Understanding the utility of active learning by conducting science to teach science

by

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Abstract

To retain students in science, technology, engineering, and mathematics (STEM) various pedagogical techniques, such as active learning, have been incorporated in STEM courses. Despite the popularity of active learning, it has rarely been empirically tested with a comparison or control group. To compare the benefits of active learning to conventional lecture learning, the aims of this project were twofold: conduct a physiological research project investigating thermoregulation in large ruminants and evaluate the effectiveness of active learning tools for an undergraduate course that focused on core concepts in physiology. Physiological concepts from the thermoregulation study were integrated into the educational portion of the study.

Holstein heifers were used as the model organism for the thermoregulation study. Initially, 8 heifers were fitted with indwelling jugular catheters and serial blood samples were collected at 3-minute intervals during exercise of increasing intensity. Blood lactate was then measured and it was determined that the lactate threshold of Holstein heifers occurs on average at a heart rate of 145bpm $\pm$ 7.7 and at an exercise speed of 6.44km/h $\pm$ 0.3. The results from the lactate threshold test were used to design two 8-week exercise trials with the objective to examine the effects of exercise on heat stress using high-intensity (short bouts of exercise above the lactate threshold), low-intensity (relatively long bouts of exercise below the lactate threshold), and sedentary control treatments. No differences were found in weekly skin temperatures or core body temperatures ($p > .05$). Expression of lactate dehydrogenase A in skeletal muscle cells decreased in the high-intensity treatment ($p = .03$). Expression of lactate dehydrogenase B in skeletal muscles decreased in the high-intensity ($p = .02$) and sedentary control ($p = .02$) treatments. Skin and core body temperatures from all treatments were correlated with THI ($p < 0.001$). There were no differences in body weight, body condition score, or

conception rate between treatments ($p > .05$). Although not significant ($p > .05$), after low-intensity exercise training, skin temperatures increased while core body temperatures decreased. Trends from this study warrant further investigation on the effects of low-intensity exercise on thermoregulation in Holstein cattle.

The second component of this project involved the incorporation of core physiological concepts from the thermoregulation project, in conjunction with those identified by the American Physiological Society, into an educational study where the utility of active learning for teaching difficult concepts within an undergraduate physiology course was investigated. Using a design-based research approach, two distinct pedagogical tools were developed: an active learning tool in the form of a case study and a conventional lecture. It was hypothesized that, compared to a conventional lecture, students receiving the active learning tool would perform significantly better on tests measuring their ability to comprehend, apply, and transfer the information to novel scenarios. Results from this project did not support the hypothesis but instead led to the question of is it the method or is it the student? Prior knowledge of students was evaluated using a physiology knowledge assessment. Students with low prior knowledge had greater learning gains from the use of a conventional lecture while students with high prior knowledge had greater learning gains from the use of a case study. Students with fewer college credit hours completed and those with lower ACT scores had larger learning gains after receiving a conventional lecture compared to the use of a case study. Students with more college credit hours completed and those with higher ACT scores had larger learning gains from the use of a case study compared to receiving the information from a conventional lecture. Furthermore, students who relied on memorization for learning new information benefitted more from a conventional lecture, while students who relied on elaboration for learning new information

benefitted more from a case study. Thus, the success of active learning likely depends on specific student characteristics. A one-size-fits-all approach to teaching and learning will not suffice; we must first consider the population of students and then select the proper instructional approach.

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Dedication

I dedicate this to my Old Grandmother (OG). I would not be the woman I am without you, thank you.

General Introduction

This project consisted of two studies including a scientific and an educational research project. The scientific research project consisted of investigating the effects of high and low-intensity exercise training on heat stress in dairy cattle. There were many ill-structured physiological concepts involved in this project, and in the interest of improving the presentation of such concepts to students in an undergraduate physiology course, an educational research study was also conducted. This portion of the project consisted of investigating the utility of active learning in the form of case studies in comparison to a conventional lecture. The overarching objective of this project was to integrate scientific research with educational research to improve graduate student research and teaching.

Chapter 1 - Exercise Training to Mitigate Heat Stress

Literature Review

Climate change has led to erratic precipitation patterns, increased levels of atmospheric carbon dioxide, and increased temperatures are causing concern for the U.S. livestock industry (Collins et al., 2013; Davidson et al., 2014; Key, Sneeringer, & Marquardt, 2014; Walsh et al., 2014; Walthall et al., 2013). Furthermore, climate change has contributed to reduced snowmelt runoff and increased surface water evaporation (Davidson et al., 2014; Famiglietti & Rodell, 2013; Scanlon et al., 2012; Walthall et al., 2013). Concurrently, higher temperatures for prolonged periods of time have increased water demand for many agricultural uses, including heat abatement in livestock. As such, the availability of ground-water, or water beneath the earth's surface stored in aquifers such as the High Plains Aquifer, has decreased (Famiglietti, 2014; Konikow & Kendy, 2005; McGuire, 2009; Taylor et al., 2012). As the reliance upon for ground-water increases for various agricultural needs, ground-water depletion and pumping costs for irrigated land will increase (Famiglietti, 2014; McGuire, 2009; Perlman, 2016; Taylor et al., 2012). Furthermore, inconsistent weather patterns, including increased temperatures, drought, and wind patterns, have contributed to unpredictable feed crop availability and increased pathogens and parasites among livestock (Grace, Bett, Lindahl, & Robinson, 2015; Key et al., 2014; Nardone, Ronchi, Lacetera, Ranieri, & Bernabucci, 2010; Walthall et al., 2013). These detrimental effects of climate change on livestock become increasingly important to consider as the human population continues to grow and food security for both livestock and humans continues to be a large concern (Godfray et al., 2010; Key et al., 2014; Parry & Rosenzweig, 1999; Zahid, Robinson, & Kelly, 2016).

US livestock and poultry industries generate approximately $100 billion annually, more than 50% of total US agricultural revenue (USDA, 2015). However, the implications of climate change are expected to significantly decrease livestock production, especially because of the impacts of heat stress (Key et al., 2014; St-Pierre, Cobanov, & Schnitkey, 2003; USDA, 2015). Heat stress has also contributed to an increase in mortality rates in cattle (Bishop-Williams, Berke, Pearl, Hand, & Kelton, 2015; Crescio, Forastiere, Maurella, Ingravalle, & Ru, 2010). In fact, during the summer of 2017 thousands of cattle died from heat stress in California, USA (Carr, 2017). Heat stress, which is characterized by an increased body temperature and accompanied by an increased heart rate, respiration rate, and altered blood chemistry, has had the largest effect on the dairy industry (Key et al., 2014; Polsky & von Keyserlingk, 2017; West, 2003; West, Mullinix, & Bernard, 2003). Heat stress has led to many adverse effects in the dairy industry including decreased reproduction, decreased milk production, decreased dry matter intake (DMI), decreased immunity, mastitis, electrolyte and acid-base imbalance, and even increased mortality risk (Baumgard & Rhoads, 2007; Collier, Dahl, & VanBaale, 2006; De Rensis & Scaramuzzi, 2003; Itoh, Obara, Rose, Fuse, & Hashimoto, 1985; Kadzere, Murphy, Silanikove, & Maltz, 2002; Key et al., 2014; Rhoads et al., 2009; West, 2003). These effects often contribute and interact with one another, becoming additive quickly, and exacerbating the effects of heat stress. For example, increased respiration rates have contributed to increased blood pH due to excessive release of carbon dioxide and therefore higher levels of bicarbonate. As a result of alkalosis, the kidneys will excrete excess bicarbonate, and could eventually result in acute acidosis. In addition to bicarbonate excretion, electrolytes are also lost in larger quantities in the filtrate under heat stressed conditions. Disruptions to the acid-base balance of an animal's body fluids can be very detrimental. For instance, as a result of heat stress, cattle

will release more bicarbonate via increased respiration rate and increased salavation as a cooling mechanism; a reduced amount saliva which contains bicarbonate reaching the rumen will result in rumen acidosis. In addition, blood flow increases to the periphery as a cooling mechanism which reduces blood flow to the gastrointestinal tract, this will typically result in an increase in volatile fatty-acid production within the rumen, further reducing the pH (Baumgard & Rhoads, 2007). Thus, acid-base imbalances could impact rumen health and function, and as such can have an effect on digestibility and nutrient availability (Baumgard & Rhoads, 2007; Kadzere et al., 2002; West, 2003). Furthermore, under heat stressed conditions dairy cattle have a decreased DMI which contributes to a negative energy imbalance, especially when energy requirements are high during lactation (Collier et al., 2006; West, 2003). As such, less energy is available for milk production (Fuquary, 1981; West, 2003). Due to the effects of heat stress in dairy cattle, the US Department of Agriculture has predicted a decrease in total annual milk production by 0.6-1.35% in the US dairy industry by the year 2030 (Key et al., 2014). This leads to the increasing concerns for food availability as the human population continues to rise.

Current Heat Abatement Strategies and Possible Alternatives

Some of the commonly used heat abatement strategies in the dairy industry include shade structures, water spraying systems, and fans (Collier et al., 2006; Ortiz et al., 2015). Shade structures can reduce the heat load from solar radiation, however they do not alleviate the effects of high ambient temperatures or humidity. Nevertheless, ventilation and water systems, such as misting or sprinkler systems, have been used to cool dairy cows via conductive, convective, and evaporative cooling. As a result, these strategies have effectively alleviated symptoms of heat stress (Chen, Schütz, & Tucker, 2015, 2016; Collier et al., 2006). Water systems designed to cool cattle are more efficient when sprayed for brief yet frequent periods of time (Collier et al.,

2006). During the hottest seasons of the year, between 23 to 256 L of water can be used to cool a single dairy cow per day (Chen et al., 2016). Although water systems and fans can reduce heat stress, these strategies can be energy intensive and may contribute to a decrease in natural resources, especially water depletion (Godfray et al., 2010; Hoekstra, 2012; von Keyserlingk et al., 2013). Due to an increase in water and energy use, the economic costs of current heat stress mitigation strategies in the US dairy industry may outweigh the profits (Key et al., 2014), yet they remain necessary to prevent serious health issues in cattle and depletion of the milk supply. To conserve resources while mitigating heat stress in dairy cattle, alternative heat abatement strategies should be considered.

Effects of Exercise Training on Body Temperature and Skeletal Muscle Cells

Exercise training has been shown to alleviate heat stress in humans, dogs, rats, and horses (Fox, Goldsmith, Kidd, & Lewis, 1963; Geor, Mccutcheont, & Lindingers, 1996; Houmard, Costill, Davis, Mitchell, Pascoe, & Robergs, 1991; Lorenzo, Halliwill, Sawka, & Minson, 2010; Nielsen, Hales, Strange, & Juel, 1993; Roberts, Wenger, Stolwijk, & Nadel, 1977). Heat acclimation in humans and horses via exercise training has been especially effective when exercised in high ambient temperatures. (Geor et al., 1996; Marlin et al., 1999; Roberts et al., 1977) Physiological characteristics of heat acclimation through exercise training include decreased core body temperature, increased sweat rate, improved blood flow to the dermis and skeletal muscle tissues, and increased plasma volume (Charkoudian, 2010; Fox et al., 1963; Geor, Mccutcheon, et al., 1996; González-Alonso & Teller, 1999; González-Alonso, Crandall, & Johnson, 2008; Simmons, Wong, Holowatz, & Kenney, 2011). During exercise, a large increase blood flow to the skeletal muscle tissues results in an increase in shear stress, otherwise known as the mechanical stress placed on blood vessels surrounding the tissue. Over time, as a result of

shear stress within the vasculature of skeletal muscle tissue, exercise training can result in vascular development or remodeling. There are two mechanisms responsible for vascular development: vasculogenesis and angiogenesis. Vasculogenesis is de novo synthesis of vasculature which involves differentiation of epithelial cells to form primitive vascular networks. Typically, vasculogenesis occurs during embryonic development or under pathological conditions such as wound healing or tumor formation, similar to the stress of exercise (Patan, 2000; Vailhé, Vittet, & Feige, 2001). Angiogenesis is defined as remodeling of the vasculature or an increase in existing vasculature; this process involves migration and proliferation of endothelial cells lining the vasculature towards an angiogenic stimulus (Auerbach, Lewis, Shinners, Kubai, & Akhtar, 2003; Patan, 2000; Prior, Yang, & Terjung, 2004; Semenza, 2007; Vailhé et al., 2001). As a result of angiogenesis or vasculogenesis from exercise training, more nutrients can be delivered to skeletal muscle cells and more waste can be removed. This has also been termed as an increase in oxidative capacity, or an increase in capillary volume.

Exercise training in humans, horses, and dogs has been shown to enhance oxidative capacity through capillary expansion and increased mitochondrial volume and size within skeletal muscle cells (Egan & Zierath, 2013; Eivers et al., 2010; Gollnick & King, 1969; Holloszy, 1967; Holloszy & Coyle, 1984; Ingjer, 1979; Williams, Salmons, Newsholme, Kaufman, & Mellor, 1986; Wu et al., 2002; Yan, Lira, & Greene, 2012; Young, Mosher, Erve, & Spector, 1959). Oxidative capacity of skeletal muscle cells varies among muscle fiber type. For example, type I skeletal muscle fibers, or slow twitch fibers typically have higher capillary and mitochondrial densities than type II, or fast twitch muscle fibers (Laughlin & Roseguini, 2008; Staron, Hikida, Hagerman, Dudley, & Murray, 1984; Westerblad, Bruton, & Katz, 2010). However, oxidative capacity of particular muscle fiber types can be altered through different

forms of exercise training. For instance, high-intensity exercise training has been shown to recruit primarily type II muscle fibers while low-intensity exercise training has recruited primarily type I muscle fibers, and both forms of exercise increased oxidative capacity of the predominate muscle fibers that were recruited via vasculogenesis and angiogenesis (Gibala, Little, Macdonald, & Hawley, 2012; Gute, Fraga, Laughlin, & Amann, 1996; Laughlin & Roseguini, 2008; Westerblad et al., 2010).

Exercise Fitness Measures and Training Protocols

While the effects of exercise training on thermoregulation have been investigated in many mammals, few studies have investigated these effects on large ruminants such as Holstein cattle (Bos Taurus). However, there has been some exploration of exercise training dairy cattle by Davidson & Beede (2003) who found that exercise training reduced heart rates and plasma lactate concentrations in pregnant dairy cows. However, others have found that combining low- and high-intensity exercise training did not have an impact on heart rates, but did increase milk protein content and lower skin temperatures after a mixed low- and high-intensity exercise training regimen in pregnant dairy heifers (Johnson, Steichen, & Rozell, 2016). While there is evidence that exercise can improve fitness and heat tolerance in dairy heifers, the physiological outcomes for enhanced thermotolerance, or heat tolerance, and its effects on dairy production are not well understood.

Poor understanding of how exercise impacts thermotolerance in dairy heifers could be partly due to inconsistent exercise training regimens or imprecise fitness measures. Physical fitness in heifers was formerly assessed by increasing the speed or slope of a walking surface over time until volitional fatigue or the animal's heart rate exceeded 180 bpm (Davidson & Beede, 2008; Johnson et al., 2016). While this can allow for measurement of changes in physical

fitness, these indicators may not assess fitness in all heifers or cows accurately as resting heart rates may vary. In addition, the intensity and duration of exercise training were previously based on the comfort level of the heifer or cow using heart rates, respiratory rates, and visual indicators of fatigue (Davidson & Beede, 2008; Johnson et al., 2016). However, in humans, horses, dogs, and rats, carefully designed fitness measures and exercise training protocols were used to accurately measure changes in physical fitness and improved thermoregulation (Allen, van Erck-Westergren, & Franklin, 2016; Guerreiro, Pereira, Martins, Wally, & Goncalves, 2009; Laughlin, Diana, & Tipton, 1978). Thus, to gain a more accurate understanding of how exercise training impacts thermoregulation in dairy cattle, more precise fitness measures and exercise training regimens should be considered.

Lactate Threshold and Skeletal Muscle Lactate in Holstein Heifers

A commonly used physical fitness measurement is the lactate threshold exercise test. Lactate threshold (LT) during exercise is the point at which blood lactate levels begin to exponentially increase as the amount of lactate being produced and released from exercising skeletal muscle cells exceeds the amount that can be utilized by cells as an energy source. The LT test is commonly used to understand levels of fitness in humans (Esfarjani & Laursen, 2007; Heck et al., 1985; Ivy, Withers, Van Handel, Elger, & Costill, 1980; Poole & Gaesser, 1985) and horses (Eaton & Rose, 2006; Gondim, Zoppi, Pereira-da-Silva, & de Macedo, 2007; Kronfeld, Ferrante, Taylor, & Custalow, 1995). However, the LT has not been described in dairy heifers. While there are other fitness measures that are commonly used in humans, such as the maximal oxygen consumption (VO2 max) test, this is a very difficult test to conduct using dairy heifers.

While lactate has been thought to be a waste product that is only produced under anaerobic conditions, lactate is actually an energy source produced constantly from pyruvate as

long as the appropriate enzyme isoform of lactate dehydrogenase is available (Brooks, Dubouchaud, Brown, Sicurello, & Butz, 1999; Brooks, Brown, Butz, Sicurello, & Dubouchaud, 1999; Cruz et al., 2012; Donovan & Brooks, 1982; Dubouchaud, Butterfield, Wolfel, Bergman, & Brooks, 2000; Gladden, 2004; Rogatzki, Ferguson, Goodwin, & Gladden, 2015). Lactate dehydrogenase (LDH), a tetramer enzyme with five isoforms, is responsible for the conversion of pyruvate to lactate or lactate to pyruvate, depending on the isoform. The tetramer is formed by two different protein subunits, LDH-M (muscle-type), coded for by LDHa mRNA, and LDH-H (heart-type), coded for by LDHb mRNA (Markert, Shaklee, & Whitt, 1975). In mammals, LDH-M and LDH-H protein subunits form five different LDH complexes LDH1(H4), LDH2 (H3M1), LDH3 (H2M2), LDH4 (H1M3), and LDH5 (M4) (Apple & Rogers, 1986; Brooks et al., 1999; Cruz et al., 2012; Dawson et al., 1964; Dubouchaud et al., 2000; Friedmann et al., 2004; Gay, McComb, & Bowers, 1968; Leberer & Pette, 1984; Liang et al., 2016; McCullagh et al., 1997; Summermatter, Santos, Perez-Schindler, & Handschin, 2013). The conversion of lactate to pyruvate has been found to be catalyzed by LDH1 while the conversion of pyruvate to lactate has been found to be catalyzed by LDH5; LDH isoforms 2, 3, and 4 have been considered intermediates (Dawson et al., 1964; Krieg, Rosenblum, & Henry, 1967; Liang et al., 2016; Porporato et al., 2011). The "muscle-type" and "heart-type" descriptions of these enzymes do not represent their location accurately because levels of specific LDH isoforms, LDH-M and LDH-H, within certain tissues depends on the oxidative capacity of the tissue (Cruz et al., 2012; Dubouchaud et al., 2000; Fritz & Jacobson, 2017; Gay et al., 1968; Krieg et al., 1967; Liang et al., 2016; Markert et al., 1975; McCullagh et al., 1997). To examine LDH isoform expression, LDHa and LDHb mRNA subunits have been investigated in horses, sheep, dogs, cats, and rabbits; however, only a few studies have investigated these mRNA subunits in Bos Taurus

(BosLDHa & BosLDHb) skeletal muscle cells (Brown et al., 2015; Echigoya, Sato, Itou, Endo, & Sakai, 2009; Washizu et al., 2002). Because LDHa and LDHb mRNA are the precursor for the LDH protein subunits, examining the quantity of these within skeletal muscle cells could predict which LDH isoforms are being upregulated in response to a stressor. The LDH isoform available within a skeletal muscle cell becomes increasingly important during exercise as it determines whether lactate will accumulate and diffuse out of the skeletal muscle cell or be utilized as an energy source within the cell (Dawson et al., 1964; Dubouchaud et al., 2000; Hashimoto, 2006; Krieg et al., 1967; McCullagh et al., 1997; McGinley & Bishop, 2016).

