during the final 3 days of the exercise intervention. You will be asked to keep a food diary during the first 3 days of glucose monitoring. During the second 3 day period of glucose monitoring, it will be very important that you repeat the timing, content and quantity of food consumption. It is very important that you consume adequate carbohydrate during these periods. We will provide sample menus that you will be asked to follow. We may also provide some standardized meals and/or meal vouchers for local eateries. You will also be asked to wear a pedometer (a small device that clips onto your belt or pants and counts the number of steps you take each day) throughout the study and record your daily steps using an accelerometer.

Visit 5 will include having the CGMS removed, and will last approximately 15 minutes.
- ✓ You will have to come to McKee Gymnasium to have the CGMS monitor removed and to turn in your diet log.

Visit 6 will take place at McKee Gymnasium, and will last approximately 10 minutes.
- ✓ You will come to McKee Gymnasium and get the same study food which you consumed during the first phase of the study and receive a second diet log.

Visit 7 will take place at McKee Gymnasium. This visit will last approximately 45 minutes and will include the following:
- ✓ Continuous Glucose Monitoring: as described in Visit 4.

Visit 8 will take place at McKee Gymnasium, and will last approximately 90 minutes
- ✓ You will come to McKee Gymnasium and exercise on a treadmill for 60 minutes. The treadmill will be set at a speed and grade to produce a heart rate which is 75% of the maximum heart rate you achieved during the treadmill max test. During the exercise you will have access to water, towels, fans, and television if you would like them. Before and after the exercise session, your blood glucose and blood pressure will be taken to ensure that you are not at risk. If your blood glucose becomes too low food, juice, or glucose tabs will be available to ensure your safety.

Visit 9 will include having the CGMS removed, and will last approximately 15 minutes.
- ✓ You will have to come to McKee Gymnasium to have the CGMS monitor removed and to turn in your diet log.

Your decision in the Optional portions below will not affect your participation in this study.

Optional: Because new markers for health and disease are discovered frequently, and could possibly provide invaluable information, we would like to save small amounts of your blood to be used for later analysis. We ask that you give approval for these tests to be performed using these samples. Your samples would be destroyed after a maximum of 5 years. If you change your mind in the future you can call Dr. Thyfault and the blood samples will be immediately discarded.

Do you Accept_____or Decline_____ for us to save blood?
Initial _____

We also ask your permission to share your samples with other investigators for use with similar research involving the metabolic syndrome, cardiovascular disease or diabetes. Any information identifying you will not be shared with other investigators. Because the samples will be shared without identifiers there will be no way to track your samples or destroy them in the future if you change your mind.

Do you Accept_____or Decline_____ for us to share blood?
Initial _____

I authorize the investigators to keep this information and any information from my participation in their studies in a database so that they may contact me regarding future studies.
YES _____ **NO** _____

HOW LONG WILL I BE IN THE STUDY?

You will be in the study for approximately 3 weeks. This will include an initial consent, and three days of filling out a food log. Then there will be up to five days of study diet, four of which you will wear the CGMS monitor. This will be followed by a 5 to 15 day dietary wash out period. Finally there will be up to five more days of study diet. On the second day you will perform 60 minutes of exercise, and wear a CGMS for up to three days.

The investigator and/or your doctor may decide to end your participation in the study, if in their judgment that it is in your best interest.

You can stop participating at any time. Your decision to withdraw from the study will not affect in any way your medical care and/or benefits.

WHAT ARE THE RISKS OF THE STUDY?

While on the study, you are at risk for the side effects described below. You should discuss these with the investigator and/or your doctor. There may also be other side effects that we cannot predict. Many side effects go away shortly after the testing and intervention are stopped, but in some cases side effects can be serious or long-lasting or permanent.

Risks and side effects related to the procedures in this study include:

Graded Exercise Test: Participants may experience dizziness, nausea, fainting, or muscle fatigue and/or soreness during and/or following the graded exercise test. Most symptoms disappear within minutes of stopping the test. Muscle soreness usually fades within 48-72 hours. There is also a very small risk of heart complications during this test. In the event you experience any heart problems, we will stop the exercise session immediately and evaluate if you need medical attention. This test will be performed by a physician.

ECG: Some people may have a skin irritation from the patches that connect the wires on your chest to the computer. Skin and hair are pulled slightly when the patches are removed after the test. Research personnel will attach and remove the patches as carefully as possible.

Exercise Training: You may experience muscle soreness or fatigue after exercise.

Blood Sampling: Possible risks associated with the blood sampling are nausea, bruising, and a small chance of infection. To minimize risk, the procedure will be performed with sterile techniques by qualified personnel.

Glucose monitoring: There is a small risk of infection, bruising, or nausea associated with the continuous glucose monitoring. To minimize risk, the procedure will be performed with sterile techniques by qualified personnel in the Exercise Physiology Laboratory.

Blood pressure cuff inflation: The blood pressure cuff will squeeze your arm tightly; however, any discomfort will be alleviated as soon as the pressure in the cuff is released.