During exercise, lactate accumulates within skeletal muscle cells in response to ATP production. This byproduct can have many fates such that it can diffuse out of the cell or into the mitochondria within the cell to be utilized for energy production (Donovan & Brooks, 1982; Eaton & Rose, 2006; Hashimoto, 2006; Ivy et al., 1980; Pilegaard et al., 1999). Lactate diffuses across membranes via lactate transporters, monocarboxylate transporter-1 and -4 (MCT1, MCT4). These transporters have been localized within the inner mitochondrial membrane and sarcolemma of skeletal muscle cells, respectively (Brooks et al., 1999; Dubouchaud et al., 2000; Hashimoto, 2006; McCullagh et al., 1997). These skeletal muscle lactate transporters are responsible for the diffusion of lactate, and free hydrogen ions, within the cell and mitochondria (Brooks et al., 1999; Dubouchaud et al., 2000; Passarella et al., 2008). Through exercise training, these transporters within skeletal muscle cells have been shown to increase within the sarcolemma and mitochondrial membrane. There are other features that lead to enhanced oxidative capacity such as capillary density, mitochondrial density and volume, and the amount of LDH produced within the cell (Brooks, Dubouchaud, Brown, Sicurello, & Butz, 1999; Brooks, Brown, Butz, Sicurello, & Dubouchaud, 1999; Dubouchaud, Butterfield, Wolfel,

Bergman, & Brooks, 2000; Hashimoto, 2006; Hood, 2001). In addition, an increase in MCT transporters on the sarcolemma can increase lactate export from skeletal muscle cells to other tissues. An increase in MCT transporters on the mitochondrial membrane can improve the ability of skeletal muscle cells to utilize lactate for ATP production. As a result of an increase in oxidative capacity of skeletal muscle tissue, the LT will increase as lactate becomes more readily used by skeletal muscle cells for ATP production and less lactate will accumulate in the blood (Acevedo & Goldfarb, 1989; Eaton & Rose, 2006; Gladden, 2004; Marcinik et al., 1991; Poole & Gaesser, 1985). Thus, the LT test has been a reliable indicator of overall fitness and has been used to determine intensities for exercise training in humans and horses (Eaton & Rose, 2006; Ferraz et al., 2008; Gladden, 2004; Kim, Tanaka, & Maeda, 1991; McGinley & Bishop, 2016; Ocel et al., 2003; Périard, Travers, Racinais, & Sawka, 2016). As shown in other species, exercise intensities for dairy cows should be based on their lactate threshold as a precise measure of fitness gains to improve thermotolerance.

Materials and Methods

To investigate the impacts of precise exercise training protocols using the lactate threshold as a baseline measure on thermoregulation in dairy cattle, the objectives of this study included: 1. Establish the lactate threshold in dairy heifers 2. Examine the effects of exercise intensity on heat tolerance.

Animals, Housing, and Diets

Experimental procedures were approved by the Kansas State Institutional Animal Care and Use Committee (IACUC, protocol #3710). A total of 57 Holstein heifers, nine to ten months in age, were obtained from the Dairy Teaching and Research center at Kansas State University. Eight heifers were used to determine LT and LTHR. To investigate potential changes in physiological parameters from exercise training based around the LTHR of Holsteins, 49 heifers were used; 6 of these heifers were removed from the study for health reasons unrelated to exercise, including 2 heifers from trial 1 and 4 heifers from trial 2.

Heifers were housed in a free stall barn and offered a mixture of wet corn gluten feed (WCGF) 1-4 times daily ad-libitum. The WCGF diet included: sweet bran (62% DM), corn silage (34% DM), grass hay (88% DM) and grain mix (90% DM). Heifers in all treatment groups were fed at the same time of day. The diet used in this study was fundamentally the same as the experimental diet used by (Mullins, Grigsby, Anderson, Titgemeyer, & Bradford, 2010).

Experimental Design

This study consisted of two main components: 1. determination of the LT and LTHR in dairy heifers 2. exercise training slightly above or below lactate threshold heart rate as an

intervention for heat stress (Figure 1.1). The lactate threshold (LT) of eight Holstein heifers was determined by assessing lactate in serial blood samples taken from the jugular vein during an incremental exercise test. Results from the LT test were used to design exercise training protocols for two 8-week exercise trials with three treatment groups (high-intensity, low-intensity, sedentary control). Trial 1 originally consisted of 24 heifers; 8 heifers within each treatment. Trial 2 began with 25 heifers; high-intensity (n=9), low-intensity (n=8), and sedentary control (n=8). A total of 6 heifers were removed from the study and thus n was reduced (trial 1, n=22 and trial 2, n=21). Muscle biopsies were taken from the right semitendinosus before and after each exercise training trial (pre and post-exercise muscle biopsy). Heifers from each trial were acclimated to the exerciser 3 days before exercise training commenced. Trial 1 was conducted between 16 May and 18 July of 2016 and trial 2 was conducted between 25 July and 26 September of 2016.

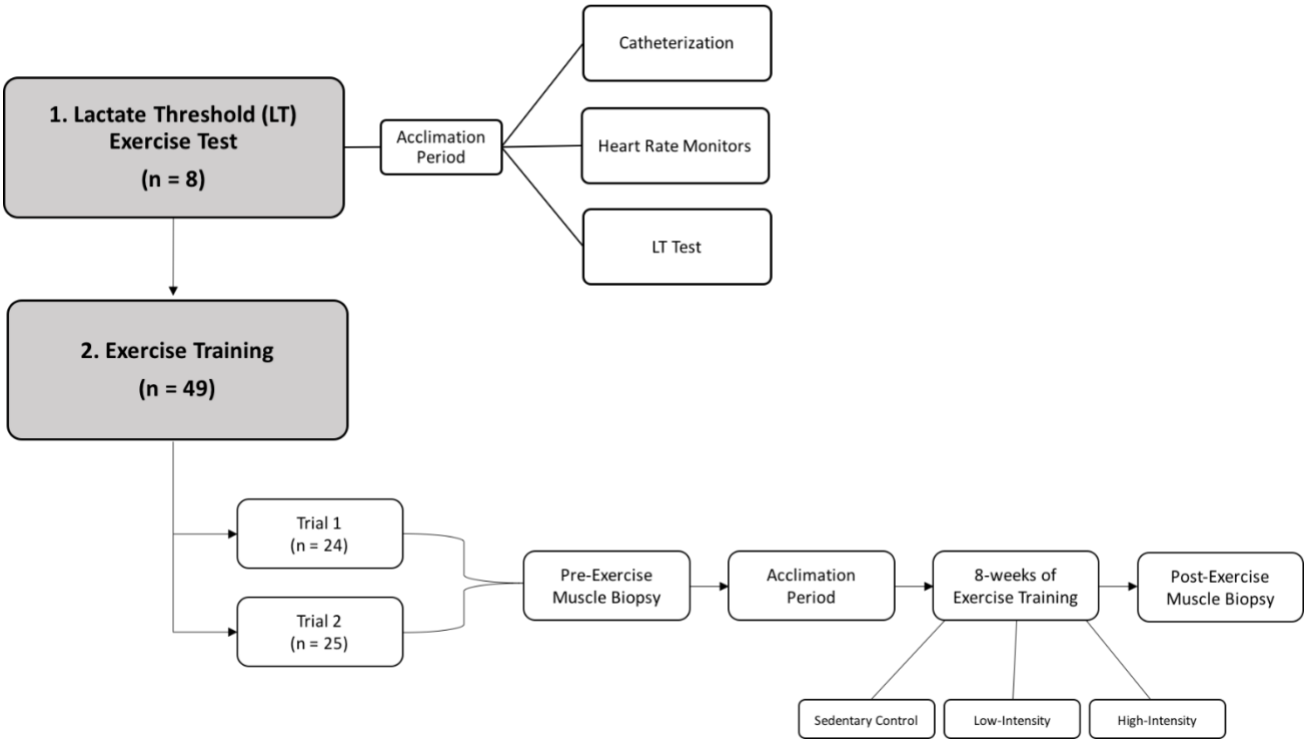

Figure 1.1. Experimental design.

A lactate threshold test was first conducted and the results were used to design two 8-week exercise trials to compare low- and high-intensity exercise programs consisting of activities below or above the lactate threshold heart rate, respectively. Muscle biopsies were taken from each heifer before and after exercise training and weekly core body and skin temperatures were recorded throughout the exercise period.

Lactate Threshold

Acclimation period

Two days prior to the lactate threshold (LT) test, 8 Holstein heifers, 9-10 months in age, were acclimated to an eight-panel motorized exerciser located outdoors (Priefert, 8-horse exerciser, Mount Pleasant, TX). To accomplish this, heifers were transported on a trailer from their group holding pen to the exerciser (distance approximately 0.4km) and individually walked in an exercise bay. Heifers walked for approximately 10 minutes at 3.2km/h then returned to their group holding pen.

Catheterization

Heifers were restrained in a head catch to allow access to the jugular vein. The area was prepared for surgery by removal of hair and then application of Betadine Surgical Scrub (Doctors Fosters and Smith©, Rhinelander, WI) followed by 70% ethanol. Prior to catheterization, 2% lidocaine HCl was administered subcutaneously, superficial to the left jugular vein. An indwelling catheter (Extended Use MILACATH-14Ga x 13cm catalog #1411, Medical Instruments for Animals (MILA) International Inc., Erlanger, KY) was inserted into the jugular vein. Catheters were secured using 8690 G prolene suture material (Ethicon©, Somerville, NJ), Co-Flex®, and surgical tape. Heifers were monitored in the treatment pens for approximately four hours until the lactate threshold test began.

Application of Heart Rate Monitors

Heifers were fitted with heart rate (HR) monitors (Polar, RS800CX, Kemple, Finland) to record specific HR responses during the LT test. The heart rate monitors were attached on a

transverse plane encircling the thoracic vertebrae and sternum, caudal to the acromial end of the scapula. Embrocation (Up & Up, Target©, Minneapolis, MN) and electrode gel (Old Spice High Performance Shower Gel, Proctor & Gamble ©, Cincinnati, OH) were applied under the electrodes to increase conductivity and sensitivity for ECG monitoring.

Blood Sampling and Analyses

After the HR monitors were attached, each heifer was loaded into an individual bay of the exerciser. Jugular catheters were flushed with heparinized saline (100U/mL) and blood was collected from the catheters at each sampling time using 10mL EDTA (k3) heparinized conical tubes (Monoject, Mansfield, Ma). All blood samples were immediately placed on ice and transported to the lab for centrifugation (Beckman Coulter, J6-B, Brea, CA; 20 min, 1,200 x g, 5°C). Plasma was stored at -20°C for 12 hours and then analyzed for glucose and L-lactate using a YSI 2300 Stat Plus Glucose and Lactate Analyzer (YSI Inc., Yellow Springs, OH) with a glucose/L-lactate reagent kit (catalog #2323, Yellow Springs, OH).

Lactate Threshold Test Design

The exercise protocol for determining lactate threshold consisted of incremental steps. There were three minutes of exercise that progressively increased in intensity, followed by a one minute rest period. Blood samples were collected and heart rates were recorded at minute 0 and during the one minute rest period following each incremental exercise stage (Figure 1.2). After the final bout of exercise and rest period, heifers were individually moved into a chute for removal of sutures, catheters, and HR monitors. Pressure was applied to the catheterization site to prevent any bleeding, followed by application of Betadine Surgical Scrub (Doctors Fosters and Smith©, Rhinelander, WI). Catheterization sites were directly observed for signs of infection twice a day for the next 5 days.

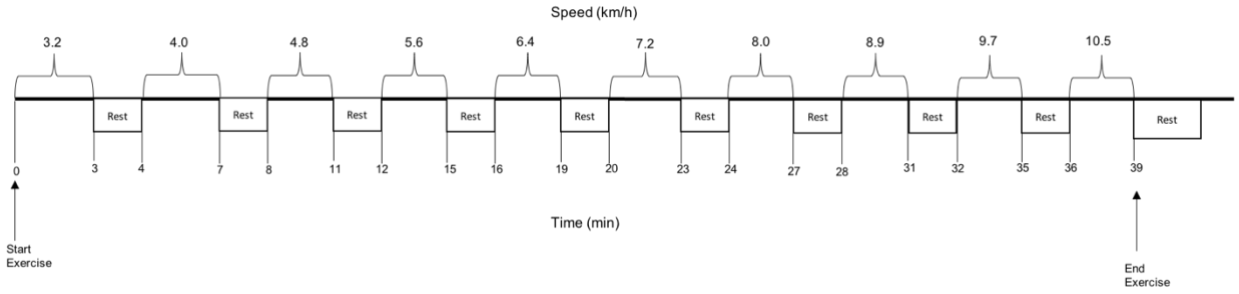

Figure 1.2. **Lactate threshold exercise test.**

Heifers were exercised at each incremental speed for 3 minutes with a 1 minute standing rest period for HR recording and blood sampling. The first blood sample and heart rate measurement was taken at time 0. The first exercise interval began at a speed of 3.2 km/h, subsequent speeds were 4.0 km/h, 4.8 km/h, 5.6 km/h, 6.4 km/h, 7.2 km/h, 8.0 km/h, 8.9 km/h, 9.7 km/h, and the final speed was 10.5 km/h. The speeds were increased between 0.7-0.9 km/h for each interval. There was a total of 10 exercising intervals.

Determination of Lactate Threshold

Results from the LT test were used to design subsequent exercise regimens for low-intensity and high-intensity exercise. Displayed in Figure 1.3 is a representative graph of blood lactate accumulation from one heifer in the LT test. The exerciser speed at each incremental exercise step was plotted against blood lactate (mmol/L) and HR (bpm). Lactate threshold was determined by the observed exponential increase in mean blood lactate levels (Figure 1.4). Blood lactate levels began to exponentially increase for heifers on average at an exercising speed of 6.44km/h with an average heart rate of 145bpm. The HR associated with LT, or lactate threshold heart rate (LTHR), was used to design each exercise training treatment (Figure 1.4)

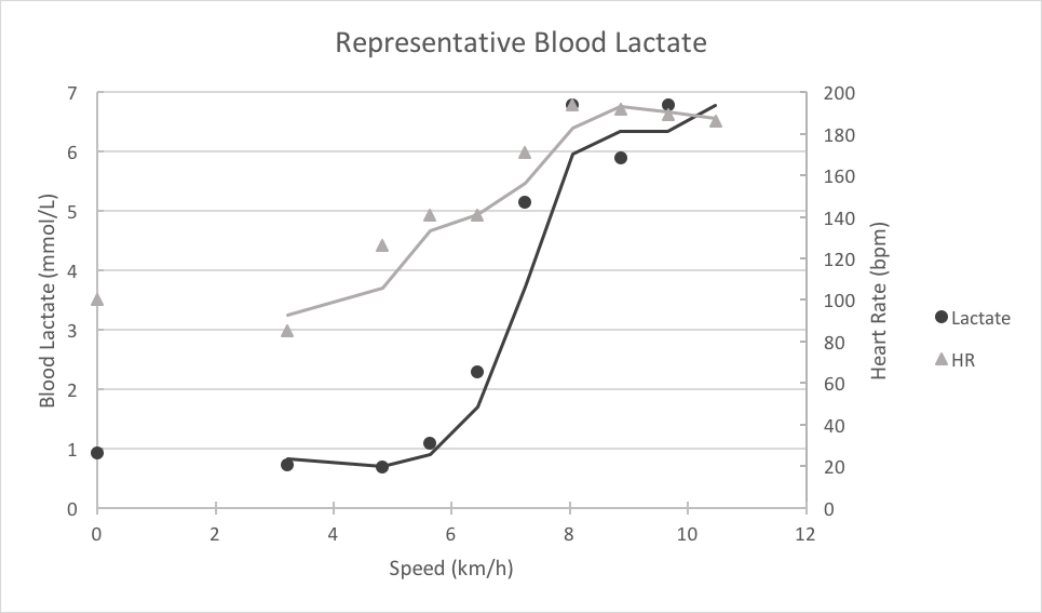

Figure 1.3. **Representative blood lactate and heart rate response during incrementally increasing exercise intensities.**

These results were gathered from one heifer and include blood lactate (mmol/L) and heart rate (HR, bpm) measures at a given exercise speed (km/h) during the LT test. Due to issues experienced during blood collection, one data point is missing on this graph at the 4km/h speed. The initial blood lactate and HR measurements were taken at time point 0, and thus the speed is 0 km/h.

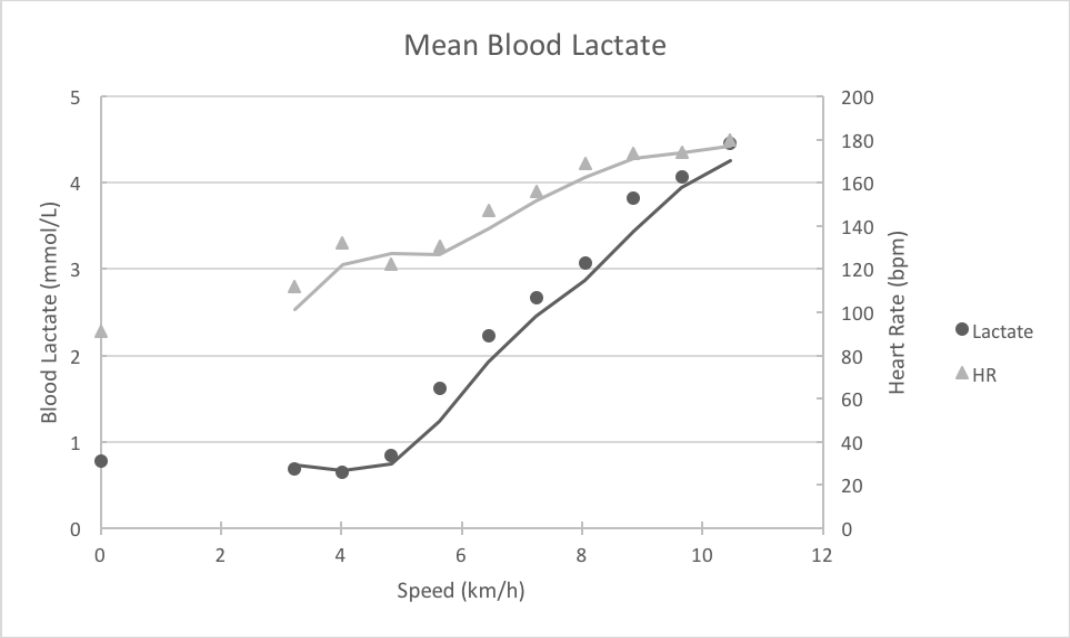

Figure 1.4. **Mean blood lactate.**

Blood lactate (mmol/L) and heart rates (HR, bpm) were averaged among all heifers used for the LT test at each speed (km/h). Results were used to identify the lactate threshold heart rate (LTHR) where blood lactate levels exceeded 2mmol/L; this occurred at a speed of 6.44km/h when mean blood lactate was 2.2mmol/L and HR was 145bpm. The initial blood lactate and HR measurements were taken at time point 0, and thus the speed is 0 km/h.

Exercise as an Intervention for Heat Stress

An exercise trial was conducted between 16 May and 18 July of 2016; this trial was replicated between 25 July and 27 September of 2016. For each trial, heifers (8-10 months of age) were stratified according to age in days (youngest to oldest) and the ratio of black to white coat color (lowest to highest). Heifers were then randomly assigned to each exercise treatment (sedentary control, low-intensity exercise, and high-intensity), starting with the first 3 youngest and lowest color ratio through the oldest and highest color ratio. Color ratios were determined by averaging three estimates of black: white coat color determined by individual researchers. All exercise activities were initiated at 0530 and completed on most days by 0800.

Exercise Treatments

Heifers were transported in a stock trailer to the exerciser (Priefert, 8-horse exerciser, Mount Pleasant, TX) and exercised 3 days per week for 8 weeks in each trial. Alternate treatment times were used for high-and low-intensity groups such that if one group went first on one day, the other group would be the first to go on the next day of treatment. The exerciser could rotate in a clockwise or counterclockwise direction, and the direction of the exerciser was alternated after each day of treatment. Heifers in the sedentary group were transported to the exerciser and directly back to the group holding pen.

The first week of exercise training was used to acclimate all heifers to exercise; therefore, heifers in the low- and high-intensity treatments participated in the same duration and intensity of exercise (Table 1.1). Heifers in the high-intensity treatment were subjected to exercise speeds that aimed to be above the LTHR for short periods of time. As shown in Table 1.1, high-intensity interval speeds ranged between 5.47km/h to 7.40km/h with an active rest period

between each interval (speed at 3.21km/h). The number of sprint interval sets and exercise speed increased gradually. Heifers in the low-intensity group exercised for longer periods of time at a constant speed (4.5km/h) or 70% of the LTHR; duration of exercise ranged from 20 to 60 minutes.

Table 1.1. **High-intensity vs. low-intensity exercise training.**

High-and low-intensity training protocols for both 8-week exercise trials. Time of exercise, speed of the exerciser, and % of LTHR are listed. The 2X and 3X indicate the number of sets for the high-intensity treatment.

Week	**High-Intensity**	**Low-Intensity**
W1	15 min walk (4.5 km/h), 70% LTHR	15 min walk (4.5 km/h), 70% LTHR
W2	10 min (5.47 km/h), 85% LTHR	20 min (4.5 km/h), 70% LTHR
W3 & W4	2X: 1 min (6.11 km/h) 2 min (3.21 km/h) 1 min (6.11 km/h) 2 min (3.21 km/h) 1 min (6.11 km/h) 5 min (3.21 km/h), 95% LTHR	30 min (4.5 km/h), 70% LTHR
W5 & W6	3X: 1 min (6.76 km/h) 2 min (3.21 km/h) 1 min (6.76 km/h) 2 min (3.21 km/h) 1 min (6.76 km/h) 5 min (3.21 km/h), 105% LTHR	45 min (4.5 km/h), 70% LTHR
W7 & W8	3X: 1 min (7.40 km/h) 2 min (3.21 km/h) 1 min (7.40 km/h) 2 min (3.21 km/h) 1 min (7.40 km/h) 5 min (3.21 km/h), 115% LTHR	60 min (4.5 km/h), 70% LTHR

Data Collection

Muscle biopsies from the right semitendinosus muscle were performed one week before each exercise trial (week 0) and the week after the end of the exercise trial (week 8). The pre-exercise biopsy, obtained from both trials, was taken approximately 1 cm lateral to the midline of the muscle and the post-exercise biopsy was taken from the same muscle 1 cm medial to the midline. The biopsy site was shaved and surgically prepared with an iodine solution (Betadine Surgical Scrub, Doctors Fosters and Smith©, Rhinelander, WI) followed by 70% ethanol application. The biopsy procedure began with a sterile piercing needle to penetrate the dermis to prepare the area for 3-5 samples per biopsy using a 10ga x 5cm long Quick-Core Biopsy Needle (Cook Medical, Chicago, IL). Samples were frozen in LN2 and stored at -80°C until qPCR analysis.

Heifers from all treatment groups were brought into a central handling facility each week on a non-exercise day for data collection throughout each of the 8-week treatment periods among both trials. Body weights were collected to examine growth trends between treatments by using a scale specially designed for livestock (GSE Inc., LBS Scales 350, Livonia, MI) . Additionally, body weights were used to calculate surface area using the equation from Brody, Comfort, and Matthews (1928): Surface Area = $(0.14*BW^{0.57})$. Body condition scores (BCS) were determined by a trained technician at the time of body weight determination during weeks 1-3, then weeks 5 and 8 of the exercise period for trial 2 only. Core body (Tb) and skin temperatures were collected weekly throughout both trials. Rectal temperatures (Tb) were assessed using a digital thermometer (Rectal Thermometer M700, GLA Agricultural Electronics, San Luis Obispo, CA) and skin temperatures were measured using an infrared thermometer (Raytek, RAYMT4U, Mini Temp Portable IR Gun, Quebec City, Canada). Skin temperatures were measured in the following

areas: center of the masseter muscle (cheek), cranial center of the right pinna (ear), dorsal ridge between the scapulae (withers, in the center of a line between the ilium and ischium (thurl), and at the caudal midline between the two hindquarters (udder). Skin temperatures were individually examined (Appendix A1-5) and averaged across all measurement sites and used as a metric termed "mean skin temperature" (MST). Mean skin temperatures were divided by core body temperatures to determine temperature ratios (MST:Tb) between the core and skin. Conception rates were collected from PCDART (Dairy Records Management Systems, PCDart, Raleigh, NC) and calculated by dividing the service in which conception occurred (1) by the total number of services.

RNA isolation and quantitative Reverse Transcription PCR

RNA isolation, reverse transcription, and quantitative PCR (qPCR) were performed on 150 mg of skeletal muscle tissue from the semitendinosus muscle, as outlined by Gonzalez et al. (2013). Nucleic acids were extracted by a Ribozol (Life Technologies, Grand Rapids, NY; Invitrogen)/chloroform, centrifugation process. The nucleic acid aqueous solution was filtered through a silica spin column and purified using an Invitrogen Purelink™ RNA Mini Kit (Life Technologies, Carlsbad, CA). Total RNA quality and concentration were evaluated using a NanoDrop 1000 spectrophotometer (Thermo Scientific, Waltham, MA). All RNA extracts yielded 260:230 ratios greater than 1.8 and were stored at -80°C until qPCR analysis.

Total RNA (50 ng) were combined with DNase to remove any genomic DNA and subsequently reverse transcribed using a High-Capacity cDNA Reverse Transcription Kit (Life Technologies, Carlsbad, CA). The cDNA representing the equivalent of 1 ng of total RNA were combined with gene specific primers (Table 1.2), PerfeCTa SYBR Green FastMix (Quanta

Biosciences, Gaithersburg, MD) and amplified using an Eppendorff Mastercycler Realplex^2 S PCR System (Eppendorff North America, Hauppauge, NY). Thermocycler conditions were: denaturation at 95°C for 10 min followed by 50 cycles of 15 s at 95°C, annealing at 60°C for 15 s, and an extension step at 68°C for 20 s. At the end of each program a dissociation step was conducted at 95°C for 15 s, 60°C for 30 s, and 95°C for 15 s. Relative gene expression was calculated as $2^{-\Delta\Delta Ct}$, with C_t denoting threshold cycle, expression being normalized to *18S rRNA* expression (ΔC_t), and calibrated to the average ΔC_t value of a pooled cDNA sample ($\Delta\Delta C_t$). This pooled sample represented all the samples in the current study that were amplified for each gene on each plate (Burnett et al., 2016).

Table 1.2. Genes, sequences, annealing temperatures, amplicon length, and efficiency of primers.

Gene of Interest	Forward Primer (5' to 3')	Reverse Primer (5' to 3')	T_m, ºC	Amplicon Length
BosLDHa[1]	TATTGGAAGTGGTTGCAATCTG	GCACACTAGAGTCACCATGCTC	58.9(F)[2], 64.5(R)[2]	22
BosLDHb[1]	ACGAGCTTGCTCTTGTGGAT	TGGAATTGGCAGTGACAGAG	60.4	20
18s[1]	GTAACCCGTTGAACCCCATT	CCATCCAATCGGTAGTAGCG	60.4(F)[2], 62.4(R)[2]	20

[1]Abbreviations: *BosLDHa* = Bos Taurus lactate dehydrogenase-a, *BosLDHb* = Bos Taurus lactate dehydrogenase-b, 18s

[2](F) indicates forward primer temperature, (R) indicates reverse primer temperature

Statistical Analyses

Expression of LDHa and LDHb (data output in appendix, Appendix Table A.1 & Appendix Table A.2) was analyzed as a generalized randomized complete block design using the MIXED procedure of SAS (SAS Institute Inc., Cary, NC). Core body temperature (Tb), mean skin temperature (MST), MST:Tb, and individual skin temperatures (cheek, ear, withers, thurl, udder) were analyzed using IBM's SPSS, version 24. To assess differences between treatments in Tb, MST, MST:Tb, and individual skin temperatures at each week of exercise training, data were analyzed using one-way ANOVAs. A Levene's test for homogeneity was performed and differences with a reported p-value < 0.05 in a one-way ANOVA were further analyzed using a Tukey post hoc test to determine specific treatment mean differences. If variances were not homogenous, a one-way Welch ANOVA was used to correct for variance. Differences with a p-value < 0.05 from the Welch ANOVA were followed by the Games-Howell post hoc analysis to determine which groups were significantly different.

Pearson correlations were used to analyze relationships between temperature humidity index (THI) and Tb, MST, MST:Tb, and individual skin temperatures for each treatment. Linear regressions were performed on THI and Tb, MST, and MST:Tb data for weeks 2-8. Week 1 was excluded from data collection because exercise protocols were identical for the low and high-intensity exercise trained groups. Using the constant coefficient (B_o) and the THI (B_{THI}) coefficient from the regression equations for weeks 2-8, subsequent regressions were calculated to predict whether there was a trend between treatment and Tb, MST, and MST:Tb based on a range of THI conditions between 65-95. The following equation was used to calculate each of these predictions: Temperatures (Tb, MST, or MST:Tb) = $B_o + (B_{THI} \times THI)$. Multiple linear

regressions were performed on these subsequent predictions to identify any treatment effects on predicted Tb, MST, and MST:Tb.

One-way ANOVAs were used to analyze body weight and body condition scores at each week between treatments. Conception rates were also analyzed using one-way ANOVAs between treatments.

Results

Expression of LDHa and LDHb mRNA

As shown in **Error! Reference source not found.**, there was a significant decrease of mRNA expression of LDHa from week 0 to week 8 for heifers in the high-intensity treatment, t (38.7) = 2.33, p = 0.03. There was a significant decrease in mRNA expression of LDHb between week 0 and week 8 for the control, t (42.5) = 2.35, p = 0.02, and the high-intensity treatments, t (42.4) = 2.40, p = 0.02. Data were combined for trial 1 and trial 2 for these analyses to assess whether there was an overall treatment effect on LDH subunit mRNA expression.

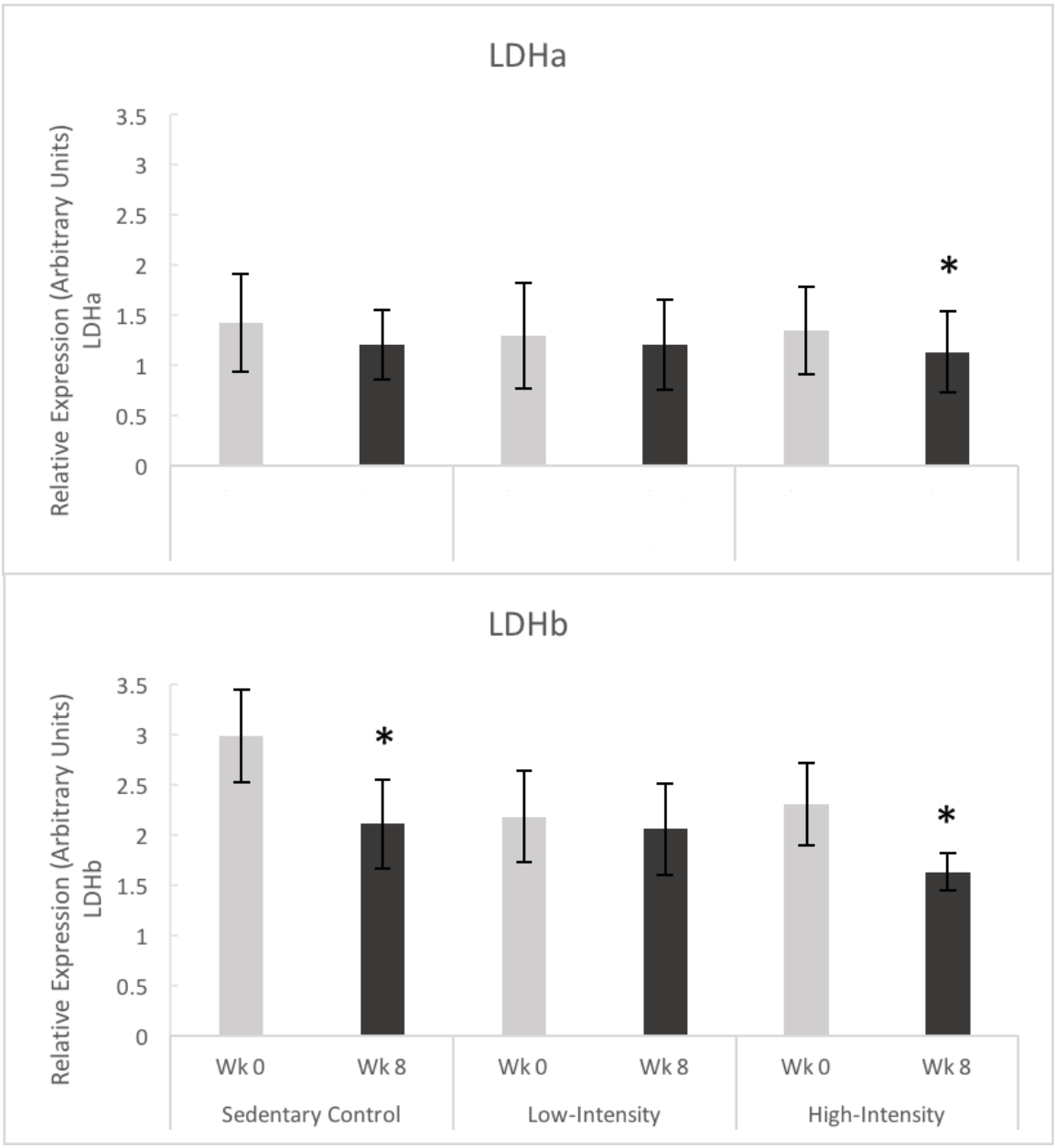

Figure 1.5. **LDH subunit mRNA expression.**

Expression of LDHa and LDHb mRNA from the semitendinosus muscle were compared between biopsy samples taken prior to and following an 8-week exercise regimen. Data were combined for trials 1 and 2 and are presented as means plus/minus SEM. The common x-axis displays treatment groups and week of biopsy sample collection. Treatments: Sedentary Control (n=14), Low-Intensity exercise (n=14), and High-Intensity exercise (n=15). Expression of *LDHa*

was reduced ($p < .05$) at Wk 8 compared to Wk 0 for the high-intensity treatment. Expression of *LDHb* subunit mRNA was reduced ($p < 0.05$) at Wk 8 compared to Wk 0 for sedentary control and high-intensity treatments.

Temperature Data

Core Body Temperature

There was a difference in core body temperature (Tb) from week 1 of trial 2 between treatments, $F(2,18) = 6.01$, $p = .01$. As shown in Figure 1.6, Tb means were significantly higher in the low-intensity treatment than the sedentary control (.55, 95% CI (.11-.98), $p = .013$) and the high-intensity treatment (.45, 95% CI (.03-.86), $p = .033$). There were no other significant differences found in Tb between treatments at each weekly measure.

Individual Temperatures

Using combined data from trial 1 and 2, there was a difference in ear temperatures between treatments, $F(2,40) = 3.60$, $p = .036$. The sedentary control treatment had significantly higher ear temperatures than the low-intensity treatment (Appendix Figure A.2) on week 2 (1.87, 95% CI (.13,3.6), $p = .032$). There were no other significant differences between treatments among the five individual skin temperatures (cheek, ear, withers, thurl, udder) across the 8-week trials.

Mean Skin Temperature

There was a significant difference in mean skin temperature (MST) on week 6 of trial 1 between treatments, $F(2,19) = 4.297$, $p = 0.029$. As shown in Figure 1.7, MSTs were significantly higher in the high-intensity treatment than the sedentary control treatment (1.89, 95% CI (.07-3.7), $p = .041$).

Mean Skin Temperature and Core Body Temperature Ratios

Mean skin temperature and core body temperature ratios (MST:Tb) were averaged weekly within treatments for trial 1 and 2. As shown in Figure 1.8, there were no differences found from MST:Tb between treatments at each weekly measure.

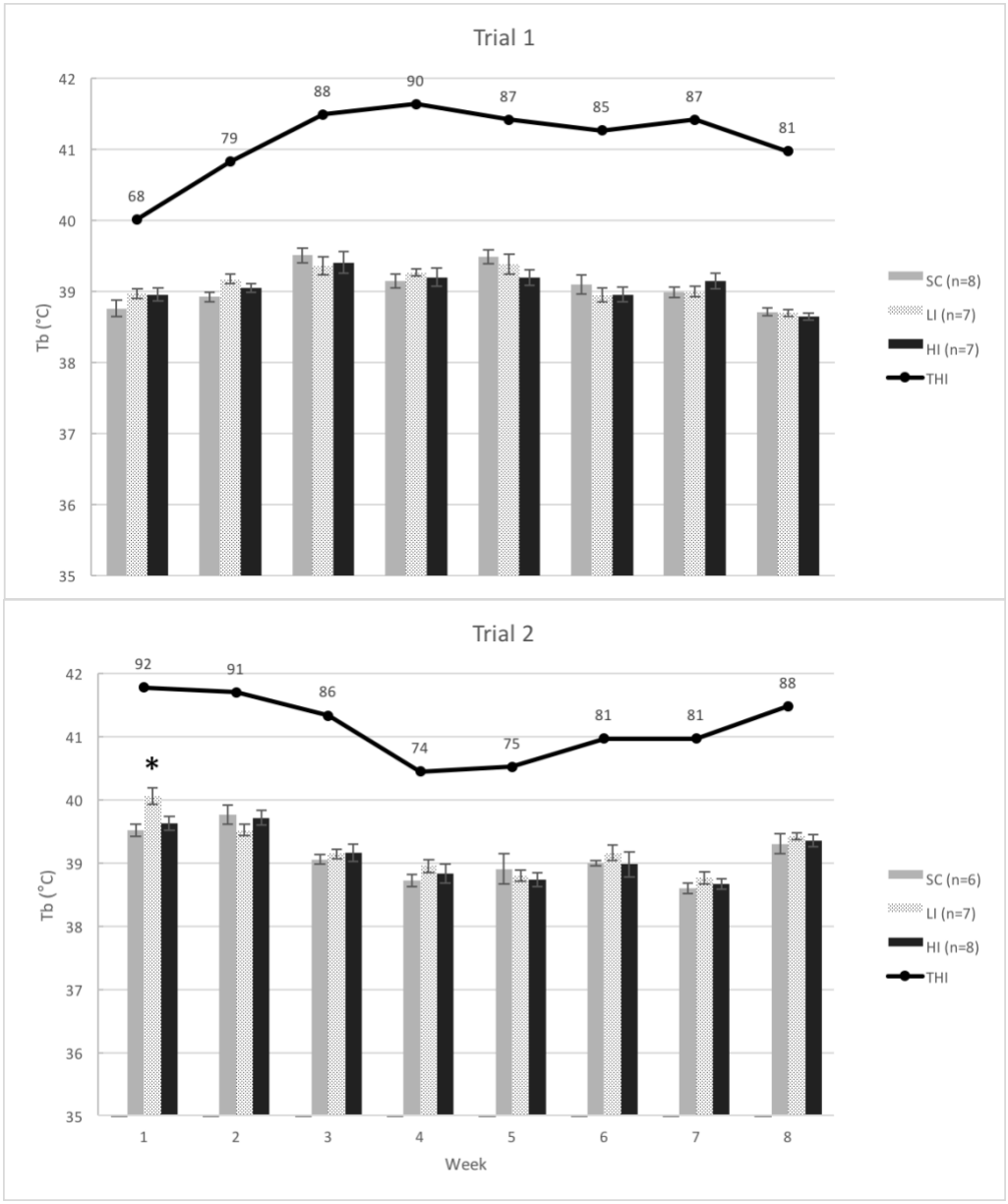

Figure 1.6. **Mean core body temperatures (Tb).**

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 data were collected between 16 May – 18 July of 2016 and trial 2 between 25 July – 26 September of 2016. The common x-axis displays the week of data collection for both

trials. Temperature humidity index (THI) for the date of each weekly measurement is indicated by the number above each data point on the line. Core body temperatures were collected on a rest day from exercise. Data are presented as mean $\pm$ SEM. The LI treatment had significantly higher Tb than the HI and SC treatments at week 1 of trial 2 ($p < 0.05$).