Reproductive risks: Because the DXA scan in this study can affect an unborn baby, if you are a woman, you should not become pregnant while on this study. If you have any questions about the reproductive issues or about preventing pregnancy, please discuss them with the investigator or your doctor.

You will be exposed to a small amount of radiation during the DXA scan. Radiation effects are cumulative. You should always inform future doctors of your participation in this study. The radiation you will receive will not exceed the amount of radiation received in one chest X-ray.

For the reasons stated above the investigator will observe you closely while giving the treatment described and, if you have any worrisome symptoms or symptoms that the investigator or his associates have described to you, notify the investigator or study physician immediately. The study physician may be reached at 573-882-4141. Dr. John Thyfault's telephone number is 573-268-2131. For more information about risks and side effects, ask the investigator or contact the study coordinator at 573-882-6892 or umchesexphysstudy@missouri.edu.

Are There Benefits to Taking Part in the Study?

If you agree to take part in this study, there may or may not be direct medical benefit to you. You may expect to benefit from taking part in this research to the extent that you are contributing to medical knowledge. We hope the information learned from this study will benefit other patients with type 2 diabetes in the future.

Other benefits include an overall assessment of your metabolic health including: fitness evaluation, dietary analysis, and continuous glucose monitoring. You are free to share this information with your personal physician if you choose.
Although most research suggests that exercise training is beneficial for individuals with type 2 diabetes, there is no guarantee that taking part in this research will result in any improvement in your condition.

WHAT OTHER OPTIONS ARE THERE?

Instead of being in this study, you have these options:
You may get some of these tests done by your personal physician and may start an exercise program on your own even if you do not take part in the study.

An alternative is to not participate in this research study.

Please discuss these and other options with the investigator and your doctor.

WHAT ABOUT CONFIDENTIALITY?

Information produced by this study will be stored in the investigators file and identified by only a code number. The code key connecting your name to your information will be kept in a separate and secure location. Your information in these records will not be given to anyone unaffiliated with the study in a form that could identify you without written consent, expect as required by law. If the investigator conducting this study is not your primary or regular doctor, he must obtain your permission before contacting your regular doctor for information about your past medical history or to inform them that you are taking part in this study.

Results of this research may be published and reports may be made to government agencies, funding agencies, manufacturers, or scientific groups, but you will not be identified in any such publication or report. In addition, the Federal Food and Drug administration, MU IRB, other government agencies, or the manufacturer of the drug(s) used in this study may inspect and copy your medical records that apply to this research. In all cases, information about you will be treated confidentially.

The results of this study may be published in a medical book or journal or used for teaching purposes. However, your name or other identifying information will not be used in any publication or teaching materials without your specific permission.

In addition, if photographs, audiotapes or videotapes were taken during the study that could identify you, then you must give special written permission for their use. In that case, you will be given the opportunity to view or listen, as applicable, to the photographs, audiotapes or videotapes before you give your permission for their use if you so request.

WHAT ARE THE COSTS?

There is no cost to you for the participating in the study.
All costs for the measurements and procedures that are part of this research study will be paid by the MU Exercise Physiology Laboratory. Your only cost will be traveling to the laboratory on a regular basis

You or your insurance company will be charged for continuing medical care and/or hospitalization not related to your participation in the study.

Will I be Paid for Participating in the Study?

You will be compensated up to $200 for completing the entire study

What if I am Injured?

It is not the policy of the University of Missouri to compensate human subjects in the event the research results in injury. The University of Missouri, in fulfilling its public responsibility, has provided medical, professional and general liability insurance coverage for any injury in the event such injury is caused by the negligence of the University of Missouri, its faculty and staff. The University of Missouri also will provide, within the limitations of the laws of the State of Missouri, facilities and medical attention to subjects who suffer injuries while participating in the research projects of the University of Missouri. In the event you have suffered injury as the result of participation in this research program, you are to contact the Risk Management Officer, telephone number (573) 882-1181, at the Health Sciences Center, who can review the matter and provide further information. This statement is not to be construed as an admission of liability.

What Are My Rights as a Participant?

Participation in this study is voluntary. You do not have to participate in this study. Your present or future care will not be affected should you choose not to participate. If you decide to participate, you can change your mind and drop out of the study at any time without affecting your present or future care in the University of Missouri Hospital. Leaving the study will not result in any penalty or loss of benefits to which you are entitled. In addition, the investigator of this study may decide to end your participation in this study at any time after he has explained the reasons for doing so and has helped arrange for your continued care by your own doctor, if needed.

You will be informed of any significant new findings discovered during the course of this study that might influence your health, welfare, or willingness to continue participation in this study.

Whom Do I Call if I Have Questions or Problems?

Questions or problems about the research can be addressed to the Study Representative at 573-882-6892. In addition, any concerns or complaints about the research can be addressed to the investigator at 573-882-9818.

If you have any questions regarding your rights as a participant in this research and/or concerns about the study, or if you feel under any pressure to enroll or to continue to participate in this study, you may contact the University of Missouri Health Sciences Institutional Review Board (which is a group of people who review the research studies to protect participants' rights) at (573) 882-3181.

You may ask more questions about the study at any time. For questions about the study or a research-related injury, contact the study coordinator at 573-882-6892.