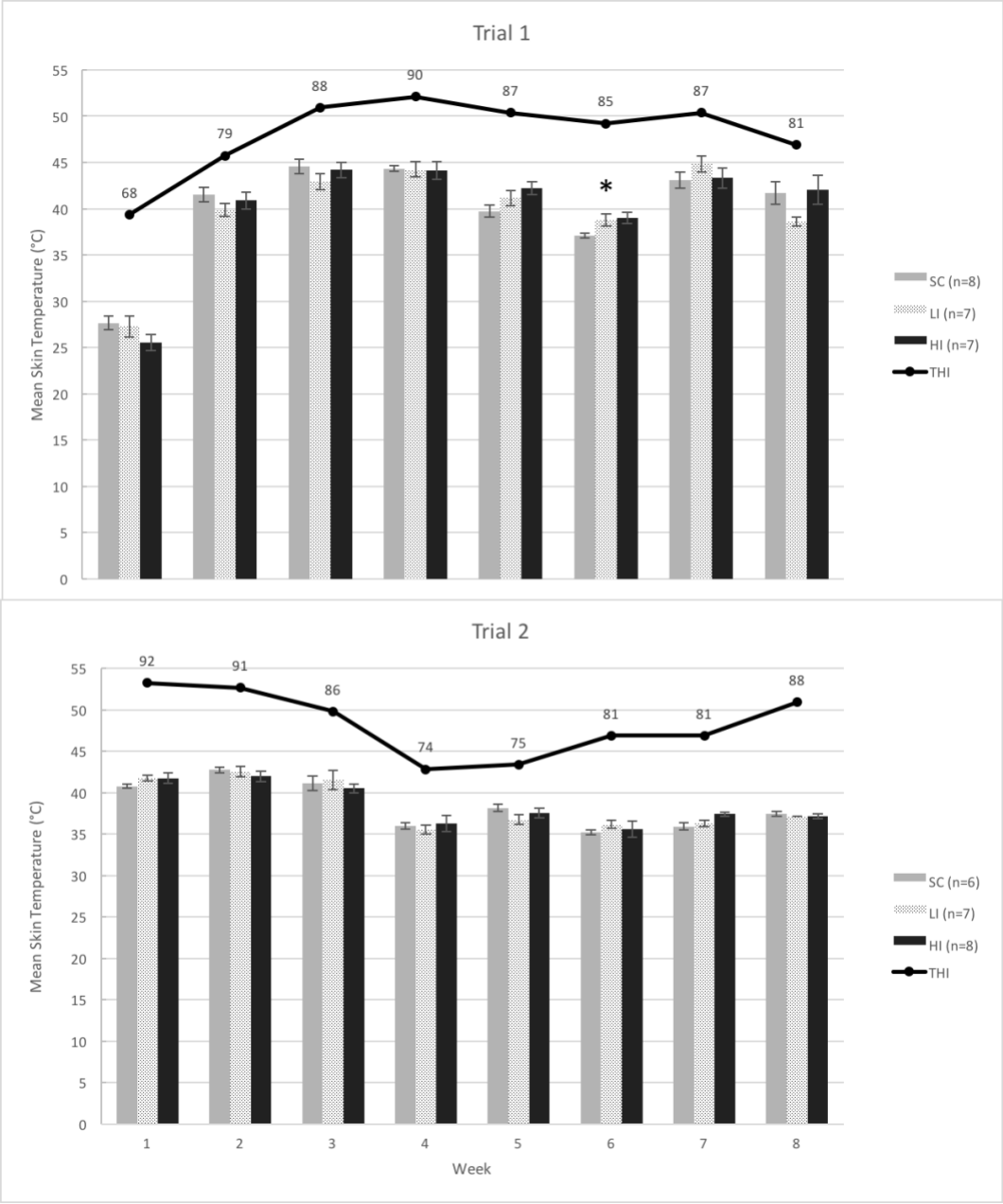

Figure 1.7. **Mean skin temperatures.**

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 data were collected between 16 May – 18 July of 2016 and trial 2 between 25 July – 26 September of 2016. The common x-axis displays the week of data collection for both trials. Temperature humidity index (THI) for the date of each weekly measurement is indicated by the number above each data point on the line. All data shown were collected on a rest day

from the center of the masseter muscle, the right pinna, dorsal ridge between the scapulae, in the center of a line between the ilium and ischium, and at the caudal midline between the two hindquarters. Data are presented as mean ± SEM. The HI treatment had significantly higher MST than the SC treatment at week 6 of trial 1 ($p < 0.05$).

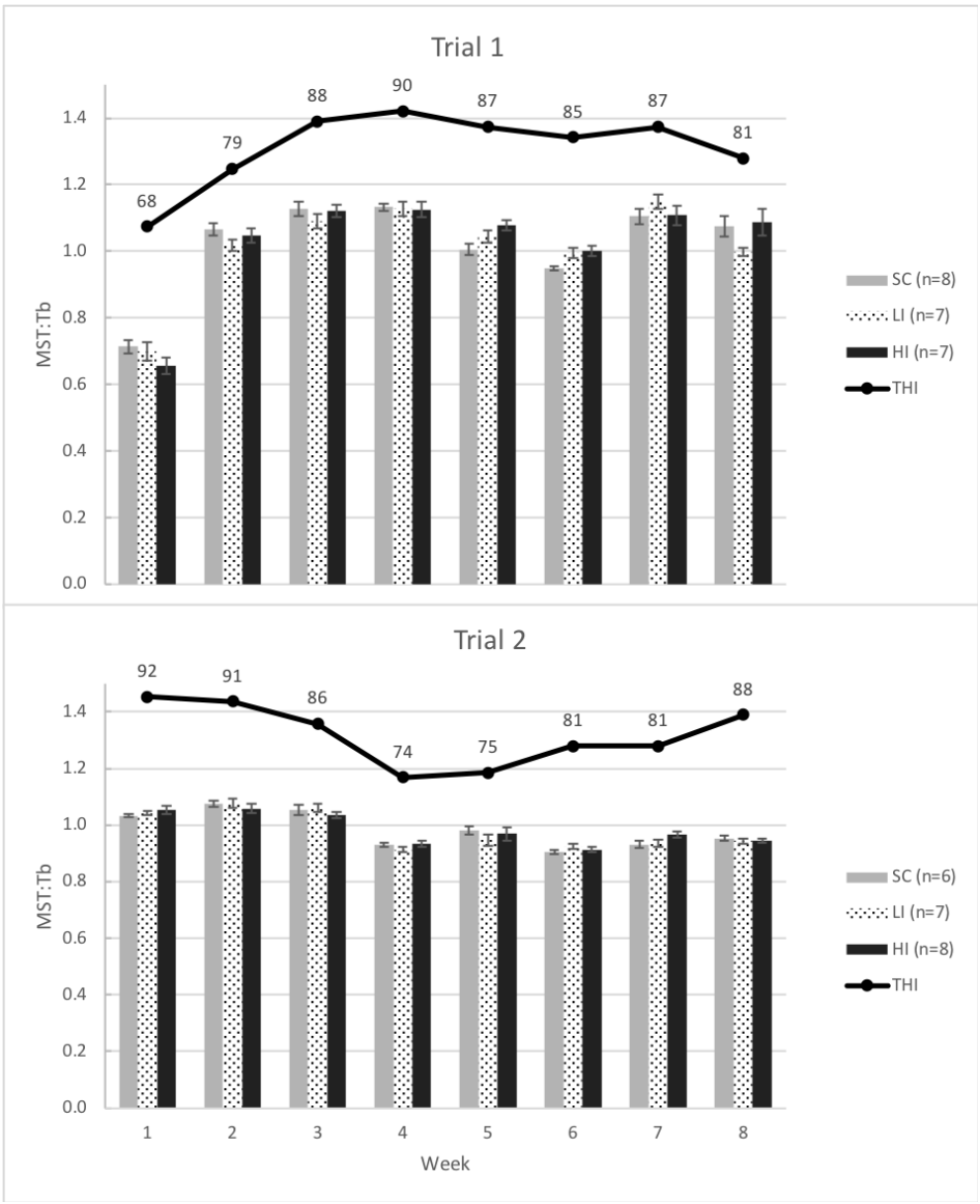

Figure 1.8. **Mean skin temperature (MST): core body temperature (Tb).**
Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 data were collected between 16 May – 18 July of 2016 and trial 2 between 25 July – 26 September of 2016. The common x-axis displays the week of data collection for both trials. Temperature humidity index (THI) for the date of each weekly measurement is indicated by the number above each data point on the line. Data are presented as mean ± SEM.

Pearson correlations

There were moderate to strong and positive correlations between THI and core body temperature (Tb), mean skin temperature (MST), and mean skin temperature:core body temperature (MST:Tb) for all treatments (Table 1.3). There were strong and positive relationships between the 5 individual skin temperatures and THI (Table 1.4). All correlations were significant at the level of $p < 0.001$. To observe relationships within individual treatments, data are combined from both trials and include all weeks of exercise.

Table 1.3. **Pearson Correlations between THI and core body temperature (Tb), mean skin temperature (MST), and MST:Tb.**

Data sets were combined between trial 1 and trial 2 for all weeks of exercise. Treatments: sedentary control (observations=112), low-intensity (observations=112), and high-intensity (observations=120).

Treatment		Tb	MST	MST:Tb
Sedentary Control	*THI*	0.577**	0. 393**	0.657**
Low-Intensity	*THI*	0.549**	0. 463**	0.730**
High-Intensity	*THI*	0.522**	0. 467**	0.675**

Correlations were significant (**) at the level of $p < 0.001$.

Table 1.4. Individual Skin Temperature correlations with THI.

Treatment		Cheek	Ear	Withers	Thurl	Udder
Sedentary Control	*THI*	0.759**	0.703**	0.611**	0.652**	0.715**
Low-Intensity	*THI*	0.816**	0.786**	0.734**	0.729**	0.736**
High Intensity	*THI*	0.733**	0.724**	0.647**	0.644**	0.675**

Correlations were significant (**) at the level of $p < 0.001$.

Linear Regressions

Using multiple linear regression analyses (Figures 1.9, 1.10, and 1.11), no significant differences were found between treatments in predicted core body temperature (Tb), mean skin temperature (MST), or MST:Tb. Regression equations are presented in Appendix Table A.3.

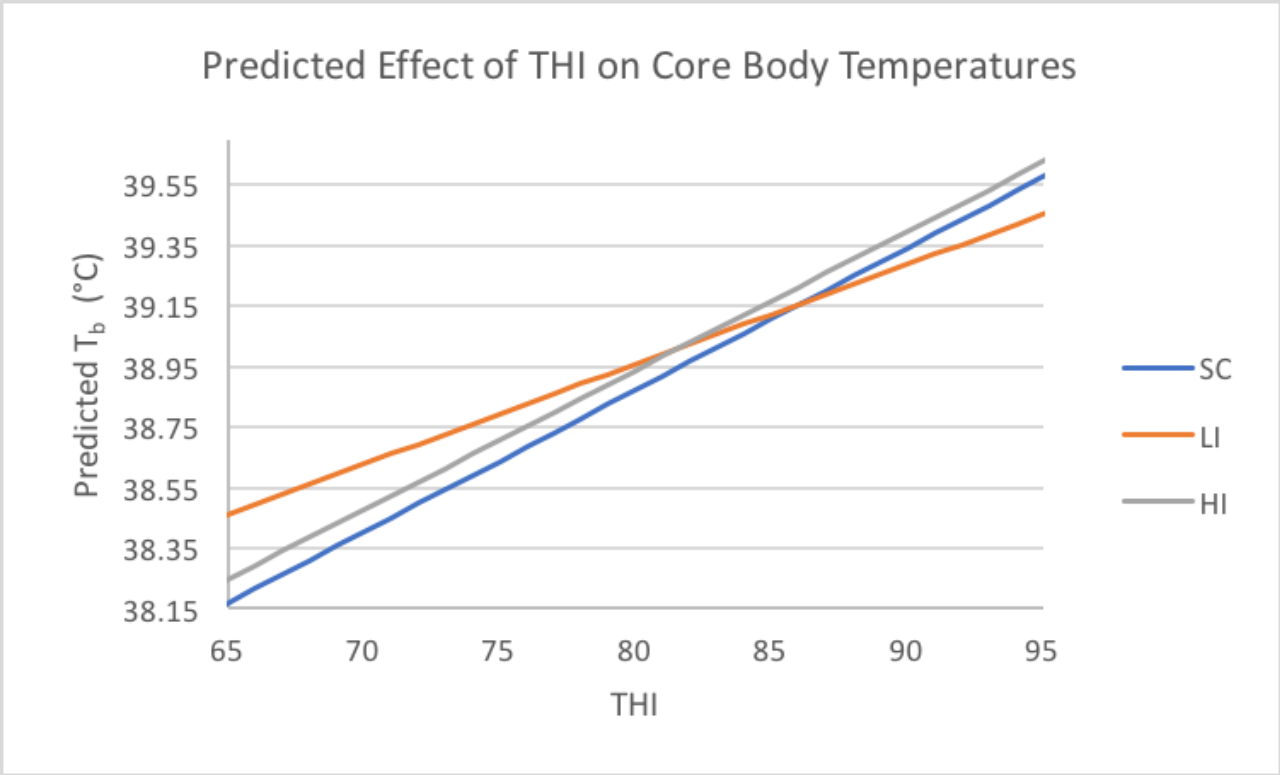

Figure 1.9. **Linear regression results of predicted THI on core body temperature (Tb).** Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Linear regressions were performed on THI and Tb data collected on rest days from trials 1 and 2 to generate regression equations, which were then used to calculate predicted Tb at a range of THI conditions between 65 and 95. No significant differences ($p > .05$) were found between these predictions by treatment.

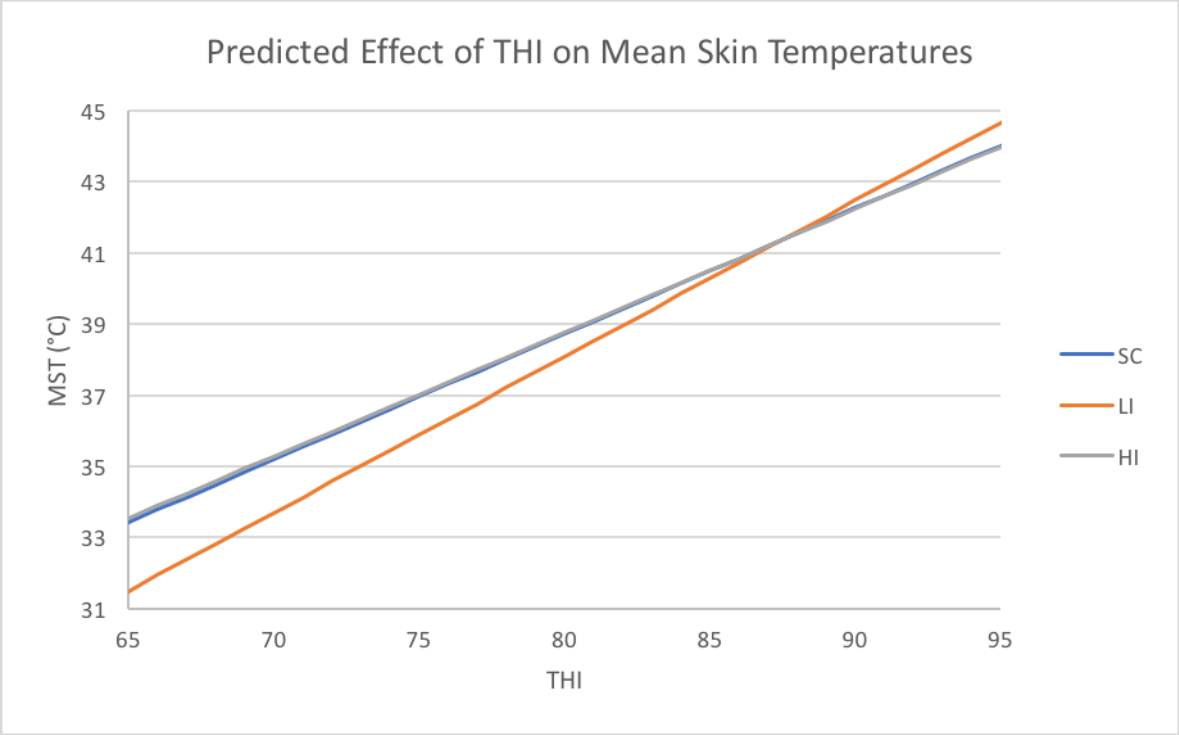

Figure 1.10. **Linear regression results of predicted THI on mean skin temperature (MST).** Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Linear regressions were performed on THI and MST data collected on rest days from trials 1 and 2 to generate regression equations, which were then used to calculated predicted MST at a range of THI conditions between 65-95. was used to calculate predicted MST. No significant differences ($p > .05$) were found between these predictions by treatment.

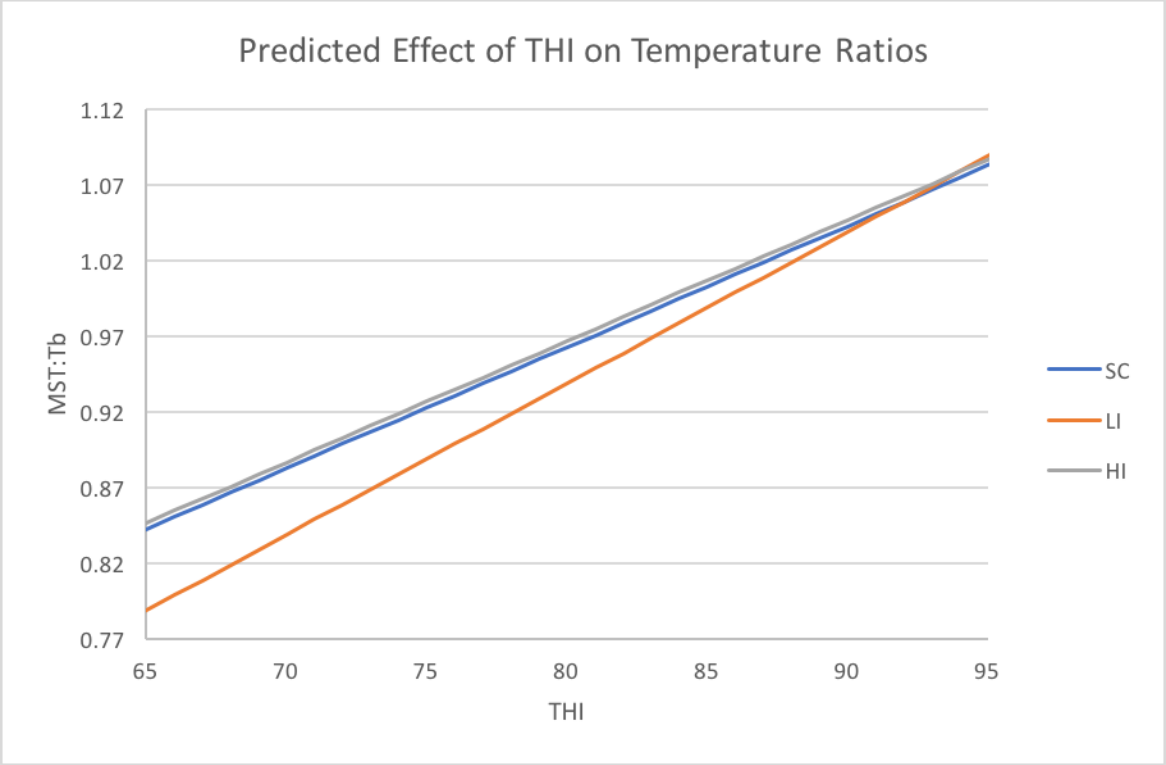

Figure 1.11. Linear regression results of predicted THI on mean skin temperature (MST):core body temperature (Tb).

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Linear regressions were performed on THI and MST: Tb data collected on rest days from trials 1 and 2 to generate regression equations, which were then used to calculate predicted MST: Tb at a range of THI conditions between 65 and 95. No significant differences ($p > .05$) were found between these predictions by treatment.

Body Weights and Body Condition Scores

There were no differences in body weight between treatments for trial 1 or trial 2 (Figure 1.12). Additionally, there were no differences in body condition scores for trial 2. Body condition scores (BCS) within each treatment were averaged for weeks 1, 2, 3, 5, and 8 (Figure 1.13).

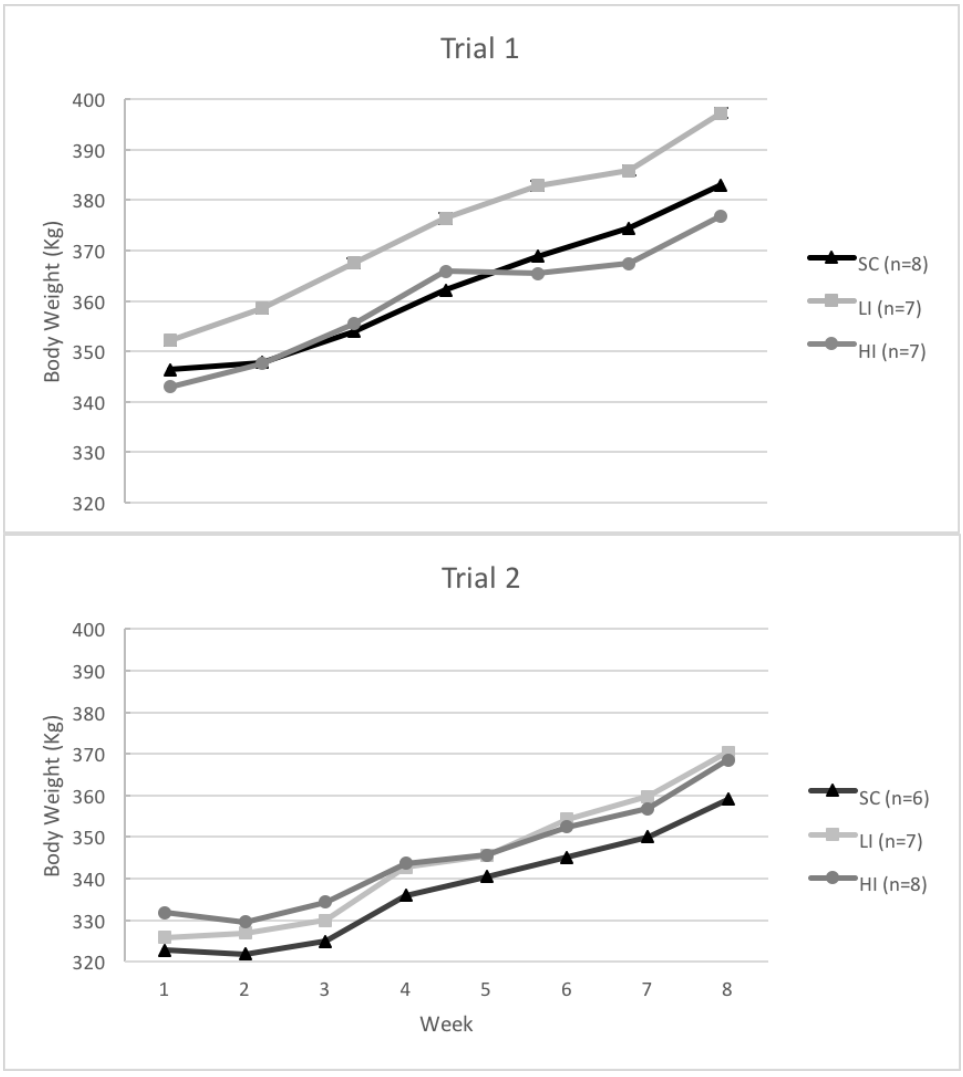

Figure 1.12. **Mean body weights.**

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 data were collected between 16 May – 18 July of 2016 and trial 2 between 25

July – 26 September of 2016. The common x-axis displays the week of data collection for both trials. Each point represents the mean body weight for each week for a particular treatment.

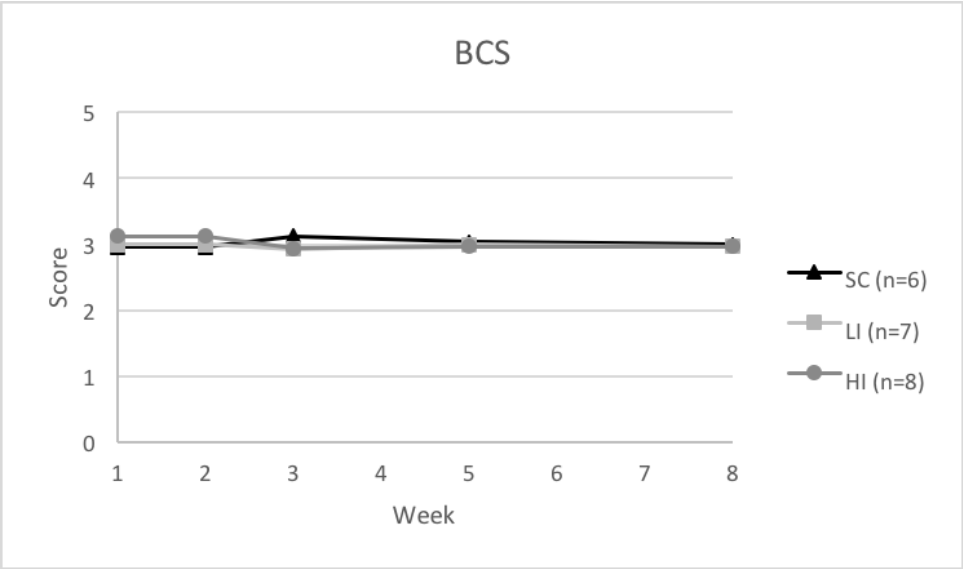

Figure 1.13. **Mean body condition scores (BCS) for trial 2.**
Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Lines represent the average BCS for each treatment taken at weeks 1, 2, 3, 5, and 8. The scale used for body condition scores ranged from 0-5; mean scores in the dataset ranged from 2.93 to 3.13.

Conception Rates

Conception rates were not significantly different between treatments for trial 1 or 2. Seven heifers were removed from the dataset due to injury, sickness, or failure to conceive and no conception rates were recorded. Therefore, *n* is reduced in trial 1: sedentary control (n=7), low-intensity (n=5), and high-intensity (n=7), and trial 2: sedentary control (n=5), low-intensity (n=5), and high-intensity (n=7).

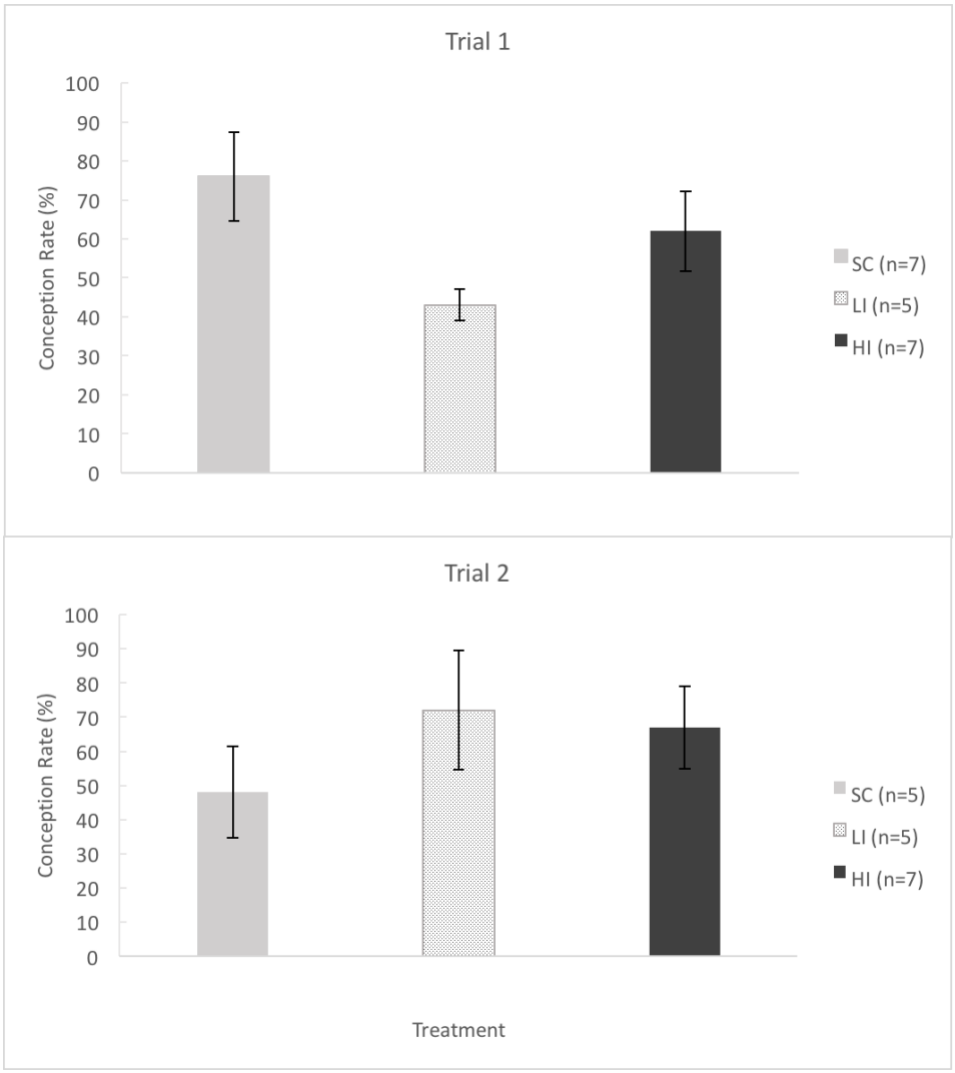

Figure 1.14. **Overall conception rates.**

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 data were collected between 16 May – 18 July of 2016 and trial 2 between 25 July – 26 September of 2016. The common x-axis displays treatment groups. Data are presented as mean $\pm$ SEM of the overall mean conception rate within each treatment.

Discussion

Using results from the first known LT test reported in Holstein heifers, the effects of low- and high-intensity exercise training on thermoregulation were investigated. Neither low- nor high-intensity exercise had consistently significant effects on thermoregulation in dairy heifers as indicated by core body temperature Tb and mean skin temperature (MST) measured at each week across two eight-week exercise trials. There were differences in mean skin temperatures between the high-intensity and sedentary control treatments, could have been because heifers entered the chute randomly for data collection and the majority of heifers in the high-intensity treatment (57%) were last to enter the chute (Appendix Table A.4). The order in which heifers entered the chute could also explain significant increases in Tb in the low-intensity treatment at week 1 of trial 2; as data were gathered from most of these heifers (57%) towards the end of collection (Appendix Table A.5). Differences could have been due to extended periods of heat exposure, from the natural environment or from being in the chute, in one treatment; although because significant differences in MST were not parallel to differences in Tb, these results remain inconclusive. In addition, temperature measurements were only collected one time per week, which could have contributed to errors in data collected.

While differences in weekly skin and core body temperature measures between treatments were inconsistent, there were moderate to strong, positive correlations between THI and Tb, individual skin temperatures, mean skin temperatures, and mean skin temperatures:Tb. Correlation results led us to further investigate these relationships via linear regression analyses; and although not significant, there were some thermoregulatory trends that may be a result of the type of exercise treatment. Heifers from the low-intensity treatment appeared to have a lower predicted Tb and higher predicted mean skin temperatures as THI increases. However, Tb and mean skin temperatures of heifers in the high-intensity and sedentary control treatments were

predicted to increase with THI. These trends led us to consider that low-intensity exercise training could perhaps lead to increased blood flow to the dermis at higher THIs as a response to lower Tb.

Blood flow to or through the skin was not directly measured, the predicted Tb and mean skin temperatures from low-intensity exercise could be an indicator of increased blood flow to the dermis, which is similar to results found in other mammals. Others have found that exercise training can lead to an increased rate and volume of cutaneous blood flow to dissipate heat, and thus decrease core body temperature, especially through exercise training in the heat (Fritzsche & Coyle, 2000; Geor, McCutcheon, et al., 1996; González-Alonso & Teller, 1999; González-Alonso et al., 2008; Hessemer, Langusch, Brück, Bödeker, & Breidenbach, 1984; Périard et al., 2016). Decreased Tb through shunting blood to the skin has been reported as an effective thermoregulatory response humans and horses; the onset of an increase in blood flow to the skin is dependent upon the stimulus or type of exercise (Art & Lekeux, 2005; Geor, McCutcheon, et al., 1996; Lindinger, McCutcheon, Ecker, & Geor, 2000; Rowell, Brengelmann, Blackmon, & Murray, 1970; Wyss, Brengelmann, Johnson, Rowell, & Niederberger, 1974). Furthermore, exercise intensity has been thought to influence improved thermotolerance in humans and horses; for example, as exercise intensity amplified, skin blood flow and sweat rate increased while Tb decreased (Kondo et al., 1998; McCutcheon & Geor, 2000). However, others have shown that low-intensity or endurance exercise training improved heat tolerance; this was quantified by a reduction in core body temperature and increased skin blood flow in humans and horses (Flaminio & Rush, 1998; Fritzsche & Coyle, 2000; Hessemer et al., 1984). Humans and horses rely on evaporative cooling, or sweat production, to dissipate heat. As core body temperature (Tb) rises in response to exercise in humans, blood flow to the dermis, sweat rate, and sweat

gland number increased (Simmons et al., 2011). Although some mammals can regulate body temperature efficiently through evaporative cooling like sweating, there are very few eccrine sweat glands found in the dermis of cattle. Instead, cattle rely on increasing respiration and panting to regulate body temperature when ambient temperatures rise above the upper critical temperature (25ºC) (Berman et al., 1985; Carvalho, Lammoglia, Simoes, & Randel, 1995; Finch, 1986). However, it has been reported that some cattle have acclimated to tropical or warmer climates such that they increased the number and size of sweat glands in the dermis (Carvalho et al., 1995). Therefore, it can be speculated that if cattle that are exposed to hot and humid environments are exercised in the heat, there may be a similar response to that found in humans and horses.

An increase in blood flow to the dermis for extended periods of time can result in physiological changes resulting in enhanced blood flow and thus reduced heat load. Some studies have shown that shear stress, or mechanical stress from an increase in blood flow to the dermis, can increase cutaneous vasculature, similar to changes in skeletal muscle vasculature (Simmons et al., 2011). Shear stress is one type of angiogenic stimulus which has been shown to activate one of many vascular endothelial growth factor (VEGF) receptors. The VEGF receptors are mitogenic and chemotaxic proteins that are thought to promote angiogenesis (Auerbach et al., 2003; Prior et al., 2004). Vasodilation and angiogenesis will occur when energy demand exceeds energy supply (Auerbach et al., 2003; Crawford et al., 2006; Patan, 2000; Semenza, 2007; Vailhé et al., 2001). In light of these previous findings with vascular remodeling or development, shear stress from low-intensity exercise training in dairy heifers could have elicited angiogenesis in the dermis; however, this experiment was not designed to measure these outcomes.

Vascular remodeling can also occur in other tissues such as skeletal muscle. Exercise training has been shown to increase the number of capillary beds within skeletal muscle tissue in horses, humans, and dogs (Egan & Zierath, 2013; Eivers et al., 2010; Gollnick & King, 1969; J. Holloszy, 1967; Holloszy & Coyle, 1984; Ingjer, 1979; Williams et al., 1986; Wu et al., 2002; Yan, Lira, & Greene, 2012; Young, Mosher, Erve, & Spector, 1959). To identify changes in the vasculature, specific cellular mRNA or proteins can be measured to predict such physiological changes within skeletal muscle cells (Dubouchaud et al., 2000; Friedmann et al., 2004; Gibala et al., 2012; Liang et al., 2016; McGinley & Bishop, 2016; Summermatter et al., 2013). One such protein includes lactate dehydrogenase (LDH). The lactate dehydrogenase enzyme, found within skeletal muscle cells and other tissues, can be observed by isoform type (LDH1, LDH2, LDH3, LDH4, and LDH5) or the LDH mRNA subunit expression (LDHa and LDHb). Using mRNA subunits to measure changes in LDH in skeletal muscle, there were significant decreases in LDHa within skeletal muscle cells after high-intensity exercise training. Additionally, LDHb was significantly reduced in skeletal muscle cells within the sedentary control and high-intensity treatments. These results were not consistent with others who have reported changes in LDH protein isoforms (Apple & Rogers, 1986; Booth & Thomason, 1991; Leberer & Pette, 1984; Summermatter et al., 2013); however, these results were similar to others who did not find significant changes in LDH subunit mRNA expression within skeletal muscle cells from humans (Nordsborg, Lundby, Leick, & Pilegaard, 2010) and horses (Eivers et al., 2010). Thus, measuring LDH mRNA versus protein isoforms could impact results. Inconsistencies in LDH subunit mRNA and protein expression could be due to biopsy timing. Based on results reported by other researchers, the amount of time between the last bout of exercise and the muscle biopsy can have implications on measuring cellular activity because mRNA and protein kinetics can

vary from exercise training (Egan & Zierath, 2013; Jensen, Pilegaard, Neufer, & Hellst, 2007; McGinley & Bishop, 2016). In this study, muscle biopsies were taken three days after the last bout of exercise. By the time the biopsy was taken mRNA activity could have been significantly reduced. Furthermore, it is important to consider that there appears to be variation of LDH isoenzymes in skeletal muscle. Researchers have reported higher quantities of LDH-M (LDH2, LDH3, LDH4, LDH5) in fast twitch skeletal muscle fibers while increased levels of LDH-H (LDH1, LDH2, LDH3, LDH4) has been found in slow twitch skeletal muscle fibers (Apple & Rogers, 1986; Booth & Thomason, 1991; Leberer & Pette, 1984; Summermatter et al., 2013). In congruence with these findings, others have found that endurance exercise training can increase LDH-H in skeletal muscle cells, along with increases in mitochondrial density, citrate synthase, and monocarboxylate transporter 1 (MCT1); all of which indicated an increased oxidative capacity within skeletal myocytes (Brooks et al., 1999; Dubouchaud et al., 2000; McCullagh et al., 1997; McGinley & Bishop, 2016). While not significant, it is interesting that LDHb mRNA was higher than LDHa from the semitendinosus muscle of all treatments; a muscle that is thought to contain fast and slow-twitch muscle fibers (Listrat et al., 2016; Wegner, Albrecht, & Fiedler, 2000). Although the mRNA results do not coincide with others who have investigated LDH protein expression after exercise training, this could be a reflection of biopsy timing and requires further investigation.

In addition to altered oxidative capacity and blood flow patterns, exercise training could have other physiological impacts, such as changes in body mass or reproduction. Much like heat stress, exercise training has influenced the ability to maintain body weight and to conceive in humans and horses; however, these issues typically arise from elite or intense exercise (Campbell, 2014; Cumming, Wheeler, & Harber, 1994; Warren & Perlroth, 2001). Exercise

intensities did not result in stunted growth or reduced conception rates in this study. It is important to note that conception rates for heifers are generally lower than cows (Kuhn, Hutchison, & Wiggans, 2006), however exercise did not affect conception in heifers. These data are consistent with those reported by Johnson et al. (2016).

Conclusions and Future Directions

Thermoregulation and heat stress in livestock are becoming more crucial factors as the climate continues to change and temperatures continue to rise. This becomes increasingly important as the human population continues to grow and food demand increases (Vorosmarty & Green, 2000; Zahid et al., 2016). To avoid depleting natural resources and increasing the cost of food, the search to overcome these challenges that will only continue to grow with global climate change and population growth must continue.

The trends found in this study warrant further investigation of the effects of low-intensity exercise training in dairy heifers. Although there were no differences in thermoregulatory responses between treatments, it is possible that heifers could require a longer period of low-intensity exercise training with a larger sample size to detect significant changes. While humans and horses have been shown to acclimate within 4 weeks of exercise training (Jensen et al., 2007), humans are more efficient thermoregulators because of the number of eccrine sweat glands in the dermis. Therefore, it would be worth exploring the effects of 12- or 16-week exercise training programs with 16-24 heifers in each treatment. Furthermore, longer exercise training programs could elicit physiological alterations within the dermis and skeletal muscle cells. To gain a more complete understanding of cellular changes within these tissues, capillary density, mitochondrial volume, VEGF, MCT-1 and -4, and LDH isoenzymes should be investigated.

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Chapter 2 - Active Learning vs. Lecture Learning: Student Characteristics Matter

Literature Review

By the year 2022, more than 9 million jobs in science, technology, engineering, and math (STEM) are expected to be added to the United States' workforce (Vilorio, 2014). Even though 92% of these STEM occupations require a postsecondary degree, only 300,000 students in the U.S. graduate from STEM fields each year; thus, there is a large gap between the number of STEM graduates actually completing a degree and the number of STEM job vacancies (Graham, Frederick, Byars-Winston, Hunter, Handelsman, 2013; Olson & Riordan, 2012). In fact, only 40% of those who enroll in STEM fields actually graduate with a baccalaureate degree in STEM (Freeman et al., 2014; Graham et al., 2013; Olson & Riordan, 2012). Compounding this issue is the loss of many skilled workers from STEM fields, largely due to increased retirement rates of the baby boomer generation. In addition, there is an increase in the number of foreign-born workers filling positions in STEM fields (Colby & Ortman, 2014; Hossain & Robinson, 2012). This amalgamation of issues will result in less native-born citizens filling secure STEM jobs within the U.S. Thus, to invest in U.S. citizens and remain competitive in the global market of science and technology, members of the President's Council of Advisors on Science and Technology (PCAST) (2012) have recommended an increase of one million STEM graduates by 2022 (Banning & Folkestad, 2012; Colby & Ortman, 2014; Freeman et al., 2014; Graham et al., 2013; Hossain & Robinson, 2012; Wang & Degol, 2014).