A copy of this consent form will be given to you to keep.

SIGNATURE

I confirm that the purpose of the research, the study procedures, the possible risks and discomforts as well as potential benefits that I may experience have been explained to me. Alternatives to my participation in the study also have been discussed. I have read this consent form and my questions have been answered. My signature below indicates my willingness to participate in this study.

Subject/Patient*	Date
Legal Guardian/Advocate/Witness (if required)**	Date
Additional Signature (if required) (identify relationship to subject)***	Date

*A minor's signature on this line indicates his/her assent to participate in this study. A minor's signature is not required if he/she is under 7 years old. Use the "Legal Guardian/Advocate/Witness" line for the parent's signature, and you may use the "Additional Signature" line for the second parent's signature, if required.

**The presence and signature of an impartial witness is required during the entire informed consent discussion if the patient or patient's legally authorized representative is unable to read.

***The "Additional Signature" line may be used for the second parent's signature, if required. This line may also be used for any other signature which is required as per federal, state, local, sponsor and/or any other entity requirements.

"If required" means that the signature line is signed only if it is required as per federal, state, local, sponsor and/or any other entity requirements.

SIGNATURE OF STUDY REPRESENTATIVE

I have explained the purpose of the research, the study procedures, identifying those that are investigational, the possible risks and discomforts as well as potential benefits and have answered questions regarding the study to the best of my ability.

Study Representative****	Date

****Study Representative is a person authorized to obtain consent. Per the policies of the University of Missouri Health Care, for any 'significant risk/treatment' study, the Study Representative must be a physician who is either the Principal or Co-Investigator. If the study is deemed either 'significant risk/non-treatment' or 'minimal risk,' the Study Representative may be a non-physician study investigator.

HS IRB USE ONLY
Approval Date: May 31, 2011
Expiration Date: April 13, 2012

Vita

NAME Douglas J. Oberlin	Position Title Graduate Research Assistant

Education

Institution	Dates Attended	Degree	Year	Field of Study
University of Missouri Columbia, MO	2003 – 2009	B.S.	2009	Nutrition and Fitness
University of Missouri Columbia, MO	2009 – 2011	M.S.	2011	Exercise Physiology

Publications

Mikus CR, Libla JL, Fairfax ST, Boyle LJ, Vianna L, Oberlin DJ, Uptergrove GM, Deo SH, Kim A, Kanaley JA, Fadel PJ, Thyfault JP (2011). Seven days of aerobic exercise augments skeletal muscle blood flow responses to a glucose load in patients with type 2 diabetes. (In preparation).

Catherine R. Mikus, Ph.D., Douglas J. Oberlin, B.S., Jessica Libla, B.S., Leryn J. Boyle, M.S., and John P. Thyfault, Ph.D. Glycemic control is improved in patients with type 2 diabetes by seven days of aerobic exercise training. (In review).

Mikus, Catherine R.; Fairfax, Seth T.; Boyle, Leryn J.; Vianna, Lauro; Oberlin, Douglas J.; Deo, Shekhar H.; Kim, Areum; Kanaley, Jill A. FACSM; Fadel, Paul J. FACSM; Thyfault, John P. (2010). Seven Days of Aerobic Exercise Improves Hyperemic Responses to Glucose Ingestion in Patients with T2DM. *Medicine & Science in Sports & Exercise*, Vol. 42, 34

Boyle, Leryn J.; Mikus, Catherine R.; Libla, Jessica L.; Oberlin, Douglas J.; Fadel, Paul J. FACSM; Thyfault, John P. (2010). GIP and GLP Responses to a Glucose Challenge after Seven Days of Exercise Training. *Medicine & Science in Sports & Exercise*, Vol. 42, 87

Presentations

Oberlin, Douglas J.; Mikus, Catherine R. (2010). Physical inactivity rapidly alters glycemic control in young, lean, previously active volunteers MU Health Sciences Research Day (*Poster Presentation*)

Oberlin, D.J.; Mikus, C.R.; Thyfault, J.P. (2010). Dietary Protein and Glucose Intake Do Not Correlate With Postprandial Glucose ACSM CS (*Poster Presentation*)

DJ Oberlin, Catherine R. Mikus, Monica L. Kearney, Justin A. Fletcher Pam S. Hinton, Jill A. Kanaley FACSM, Randy S. Rector, Heather J. Leidy, John P. Thyfault. (2011) A SINGLE EXERCISE BOUT DOES NOT IMPROVE GLYCEMIC CONTROL IN VOLUNTEERS WITH TYPE 2 DIABETES. ACSM CS (*Poster Presentation*)

Experiences and Honors

2009 – 2011	Teaching Assistant for Nutrition Concepts and Controversies and Introduction to Exercise and Fitness, University of Missouri
2009- 2011	Member of the Health Sciences Graduate Student Association
2010	Edward J. O'Brien Scholorship, University of Missouri
2010	Ben Londeree Distinguished Graduate Student in Exercise Physiology Award, University of Missouri
2009-2010	Fitness instructor at Boone Hospital's fitness center
2008	Intern for Boone Hospital's Cardiac Rehabilitation Program