Attrition of undergraduate students from STEM fields is a result of many factors including negative experiences in college courses such as dull curricular instruction, lack of confidence, lack of preparation, and in-class experiences (Graham et al., 2013; Maltese & Tai,

2011; Olson & Riordan, 2012; Rhodes & Rozell, 2014; Watkins & Mazur, 2013). Such in-class experiences include classroom environment, personal support, relationships, and student involvement (Osborne, Simon, & Collins, 2003; Roff et al., 1997; Veltri, Banning, & Davies, 2006; Watkins & Mazur, 2013). In addition, pre-college factors can impact persistence within STEM fields; including demographics, family background, and academic preparation (Chen, 2009; Chen & Soldner, 2013; Hill, Corbett, & St Rose, 2010; Radford & Horn, 2013; Rask, 2010). Furthermore, attrition in STEM has been linked to academic success and self-efficacy (Ackerman, Kanfer, & Beier, 2013; Bandura, 1997; Chen & Soldner, 2013; Gasiewski, Eagan, Garcia, Hurtado, & Chang, 2012; Majer, 2009; Watkins & Mazur, 2013; Whalen et al., 2010). Students who aim to pursue STEM careers tend to set high expectations for academic success; however, if these expectations are not met, students quickly lose self-efficacy and motivation to continue in STEM (Chen & Soldner, 2013; Graham et al., 2013; Olson & Riordan, 2012). The importance of confidence and motivation has been recognized by Graham et al. (2013) in the Journal of Science through the persistence framework which described the momentum initiated with confidence which has led to motivation in students to actively engage in information and research.

In addition to the impacts of self-efficacy on persistence in STEM, the majority of students who left STEM fields also reported poor quality instruction in STEM courses contributed to abandoning STEM fields (Watkins & Mazur, 2013). As such, instructional approach within college classrooms has become a focal point to improve persistence in STEM (Bonwell & Eison, 1991; Freeman et al., 2014; Graham et al., 2013; Olson & Riordan, 2012; Prince, 2004). In fact, members of the PCAST (2012) cited the first two years of postsecondary education as the most crucial for recruiting and retaining students within introductory and

intermediate STEM courses (Olson & Riordan, 2012). Moreover, poor performance and negative experiences in introductory courses that have led to fleeing STEM have been attributed to many factors including large classroom sizes, uninspiring pedagogy and covering copious amounts of information in short timeframes (Scott, Mcnair, Lucas, & Land, 2017). While poor classroom experiences negatively impact all students, it is possible that this could elicit greater and longer lasting impacts on students with weaker academic backgrounds and poorer preparation for science courses. According to the National Center for Education Statistics (2013), low performing students in STEM (college GPA of 2.5 or less) or those with weaker academic backgrounds are more likely to drop out of college or switch to non-STEM majors versus high performing students in STEM (college GPA 3.5 or higher) (Chen & Soldner, 2013). Additionally, many students have reported that introductory STEM courses require memorization of information without practical implications and thus, students find these courses to be discouraging or irrelevant to their lives (George, 2006; Holmegaard, Madsen, & Ulriksen, 2014; Nolen, 2003). To improve student learning experiences and encourage persistence in STEM, more effective pedagogy must be utilized within introductory and intermediate courses where students are most likely to leave STEM (Olson & Riordan, 2012). As suggested by the American Association for the Advancement of Science (2011), this begins with identifying core concepts within specific disciplines (Brewer & Smith, 2011; Michael, Cliff, McFarland, Modell, & Wright, 2017); this is especially true for ill-structured domains like physiology.

Core Concepts in the Ill-Structured Domain of Physiology

Physiology is one sector of science that is comprised of intrinsically difficult concepts for students to learn. This is largely because understanding physiological mechanisms requires a strong foundation of other sciences including chemistry, physics, and biochemistry (Michael et

al., 2017; Rhodes & Rozell, 2017). Because physiology is multifaceted and challenging for students to learn, Michael et al. (2017) have defined core concepts in physiology as those that provide a foundation to better understand future mechanisms. Furthermore, these concepts should be widely applicable or transferrable to foster problem solving across all areas of physiology. The top five core concepts in physiology as described by Michael et al. (2017) include homeostasis, cell membranes, cell-cell communication, interdependence, and flow down gradients. These concepts are thought to be related or even overlap one another such that they interact together. For example, integral cell membrane proteins are often involved in cell-cell communication and maintaining homeostasis. These are considered crucial for understanding concepts within the ill-structured domain of physiology (Rhodes & Rozell, 2017).

Ill-structured domains are difficult for students because they are, by definition, non-linear and often require multiple representations for accurate portrayal whereas well-structured concepts are linear and can be understood through one representation (Spiro, Vispoel, Schmitz, Samarapungavan, & Boerger, 1987). Ill-structured domains, often found in medicine, history, and economics, require interconnectivity such that old knowledge must be assembled with new knowledge to solve novel problems (Rhodes & Rozell, 2017; Spiro, Coulson, Feltovich, & Anderson, 1988; Spiro et al., 1987). Ill-structured concepts often have multiple avenues to approach one correct answer; these scenarios can be found frequently in the field of physiology. For example, an increase in blood pressure can lead to a response from many different body systems, each of which act to accomplish the same goal to reduce blood pressure (Hall, do Carmo, da Silva, Wang, & Hall, 2015). This example holds true for most physiological mechanisms that aim to restore homeostasis.

Establishing an interconnected and flexible web of knowledge is essential for understanding and solving problems within ill-structured domains like physiology (Rhodes & Rozell, 2017; Spiro et al., 1988; Spiro et al., 1987). A flexible web of knowledge begins with knowledge elements, or ideas that can be grouped together as a schema or knowledge structure. Schemas can be organized or grouped together to form schemata, or "chunks" of information. The integration and transfer of schemata is known as *cognitive flexibility* (Spiro et al., 1987; Young, Van Merrienboer, Durning, & Ten Cate, 2014). Cognitive flexibility is described as the ability to assimilate information into a network and apply knowledge to various scenarios; this is crucial for advanced knowledge acquisition. Advanced knowledge acquisition involves a shift away from rote memorization or exclusive schema packaging and retrieval to flexible thinking that requires assembly and synthesis of schemata (Rhodes & Rozell, 2017; Spiro et al., 1988; Spiro et al. , 1987). According to Spiro et al. (1987), to advance knowledge acquisition and approach learning on a deeper level, students must "criss-cross the landscape." In other words, students must be exposed to multiple representations of information and recognize that many routes can lead to the same answer or solution to a problem (Rhodes & Rozell, 2017; Spiro et al., 1988; Spiro et al., 1987).

Because many concepts in physiology often involve a multiplicity of correct avenues that could be taken to arrive at the correct solution, they are inherently ill-structured and thus require cognitive flexibility. However, many students approach learning physiology as if it is well-structured and simply requires retrieval of rigidly packed schemata. Previous work by Rhodes and Rozell (2017) has shown that students who approach learning within ill-structured domains through memorization perform worse on unit exams than those who elaborate or use an elaborative or a more flexible approach to learning meaning they incorporate new information

with previously learned information. To encourage flexible thinking within ill-structured domains, university faculty have been encouraged by members of the PCAST to embrace various teaching techniques that are evidence based, with a strong emphasis on active learning (Freeman et al., 2014; Graham et al., 2013; Olson & Riordan, 2012; Rhodes & Rozell, 2017).

Active Learning

Active learning has been thought to engage students in learning ill-structured information (Bonwell & Eison, 1991; Brewer & Smith, 2011; Freeman et al., 2014; Graham et al., 2013; Olson & Riordan, 2012; Rhodes & Rozell, 2017). Tackling ill-structured concepts via active learning tools is similar to how scientists actually 'do' science as students are encouraged to develop their own hypotheses about concepts, problem-solve, draw logical conclusions from a given dataset and connect knowledge from different domains (Herreid, 1994) and is believed to increase cognitive flexibility and problem solving. Thus, active learning appears to be a logical approach to training future scientists, but students without adequate preparation may find educational approaches that require application of information to be intimidating and very complex (Brookfield, 1997).

Active learning can be broadly defined as any learning approach that requires students to be cognitively engaged with content. This form of learning can be implemented using a variety of teaching techniques as there is no one definition of what makes a learning activity 'active' as opposed to 'passive.' Instead, active learning is a term applied to any activity that emphasizes higher-order thinking rather than listening to or watching an instructor, and can include a myriad of instructional approaches such as engaging students in effective question and answer sessions, small group discussions or intimate debates, problem-based learning, case studies, collaborative learning, concept mapping, one minute papers, team-based learning, role playing/drama, flipped

classroom approaches, and even the use of carefully constructed and timed worksheets (Bonwell & Eison, 1991; Chi & Wylie, 2014; Cleveland, Olimpo, & Dechenne-peters, 2017; & Schiller, 2013; Knight & Wood, 2005; Menekse, Stump, Krause, & Chi, 2013; Rybarczyk, Baines, McVey, Thompson, & Wilkins, 2007; Stull & Mayer, 2007; Thistlethwaite et al., 2012; Zayapragassarazan & Kumar, 2012). Regardless of the form or format of the activity, the overarching goal of active learning is to increase the ability of students to reason with ill-structured concepts and apply what they have learned to novel situations. This could be especially helpful for students who are struggling in STEM courses and may help ease attrition issues.

Some researchers have found that active learning approaches improve learning gains when compared to traditional instructional approaches, such as the conventional lecture (Bonwell & Eison, 1991; Chickering & Gamson, 1987; Cleveland et al., 2017; Freeman et al., 2007, 2014; Graham et al., 2013; Knight & Wood, 2005; Prince, 2004; Roehl, Reddy, & Shannon, 2013; Rybarczyk et al., 2007). Bonwell & Eison (1991) described the conventional lecture as a passive and unengaging teaching approach. They reported many studies that found active instruction to be more effective, including pause lectures (lectures that provide a brief moment to think, question or discuss), discussion within the classroom, and other active learning techniques. Additionally, Knight & Wood (2005) found that implementing group work, class discussion, and the use of clicker questions resulted in greater learning gains than the traditional lecture. However, others have found that active learning approaches did not provide additional learning benefits (Andrews, Leonard, Colgrove, & Kalinowski, 2011; Colliver, 2000; Menekse et al., 2013; Sadler, 2002; Stull & Mayer, 2007; Wilson, 1999). Stull & Mayer (2007) conducted a large study on retention of scientific concepts that included multiple experiments and examined

the effect of scaffolding information for students versus requiring students to construct all information individually. Researchers concluded that learner generated graphics were not the best approach for all students. Wilson (1999) found that student interaction with a computer program was no more effective on memory than passively watching the virtual simulation. In addition, Andrews et al. (2011) sampled an array of introductory biology courses across the U.S., each course instructor reported whether the course consisted of primarily active learning approaches or traditional lectures. Andrews et al. (2011) then measured student learning outcomes on natural selection, a core concept for introductory biology students, within these courses and reported no differences between courses that used active learning vs. passive learning.

Part of the inconsistency in the literature likely arises from the lack of a clear definition of what specifically constitutes an active learning activity. For example, pause lectures, class discussion, case studies, collaborative learning, graphic designing, one minute papers, and flipped classrooms have all been considered forms of active learning (Bonwell & Eison, 1991; Chi & Wylie, 2014; Cleveland et al., 2017; Herreid & Schiller, 2013; Knight & Wood, 2005; Menekse et al., 2013; Rybarczyk et al., 2007; Stull & Mayer, 2007; Thistlethwaite et al., 2012; Zayapragassarazan & Kumar, 2012). Thus, defining and operationalizing activities that are thought to be forms of active learning and then systematically evaluating their effectiveness in the classroom is crucial for ascertaining the specific benefits provided by this type of learning.

As displayed in Table 2.1, variation exists in the studies that have investigated active learning, including the definition of active learning, sample size of the study, prior knowledge levels of the students, active learning tools and forms used, time frame of the intervention,

experimental design, and findings of the study. Such variations make it difficult to decipher whether active learning is useful for all courses and populations of students.

Table 2.1. Active Learning Meta-Analysis. A review of articles investigating the utility of active learning in college courses. Active learning definitions vary across studies. Boxes containing "N/A" indicate that active learning was not defined in the publication, or further information is not provided as some of these publications were reviews, meta-analyses, or books.

Source	Active Learning Definition/ Description	Course Size	Course Level	Experimental Time Frame	Experimental Design	Findings/Conclusion
Kim, Sharma, Land, & Furlong, 2012	N/A	155	Intro to geoscience	Week 6 & 12 of the course	No experimental design	Active learning strategies increased critical thinking and enhanced student learning.
Haak et al., 2011	N/A	345 in first 5 quarters and 700 in the last quarter	Intro to biology	6 quarters	No experimental design	Active learning reduced the achievement gap between students and enhanced higher-order thinking.
Freeman et al., 2014	Group problem-solving, worksheets, peer instruction, and studio courses.	Meta-analyzed 225 studies	Various STEM courses	N/A	No experimental design	Active learning led to exam performance gains and higher grades.
Michael, 2006	Engaged students in an activity that allowed them to reflect upon ideas.	N/A	Physics, chemistry, biology, undergraduate and graduate level physiology	N/A	No experimental design	Active learning worked better than passive learning approaches.
Roehl, Reddy, Shannon, 2013	An umbrella term for pedagogies focused on student activity & engagement	N/A	N/A	N/A	N/A	Through active learning, students may develop higher order thinking and creativity.
Meneske, 2013	Involved students in the learning process.	42	Intro to science and engineering	5 days	True experimental design	Active forms of learning were more effective than passive forms.
Rybarczyk et al., 2007	N/A	157	General biology and Intro to cell biology	5 days	Quasi-experimental design	Case studies increased learning gains more than lecturing.

Wilke, 2003	Involved students reflecting on assignments.	141	Sophomore level human physiology	1 semester	Quasi-experimental design	Active learning did not result in improved achievement scores.
Nogaj, 2013	N/A	47	Intermediate molecular biology	1 semester	No experimental design	Active learning improved student performance.
Wolff et al. 2015	Allows students to retain and understand knowledge.	N/A	N/A	N/A	N/A	Incorporating active learning into lectures increased learning.
Freeman et al., 2007	N/A	345	Intro to biology	2 semesters	No experimental design	Active learning increased student performance.
Benware & Deci, 1984	Learning that is done with the expectation of using the material.	43	Intro to psychology	1 week	True experimental design	Active learning resulted in greater learning gains.
Skoloff & Thornton, 1997	N/A	444	Introductory physics	Several semesters	No experimental design	Active learning increased student understanding.
Tune, Sturek, & Basile, 2013	Used problem-based learning with or without traditional didactic lectures.	27	Graduate level physiology	1 semester	No experimental design	Student's in the flipped classroom earned higher exam grade averages than student's in a traditional classroom.
Andrews, et al., 2011	Instructor stopped lecturing and students worked on a question or task.	N/A	Intro to biology	1 semester	No experimental design	Active learning was not associated with student learning.
Cavanagh et al., 2016	Provided students with opportunities to engage in the learning process.	245	Intermediate human anatomy and physiology	1 semester	No experimental design	Student learning gains improved from student buy-in, or feelings, toward active learning.
Deslauriers, Schelew, & Wieman, 2011	N/A	382	Intro to physics	1 week	No experimental design	Active learning improved learning and engagement.
Wiggins et al., 2017	Students co-constructed knowledge with their peers.	408	Intro to biology	N/A	N/A	Student experiences with active learning influenced engagement.
Ernst & Colthorpe, 2008	Students engaged in an activity that required reflection upon ideas.	60	Pharmacy school human physiology and pharmacology	3 years	No experimental design	Voluntary active learning improved student exam scores and provided a decrease in the number of underachieving students.

Pierce & Fox, 2012	Interactive activities allow the learnerto take responsibility and ownership of learning.	71	Advanced pharmacy course	90 minutes	A design experiment	Active learning improved student performance and generated positive attitudes.
Zayapragassarazan and Kumar, 2012	Promotes proper knowledge, attitude, and skills.	N/A	N/A	N/A	N/A	Students learned better in active learning environments.
Diamond, Koering, & Iqbal, 2008	Students actively engaged with and personalized information.	69	Undergraduate marketing course	1 semester	No experimental design	Active learning increased deep understanding.
Stull & Mayer, 2007	Learning by viewing.	370	N/A	N/A	Posttest control group design.	Increased activity by the learner did not improve learning gains.
Chaplin, 2009	Allows students to connect information to personal experiences.	46-50	Introductory biology	By semester: data collected in 1998 and 2006	No experimental design	The use of case studies improved exam scores.
Burrowes, 2003	N/A	204	Intro to biology	1 semester	Quasi-experimental design	Active learning was more effective than traditional instruction.
Felder & Brent, 2009	Anything teachers ask students to do.	N/A	N/A	N/A	N/A	Active learning increased quality of learning.
Paulson, 1999	Strategies that involve activity or discussion in the classroom.	N/A	Organic chemistry	Several semesters	No experimental design	Active learning strategies increased retention.
Hall, Waitz, Brodeur, Soderholm, & Nasr, 2002	Techniques that stress student's active involvement in their own learning.	N/A	Unified Engineering	Several years	No experimental design	Students reported active learning techniques helped deepen their understanding of concepts.
Briscoe & LaMaster, 1991	Meaningful learning.	6	Community college biology	One semester	No experimental design	Students retained information and constructed knowledge in their own terms.
Dori & Belcher, 2005	N/A	809	Introductory physics	Several semesters	No experimental design	Active learning improved conceptual understanding.

In addition to a lack of what constitutes as active learning, there is a large amount of variation in the analytical methods used to evaluate its effectiveness. Some active learning studies lack of a true experimental design, use small sample sizes, or lack a defined experimental timeframe that controls for threats to validity; these inconsistencies may contribute to the conflicting results regarding the benefits of active learning for all students. For example, Dori & Belcher (2005) found technology-based active learning to be beneficial for students in an introductory physics course at Massachusetts Institute of Technology; a very specific population of students. Furthermore, Tune, Sturek, and Basil (2013) found flipped classrooms to be an effective approach for graduate students in physiology; however, that does not necessarily predict that undergraduate students will have the same success. Experimental time frame is another imperative detail that must be considered when evaluating the efficacy of an educational study. Burrowes (2003) found that active learning increased learning gains in an introductory biology course, but the time frame of study was over the course of the semester. During that time, students could have matured or even learned how to apply information better; thus, there could have been additional factors that influenced learning gains other than active learning. Lastly, it is important to note that numerous active learning studies do not have an experimental design of any sort. Conclusions made from a study without an experimental design may not be trustworthy or transferrable to other classrooms.

The lack of a general consensus regarding the effectiveness of active learning is worrisome, especially considering its current level of popularity. Perhaps most concerning is the paucity of information regarding interactions between specific forms of active learning and particular student characteristics. These are some of the most important factors to consider as younger or less experienced students may not have the foundational knowledge or skills

necessary to effectively apply and manipulate information as required by many active learning methods. This could be especially true within intermediate STEM courses where students are just beginning to get a feel for the domain and might not yet have the skills needed to benefit from a less structured learning activity. For example, Stull & Mayer (2007) found that constructing graphics was too complex and did not result in deep learning for low-knowledge students who were, on average, 18-19 years of age. Results from this study are similar to others such as Kirschner, Sweller, and Clark (2006) who reported that minimal instructional guidance is especially detrimental to students with lower prior knowledge and can lead to misconceptions and disorganized schemata. These inconsistencies within the literature led us to take a fresh look at the utility of active learning by using a very specific form of active learning, case studies, and investigating the benefits it may provide for a specific group of students.

Case Studies

While there are many forms of active learning, case studies are believed to be an effective form for improving cognitive flexibility within ill-structured domains due to increased engagement with the material (Chaplin, 2009; Dewprashad, 2013; Dinan, 2002; Gabel, 1999; Herreid & Schiller, 2013; Herreid, 1994; Herreid, 2006a; Murakami, 2013; Raju & Sankar, 1999; Rybarczyk et al., 2007; Smith & Murphy, 2016; Thistlethwaite et al., 2012). For example, case studies generally include a compelling story that provides a more friendly introduction to concepts as well as interactive features, such as discussion questions, interactive figures or graphics, interactive clickers, concept maps, videos, multimedia, and podcasts that are believed to help students understand complex information (Herreid, 1997; Herreid, 1994; Herreid, 2006a). Case studies have been used as an effective active learning tool within a variety of STEM courses: biology (Baines, Rybarczyk, Thompson, & Wilkins 2004; Rybarczyk et al., 2007),

physiology (Casotti, Beneski, Knabb, 2013), chemistry and biochemistry (Dewprashad, 2012; Hinde & Kovac, 2001; Nair, Shah, T., Seth, Pandit, & Shah, G., 2003), engineering (Raju & Sankar, 1999; Rosson, Carroll, & Rodi, 2004; Russel & McCullouch, 1990), economics and mathematics (Calson & Schodt, 1995; Rasmussen & Kwon, 2007).

Various types of case studies exist, as well as methods for presenting them to students. Case studies can be used in large lectures, small group discussion, whole class discussions, debates, or individually. Large lecture discussions or debates, which are common in many case studies, can become very difficult to manage. Furthermore, large lecture courses have students with a wide variety of academic background interests; some students might excel working with a partner while others might excel working along at their own pace. Case studies are also thought to be effective for students working on their own, at their own pace, as they independently explore concepts. The manner in which the case study is presented, and to whom, often depends on the type of case study (Herreid, 2006a).

The class discussion method is considered the classical case study method which typically involves an instructor that presents a story and then the class is asked to provide feedback; this is very common in the problem-based learning case study approach. Problem-based learning encourages students to research a topic and solve a problem within 2-3 days. For example, students might be presented with a case study and assigned a specific topic to research in order to solve a problem proposed within the case. The next class period would consist of discussing each group member's topic and how it applies to the case.

Another case-based learning method that is similar to the problem-based learning approach is the interrupted case. Interrupted cases are distinguished from problem-based learning because these cases are typically completed in one day rather than across several days.

The interrupted case could involve the instructor introducing the case to the class by asking questions about a topic or a brief overview of the story. After a brief introduction, students are asked to complete the case study individually or within groups. The interrupted case has been thought to be the best-case technique because it prompts students to think like a scientist such that they must develop hypotheses, gather information, and come to logical conclusions (Herreid, 2006a). As such, interrupted case studies were designed and implemented for this research.

Conventional Lectures

Conventional or didactic lectures are sometimes seen as the polar opposite of active learning. Many have described this format of teaching and learning as traditional, as the delivery of information is primarily done by an instructor while students listen and record the information (Bonwell & Eison, 1991; Ebert-May, Brewer, & Allred, 1997; Freeman et al., 2014; McCarthy & Anderson, 1999; Naftalin, 2014; Wolff, Wagner, Poznanski, Schiller, & Santen, 2015). As described above, active learning has been favored over the conventional lecture because the lecture is thought to result in surface level learning (Biggs, 1987; Bonwell & Eison, 1991; Freeman et al., 2014; Wolff et al., 2015). Surface level learning refers to reliance upon temporary learning and rote memorization which promotes the use of information for one single representation or task (Baeten, Kyndt, Struyven, & Dochy, 2010; Case & Marshall, 2004; Perkins, 2006). Students who favor surface level learning approaches tend to store knowledge as individual or discrete packets which are difficult to use and apply in the future as they are often rooted in a certain context or exercise and are not transferable to future scenarios. This type of knowledge is often referred to as inert knowledge (Meyer & Land, 2006; Perkins, 1999).

The conventional lecture approach has been thought to encourage surface-level learning and potentially increase inert knowledge, thus it is viewed as inferior to active learning; however,

others have reported that lectures can provide equal or greater learning value than other approaches (Azer, 2009; Bartsch & Cobern, 2003; Carter, Wesley, & Larson, 2005; Goodwin, Miller, & Cheetham, 1991; Lake, 2001; Lee, Gillan, & Harrison, 1996; Penson, 2012; Schwerdt & Wuppermann, 2011; Smits et al., 2003). For example, Smits et al. (2003) found problem-based and learning from conventional lectures equally effective on student learning gains. In addition, Lee et al. (1996) found that recitation lectures were more beneficial for certain students. Thus, results from other studies challenges the pervasive active learning approach. Due to the inconsistencies within educational literature regarding the best learning approach, the benefits of case studies versus conventional lecturing were examined to unravel what truly constitutes for "active learning" and for whom.

Theoretical Framework

The theoretical framework is the foundation of an educational research project as it guides research questions for transferability and validity. It also provides a framework for systematically investigating a phenomenon and interpreting the results.

This study was grounded in the educational theory known as the Cognitive Load Theory (CLT) which provides guidance for examining the relationship between different modes of content delivery, current levels of intellectual development of learners, and the complexity of the information being presented. Cognitive load can be defined as a multidimensional construct that represents the stress or load on a learner as they attempt to take in and process new information. This load is influenced by many factors such as the learner's intellectual ability, past history with the information, and the effort they are willing to expend to assimilate and accommodate the new information with what they already know (Kalyuga, Chandler, & Sweller, 1998; Paas, Renkl, & Sweller, 2004; Sweller, Ayres, Kalyuga, & Chandler, 2003; Young et al., 2014). The CLT

identifies potential limitations and roadblocks learners may encounter when attempting to learn information for the first time and provides suggestions for how these can be mitigated (Paas et al., 2004). For example, a student with an underdeveloped cognitive architecture, meaning they possess a limited number of interconnected schemata, or chunks of information in their long-term memory that act as a web-like network, is likely going to struggle to comprehend ill-structured information if it is presented at a very rapid pace and foundational issues such as vocabulary and basic concepts are glossed over. The opposite is often true of a learner with a very well-developed cognitive architecture. According to the CLT, the mode of instruction, or way information is presented, can help alleviate some of these issues. In other words, a learner's ability may not always be the limiting factor in the learning process (Brünken, Steinbacher, Plass, & Leutner, 2002; Nesbit & Adesope, 2013; Rhodes & Rozell, 2017; Rhodes & Rozell, 2014; Young et al., 2014).

Components of the Cognitive Load Theory

A substantial part of the CLT involves the investigation of memory systems and how these impact learning. Young and colleagues (2014) described three types of memory systems comprising the CLT: sensory, working, and long-term memory (Figure 2.1). Sensory memory systems involve sensory memory channels such as visual, auditory, touch, and smell. The learner must be actively cognizant or focused on incoming sensory information in order for this information to be incorporated into other memory systems. It is believed that the more sensory modalities or inputs utilized when learning new information, the easier the information will be to learn which reduces the strain on the next tier of memory. For example, listening to a presenter while reading the same words on a slide or in a text allows two channels to provide 'input' into working memory whereas simply reading or listening likely decreases the input due to the loss of

the other available, but not utilized channel. Sensory memory supplies the next tier of memory, which is working memory, with information. Working memory can be described as the temporary information processing center of these memory systems. Working memory is where incoming bits of information are initially interpreted and organized. If the incoming information has not overwhelmed working memory, the information can be incorporated into the next tier of memory, which is long term memory. Long-term memory is comprised of stored schemata that vary in complexity and automation (Van Merriënboer & Sweller, 2010). Schemata result from the long-term construction, addition, and refinement of schema. The more schema a learner possesses, and the more effort is invested in refining these schemas over time, the more information storage and retrieval can occur as their schemata begin to resemble a flexible web instead of a collection of linear, pre-packaged, and ridged mass of pre-figurative schemas. Thus, the number and quality of schemata a learner possesses is important. The more schemata a learner has, the less work or cognitive load is placed on their working memory when learning new information as the information likely has a place to reside or belong in long term memory, in other words, it seems more familiar to the learner. This helps the learner quickly recognize the relevancy of the new information and makes it more likely that the new information can be assimilated and accommodated efficiently with previously learned information. As a result, cognitive flexibility is enhanced (Kalyuga et al., 2003). Memory systems can be managed to process information more efficiently through organizing and connecting schemata in long-term memory (Chandler & Sweller, 1991; Paas et al., 2004; Van Merriënboer & Sweller, 2010; Young et al., 2014). The development of organized schemata is thought to happen through practice and familiarization within a subject (Young et al., 2014).

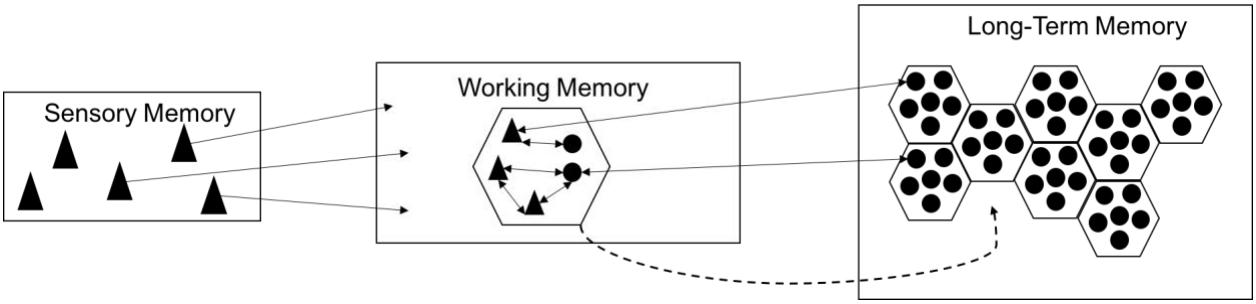

Figure 2.1. Memory Systems described by the Cognitive Load Theory (CLT).
The triangles in the sensory memory box represent sensory information input via visual, auditory, and touch sensory channels. If this information is actively attended to by the learner, it will enter the working memory box where it can be assimilated with prior knowledge (dark circles) that is retrieved from long-term memory. Initial processing of information occurs in the working memory system and can be incorporated into a schema, indicated by the hexagon; each schema is stored into long-term memory.

In addition to memory systems, the CLT describes three types of cognitive load that must be considered when designing and implementing an instructional or pedagogical tool and how these types of cognitive loads can influence the memory systems of the learner and thus ultimately the amount of information taken in, processed, and stored. As described by the CLT, there are three types of cognitive load: intrinsic, extraneous, and germane load, which can work in isolation or in an additive way on learners (Figure 2.2). *Intrinsic load* is described as the inherent complexity or ill-structured nature of a subject (Young et al., 2014). For example, physiology and medicine are ill-structured subjects because there is rarely only one correct answer to any problem and multiple avenues could be pursued that ultimately produce a correct answer (Rhodes & Rozell, 2017; Spiro et al., 1988). To prepare learners for these types of subjects, the natural complexity of these disciplines must be fully displayed and mitigated; a reductionist approach that attempts to simplify a naturally complex concept would be a

disservice to the learner now and in the future as it provides an unrealistic assessment of the field. Thus, intrinsic load should not be altered by instruction. In contrast, extraneous load can - and should be - influenced by instruction such that it is minimized whenever possible (Van Merriënboer & Sweller, 2010). *Extraneous load* is defined as any external factors that are unnecessary or superfluous to the information being presented. If extraneous load is high and requires the learner to devote extra cognitive processing or mental effort to manage this load, less is available to attend to the intrinsic load of the material making it even more difficult to learn (Young et al., 2014). An example of extraneous load could be a Microsoft Powerpoint slide used in a conventional lecture presentation that is filled with words or unnecessary graphics and animations. This can be distracting and negatively impact a student's ability to process and organize relevant information; this is especially true of learners with low prior knowledge or lower levels of motivation to learn the material. The third type of cognitive load is *germane load*. Germane load is referred to as the mental effort put forth by a learner to comprehend the information being presented. Although it seems the learner is primarily in control of germane load, other factors can optimize this load such as reducing extraneous load or scaffolding intrinsic load; both of which would ease the germane load. For instance, learning about feedback mechanisms in physiology requires a strong foundation of many core concepts in physiology and therefore could present an overwhelming intrinsic load to the student, especially if the information is presented in a way that stresses extraneous load. In this instance, the learner could become frustrated or perhaps lose confidence in his/her ability to learn this information, and as such, the amount of effort required by the learner to comprehend the information would have to increase in order to handle the intrinsic load. This task would become even more difficult if the information presented to the student contained superfluous information that is not directly related

to the concept such as how feedback systems often fail during the development and progression of a disease or how a new drug therapy has been developed to help combat this. While interesting, these types of details can negatively impact learning and understanding as they are not directly important for understanding the concept initially. As a result germane load, or the amount of effort the learner would have to put forth just to learn the basic concept, would increase. This is especially true of students with lower prior knowledge or those lacking strong academic backgrounds; the opposite is true for students with stronger academic backgrounds (Graham et al., 2013). Therefore, the CLT encourages instructors to minimize extraneous loads whenever possible, and manage or support intrinsic loads and germane loads.

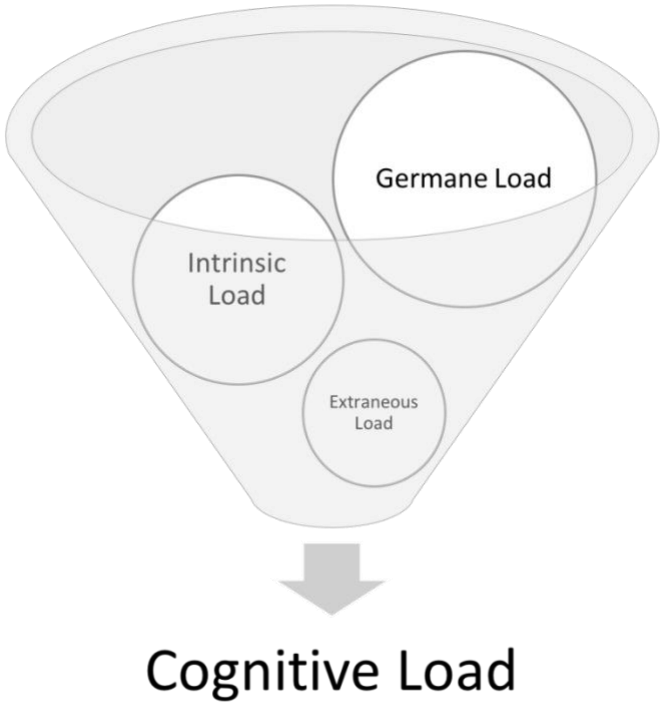

Figure 2.2. **Components of Cognitive Load.**
Extraneous, intrinsic, and germane load are additive as represented in the funnel. Designated by the size of each circle within the funnel, the aim is to keep extraneous load low, manage and support intrinsic load, and optimize germane load.

The Expertise Reversal Effect

Cognitive architecture varies among individuals primarily because of past and present experiences within a domain, which in turn influences the number and quality of schema developed (Kalyuga et al., 2003). The consideration of a learner's cognitive architecture is critical for designing effective instructional tools as the tools and strategies that may help low prior knowledge learners may actually become detrimental for high prior knowledge learners; this is known as the *expertise reversal effect* (Kalyuga et al., 2003; Kalyuga et al., 1998). It is

believed that students with little experience in a domain generally have low prior knowledge within a field have resulting in the development of very few schemata about the domain in their long-term memory system. Others, however, might have had more experience which generally results in a higher prior knowledge level, and consequently more developed, organized and refined schemata. Thus, these experiences and prior knowledge levels impact learning greatly. According to Kalyuga et al. (2003), low prior knowledge learners require more instructional guidance than high prior knowledge learners to compensate for the lack of schemata and reduce stress on working memory systems. Additionally, low prior knowledge learners likely benefit from a reduction in extraneous loads, especially when intrinsic loads are high. Conversely, high prior knowledge learners can tolerate a higher extraneous load as the intrinsic loads for them are often less due to their previous exposure with the discipline and enriched collection of schemata. Additionally, high prior knowledge learners benefit from activities that actually aim to increase the extraneous load as information that is presented too simply or too straightforward or even in a way that could be seen as redundant can detract from their potential.

Cognitive Load Theory Applications in STEM

Assessing and considering prior knowledge levels and schemata development in learners has many implications in biological, physiological, and even medical education (Grunwald & Corsbie-Massay, 2006; Leppink & van den Heuvel, 2015; Van Merriënboer & Sweller, 2010; Young et al., 2014). For example, Eva (2005) states that experts in the medical field, or those with high prior knowledge, can respond to problems through generating ideas rapidly based on recognized patterns. Although novice medical practitioners, or those with low prior knowledge, must use deliberate analytical thinking to solve a problem. Through experience, the number of schemata will increase and memory architecture will develop which results in more efficient

problem analysis (Young et al., 2014). Therefore, the CLT provides a very useful and applicable framework for investigating the effects of different instructional approaches on students, especially within introductory and intermediate STEM fields.

Conceptual Framework

The conceptual framework of a study is an outline of how a research project was conducted in reference to a specific educational theory, in this case, the CLT. Through the lens of this theory, the effects of presenting ill-structured concepts to students in an intermediate physiology course using two different instructional approaches were examined. To accomplish this, two pedagogical tools were created with different levels and types of cognitive loads: an active learning activity in the form of a case study and a conventional lecture. Each form of instruction was designed to deliver identical information but used a different format.

Interrupted case studies were designed to scaffold the information for students by using interactive charts and diagrams as well as an engaging story that asked students to examine ill-structured concepts from multiple angles with minimal to no guidance. Information with a high intrinsic load was presented with a high extraneous load in the form of case studies. The case studies required students to interact with graphics, problem solve, and transfer information through the use of a story. However, the conventional lecture was designed to present the same information without the use of a story or interactive graphics. The lecture was designed to present the same ill-structured information with a high intrinsic load in such a way that reduced the extraneous load.

Limitations and Delimitations

Many studies have limitations that are inevitable when working with voluntary students in a real-life classroom setting. Limitations within educational research, and other areas of research, are beyond what can be controlled by the researcher. This includes student characteristics such as prior knowledge, approaches to learning, academic preparation, age, interests, goals, home life, or mental and physical health. Other limitations include time allotment for the study and timing within the semester. Timing of study can be particularly important if students have academic or personal issues that may affect their performance.

The delimitations include the aspects that can be controlled in the study such as student population, experimental design, and research setting. To control for these, this study was conducted at one institution using a single course. Furthermore, while there are many forms of active learning, we investigated the effects of active learning using only case studies.

Participants

Participants were recruited from an undergraduate physiology course comprised of sophomores, juniors, and seniors with very diverse majors, career goals, and academic backgrounds at a large Midwestern university. Prerequisites for this course included chemistry I and II, Biology I (with a B or better), 3.0 GPA, and a sophomore standing (in academic credit hours). Course content was in-depth with all major physiological systems presented and was designed to prepare students for professional exams as well as their first year of professional school.

All activities were approved by the Institutional Review Board (IRB) of the university. Students were asked to sign an informed consent form presented to them on the first day of class, and then elements of informed consent were repeated verbally to students on the day of the study. Students were offered minimal extra credit for participating in the study. An alternative assignment was offered if any student did not want to participate in the study.

Methodology I: Insulin Resistance and Type II Diabetes Case Study

A mixed methods approach was used to examine the utility of active learning vs. a more traditional format within an undergraduate physiology course as well as the potential relationships that might exist between these formats of learning and individual student characteristics. The following research questions were investigated:

1. When presenting an ill-structured concept, is there a difference in overall student learning gains between using an active learning tool such as a case study compared to a conventional lecture?
2. Is there a relationship between a student's approach to learning, prior knowledge levels, ACT scores or credit hours completed and the amount learned from an active learning tool such as a case study compared to a conventional lecture?

Active Learning Tool Selection and Case Study Development

Before the study began, an active learning tool in the form of a case study was developed. The first step in this developmental process was the selection of a topic that aligned with concepts from the dairy cow exercise study and core physiological concepts known to be difficult, yet critical, for students to learn in introductory and intermediate physiology courses for success in future courses (Michael et al., 2017). The topics chosen included insulin resistance and type II diabetes. Insulin resistance and type II diabetes are multifaceted because they can vary in causation, side effects, and treatment. Many factors can contribute to the onset of insulin resistance and type II diabetes, including genetic predisposition, eating habits, lack of exercise, age, race or ethnicity. The consequences of insulin resistance and type II diabetes can lead to systemic complications such as diabetic neuropathy, retinopathy, cardiovascular disease, and

endocrinopathies. Treatment methods may include exercise, diet, surgery, an assortment of drug therapies, and even amputation of body parts. These diseases are considered to be ill-structured and can be difficult for students to learn because of the conceptual application, various avenues that can lead to the development of insulin resistance and type II diabetes, and positive feedforward effects that follow. Students must have a firm understanding of glucose regulation, which begins with a solid understanding of the core concepts identified above, to recognize the many pathways involved in insulin resistance and type II diabetes; as such, these concepts often present a high intrinsic load.

To support the high intrinsic load while mitigating extraneous load, a case study was designed that presented the effects of poor diet choices and lack of exercise on glucose regulation. High glucose diets can result in various physiological responses that disrupt blood glucose homeostasis. Responses could include: down regulation of insulin receptors within insulin-dependent cells, failure to complete the second messenger pathway that is necessary for GLUT4 channel proteins to translocate to cell membranes, and eventually could lead to cell death of pancreatic beta cells. The intricacies of these diseases cannot be memorized. Approaching these problems requires a flexible network of schemata to retrieve and connect knowledge.

The non-linear approach students must take to understand ill-structured concepts like insulin resistance and type II diabetes can be uncomfortable and frustrating, especially if students have created a habit of surface level learning approaches (Rhodes & Rozell, 2017). However, non-linear or deep learning approaches are vital to solve ill-structured problems within fields that many students in physiology are interested in pursuing, like human medicine. There are other homeostatically regulated variables in the body that are ill-structured concepts and present a high

intrinsic load such as thermoregulation, ion levels in the blood, or hormone production and release. However, insulin resistance and type II diabetes were selected topics because they are current and becoming an epidemic in the U.S. that many students can relate to on a personal level (Zimmet, Alberti, & Shaw, 2001).

Design-Based Research: Insulin Resistance and Type II Diabetes Case Study Development

Data were collected and analyzed during the Spring of 2017 using a finalized version of the case study that required several semesters and iterations. Design-based research (DBR) was used to test and refine the instructional tool (Figure 2.3) in order to improve its initial design and even subsequent iterations (Cobb, Confrey, DiSessa, Lehrer, & Schauble, 2003). This approach allows the researcher to conduct developmental studies using a research team or outside reviewers to document common themes and refine each iteration (Cobb et al., 2003). For this study, an initial iteration of the case study was designed and then refined using the DBR process which occurred during two 16-week regular academic semesters and one 8-week summer semester. Each iteration of the case study was edited and refined using feedback from students, peer reviewers and even outside reviewers.

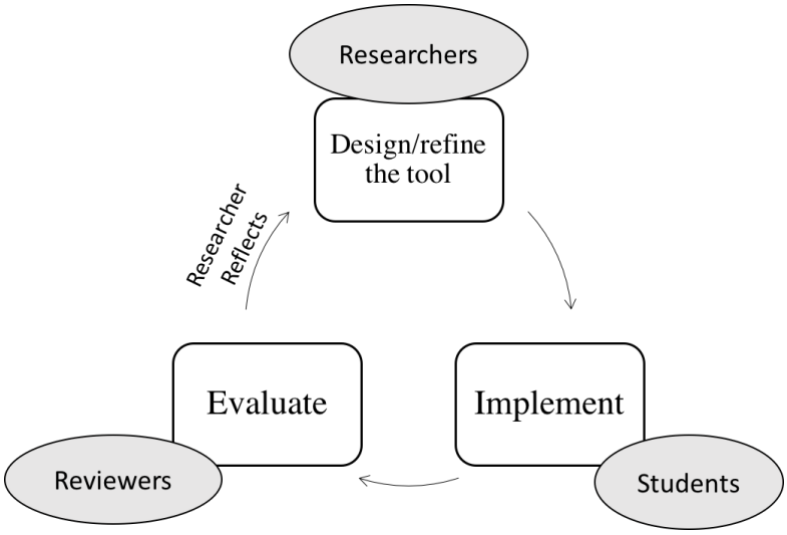

Figure 2.3. **The design-based research process.**

This approach was used to develop the case study which was implemented in a real classroom setting, and evaluated by outside reviewers. After each evaluation, emerging themes were reflected upon and used to refine the case study before reusing.

Four iterations of the case study were implemented into an undergraduate physiology course across four semesters (Figure 2.4). Each iteration was evaluated by outside reviewers. The first iteration of the case study was evaluated by five reviewers selected by the researchers, including undergraduate physiology instructors (n = 4) and a medical practitioner (n = 1). The second and third case study iterations were peer-reviewed by three reviewers selected by NCCSTS; and finally, the fourth iteration of the case study was published by the NCCSTS (http://sciencecases.lib.buffalo.edu/cs/collection/detail.asp?case_id=917&id=917). The peer review process through NCCSTS was incorporated into the design-based research process such that all comments were documented, and emerging themes were identified throughout the refinement of the case study. A summary of emerging themes can be found in Figure 2.5.

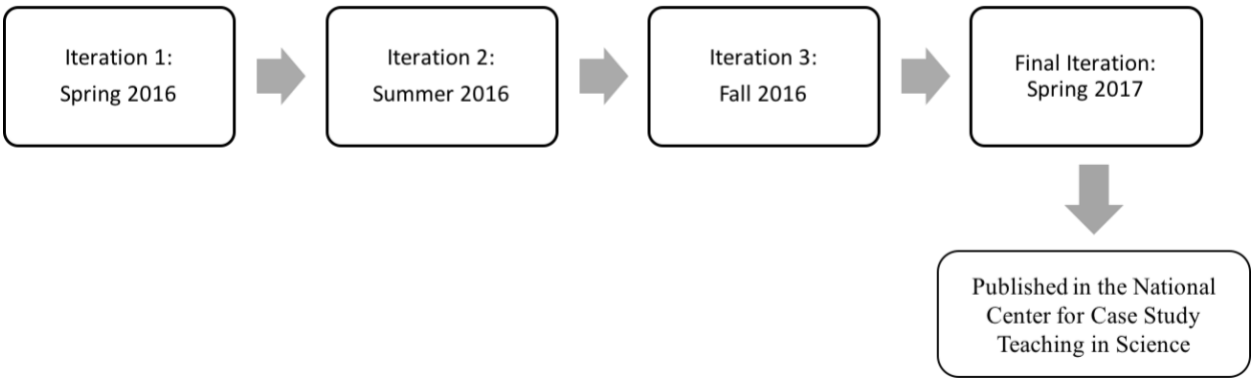

Figure 2.4. Case Study Iteration Timeline.
Each iteration was implemented into an intermediate physiology course during the semesters listed by academic semester and year.

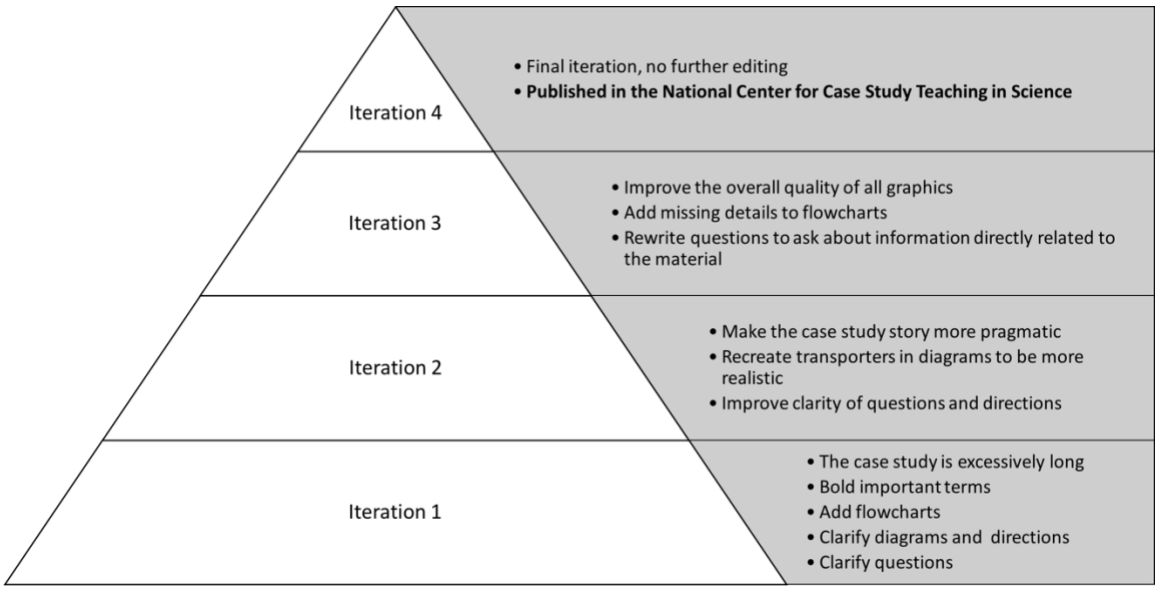

Figure 2.5. **Emerging themes from each case study iteration.**

The case study was tested and revised three times via the design-based research process. Each iteration was refined to improve the clarity of information presented, questions asked, and directions for completing the interactive graphics and diagrams within the case study. The themes listed on the right of the figure were addressed to create the subsequent iteration. Preliminary data were collected and analyzed from iterations 1-3. Data collected from iteration 4 was used for publication because this was the final and published version of the case study.

Case Study Iterations

Specific changes made in each iteration as suggested by reviewers are shown in Figure 2.7.

Iteration 1

The first case study iteration was designed to introduce in-depth information regarding blood glucose regulation and how the pathways involved can be disrupted with insulin resistance and type II diabetes. This case study was given to students enrolled in a 16-week physiology course during the spring 2016 semester. Preliminary data were collected and

analyzed from this iteration but were not used for manuscript publication because the case study was not published at this time. However, student answers, comments, and misconceptions were documented. After using the case study with students, it was submitted to five selected reviewers to critique and edit. These reviewers included college science instructors and a trained medical practitioner. All comments were transcribed, and emerging themes were identified and documented. Specific changes to iteration 1 (Figure 2.7) were used to implement the second iteration in the following semester.

Iteration 2

The second iteration was administered during the summer of 2016. Preliminary data were collected and analyzed as a part of the design-based research process. This iteration was submitted to the National Center for Case Study Teaching in Science (NCCSTS), sponsored by the National Science Foundation (NSF), for publishing. This iteration was then critiqued by three reviewers chosen by NCCSTS. After receiving feedback from these reviewers, emerging themes were identified and addressed. Specific changes to iteration 2 (Figure 2.7) were used to implement the third iteration in the following semester. The third iteration was resubmitted to NCCSTS.

Iteration 3

The third iteration was implemented in the fall of 2016 and preliminary data were collected and analyzed. Once feedback was received from the NCCSTS reviewers, the diagrams, questions, and authenticity of the story of this iteration were addressed. Additions made to this iteration tremendously improved quality and clarity of the physiological mechanisms occurring within the diagrams. Questions were rewritten to more directly ask about information presented within the case study, rather than asking about information that may have required students to

have a specific baseline of prior knowledge. Directions to complete diagrams were simplified to reduce the amount of labeling. Specific changes to iteration 3 (Figure 2.7) were used to implement the fourth and final iteration in the following semester. Revisions were resubmitted to the NCCSTS (the third submission of the case study to NCCSTS).

Iteration 4

After receiving feedback from NCCSTS reviewers, a box was added to one of the flowcharts so that all steps presented in the case study regarding insulin signaling and glucose uptake in a skeletal muscle cell could be included. Additional grammatical changes were also made. This final iteration was implemented in the spring 2017 semester. Data were collected and analyzed for publication. There was no further editing beyond this iteration. The case study was published in the NCCSTS. An example of a graphic from the final iteration can be found in Figure 2.6

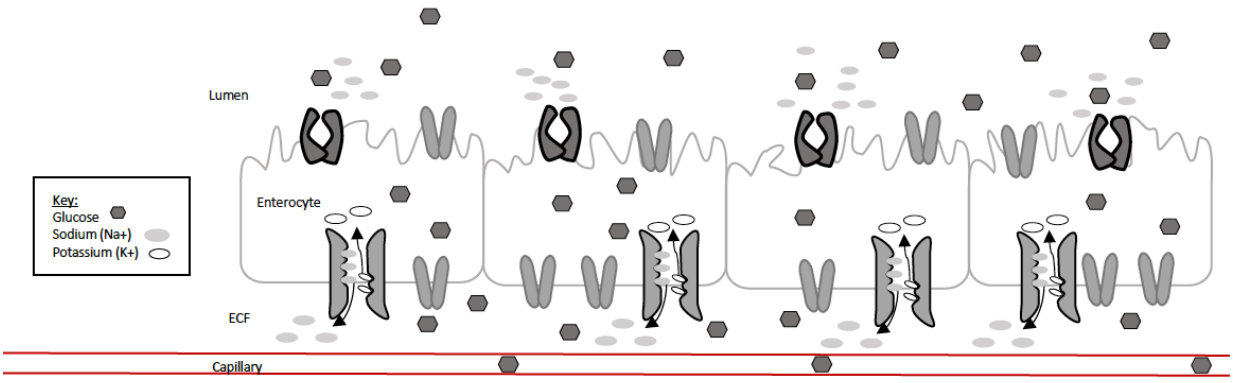

Figure 2.6. **An example of an interactive graphic from the case study.**
This graphic displays glucose absorption through enterocytes located within the jejunum of the small intestine and asks students to label transporters and the direction in which molecules are moving. Students in the case study group were asked to label the transporters and movement of molecules. This graphic was also presented to students in the conventional lecture group, but the transporters and direction in which molecules are flowing were already labeled.

Iteration 1 *Spring 2016*	**Iteration 2** *Summer 2016*	**Iteration 3** *Fall 2016*
• Using a tablet for graphic design, figures were recreated for more accurate representations of transporters and pumps within the enterocytes of the jejunum and a skeletal muscle cell • Information and questions regarding glucose uptake into pancreatic beta cells and insulin release were removed • Terms deemed as important were bolded and flowcharts were added • Preliminary data were collected and analyzed.	• Proteins found within the enterocytes of the jejunum (SGLT-1) and within the skeletal muscle cell (GLUT4) were reconstructed to appear as transporters rather than channels • GLUT4 proteins were relocated from inside of the storage vesicles to within the plasma membrane of the vesicle • Questions that were deemed irrelevant or unclear were revised to ask questions directly related to the case study • The timeline of the onset of type II diabetes was adjusted from 4 years to 25 years. • Preliminary data were collected and analyzed.	• Blank boxes were added to flowcharts that were used for students to interact with and visualize steps of physiological processes • Diagrams that were hand drawn via a tablet were recreated in PowerPoint to be more precise and accurate • GLUT2 and SGLT-1 proteins within enterocytes were recreated to more accurately represent their shapes • ATPase pumps were removed from the apical membrane of enterocytes • Preliminary data were collected and analyzed.

Figure 2.7. Specific changes in each iteration of the case study.

The chart above lists specific changes made to each iteration of the case study. Beneath the iteration number at the top of the chart lists the semester in which that iteration was implemented in italics. Preliminary data were collected from iterations 1-3. The fourth iteration is not listed as there were no further changes made to this final iteration.

Conventional Lecture Iterations

The conventional lecture was designed to present the same material found within the case study but without the inclusion of a story or interactive graphics. The purpose of the lecture treatment was to deliver information in a way that minimized extraneous load while maintaining the same intrinsic load. As with the case study treatment, iterations of the conventional lecture were refined using the design-based research process. Furthermore, the editing process used to

refine the conventional lecture treatment mirrored that used to refine the case study; for example, any information that was removed or changed in the case study was also changed in the lecture so that these two treatments presented information as identically as possible with the exception of the format and type of cognitive load associated with each format.

Pre-class Assessments

After the development of the active learning tool in the form of a case study and the conventional lecture, pre-class assessments were used to assess students' current levels of experience with core physiological concepts and their preferred method for learning new information. There were three assessments given prior to treatment administration including a prior knowledge assessment, a student approach to learning assessment, and a pretest.

Prior Knowledge Assessment

A Prior Knowledge Assessment (PKA) was administered to all students on the first day of class as a measurement of background knowledge regarding key physiological concepts (Appendix Table B.2). The PKA consisted of 10 questions that were chosen after several rounds of editing to represent major physiological concepts taught throughout the semester. For each question on the PKA, multiple answers could have been correct but to receive the full point for each question students had to have selected all the correct answers and none of the incorrect answers. This grading procedure was used to reduce guessing. Results from the PKA were used as a baseline to evaluate students' levels of prior knowledge regarding complex and ill-structured concepts in physiology.

Student Approach to Learning

A measure used to evaluate student learning strategies is the Student Approach to Learning (SAL), a validated metric that is used to assess students' cognitive learning strategies,

motivation, self-beliefs, and learning preferences (Marsh, Artelt, & Peschar, 2006). Previously the SAL survey has been used to evaluate student learning perspectives and career aspirations (Nagengast & Marsh, 2012). The measurement has also been used to evaluate how students choose to learn information (Rhodes & Rozell, 2017). Although the SAL is used to evaluate many aspects of learning, only questions from the SAL that evaluated students' self-regulated learning strategies were selected (Appendix Table B.3). Self-regulated learning strategies are defined as learning preferences or predispositions a learner has for amassing new information; such as, memorization and elaboration strategies. Using a Likert-type scale, students were asked to select a numerical value ranging from 1 to 5 to determine whether they strongly disagreed, by selecting the number 1, or strongly agreed, by selecting the number 5, with a statement. Eight questions were selected to evaluate the propensity of a student to memorize (4 questions) and elaborate (4 questions) on the SAL.

Results from the SAL survey were used to develop an individual SAL metric for each student as previously described by Rhodes and Rozell (2017). This metric was used to determine whether relationships existed between a student's approach to learning, prior knowledge levels, and performance on post-tests related to the topics covered in the case studies. To determine the SAL metric for each student, the following formula was used:

SAL Metric = (Elaboration Sum) – (Memorization Sum)

This formula resulted in a number, ranging from -16 to +16, indicating whether a student had a greater inclination to memorize or elaborate, respectively. The lowest number a student could select per question on the SAL was 1, thus the SAL metric was calculated using the following formula: [(5 X 4) – 4]. A student with a score of -16 would indicate a very strong inclination to memorize, and a student with a score of +16 would indicate a very strong inclination to

elaborate. The metric was used to assign one value to each student to prevent from counting a student twice, as a memorizer and an elaborator, in data analysis. The metric ranged from -14 to +14 in the data set used for publication.

Pretest/Posttest

The pretest was designed to ask questions about the information specifically contained within the case study including glucose regulation, insulin resistance, and type II diabetes. In comparison to the PKA, which assessed student understanding of many core concepts in physiology, the pretest focused on information unique to the case study. The purpose of the pretest was to gain a better understanding of how familiar students were with these specific concepts to observe learning gains from the case study and conventional lecture. The pretest was reviewed by the authors of the case study and revised with each iteration of the case study (4 iterations). Therefore, the pretest was also a part of the design-based research process to ensure accuracy. There were two main categories of questions on the pretest: basic and applied/transfer questions. Basic questions were defined as those that required students to simply retrieve information, such as asking about the name of a specific transporter located on an enterocyte within the jejunum. Applied/transfer questions were defined as those that asked students to apply information to a situation, such as asking about a type of drug that may slow the uptake of glucose. The pretest, which was given to all participants before treatments were administered, consisted of 10 questions that were a combination of basic and applied/transfer questions (Appendix Table B.4). To receive the full point for each question every correct answer had to be selected and none of the incorrect answers selected. This all-or-none grading scheme was used to reduce the chance of receiving full credit due to guessing. The posttest was identical to the pretest and given directly after students received each treatment. To test the validity of the

posttest and determine the difficulty of each question, an item difficulty index was used as described by Tavakol and Dennick (2011). This index evaluates the total number of students who answered each question correctly to determine the quality of each question. The formula used to determine this index was:

$$P = R/n$$

Where P = difficulty index of the item, R = number of correct answers to item, and n = total number of students. Item difficulty index scores have a range of 0 to 1. As described by Tavakol and Dennick (2011), questions with an item difficulty index score between 0.3 and 0.8 are considered to be reasonable questions, or questions with the appropriate amount of difficulty for students to answer. Questions with item difficulty index scores outside of this range are considered too easy (approaching 1.0) or too difficult (approaching 0). Results of the item difficulty index for the posttests are shown in Table 2.2. All IDI scores fell within the acceptable range, with the exception of question 10 which fell below the recommended IDI range.

Table 2.2. **Item difficulty index (IDI) analysis.**

Using the IDI equation, the posttest answers to each question within the case study and conventional lecture treatments were examined to determine the difficulty of each question. IDI scores range between 0 and 1. Bolded numbers indicate IDI scores that were below 0.3 and thus not reasonable questions.

	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Q10
Case Study	0.60	0.48	0.40	0.53	0.65	0.68	0.68	0.42	0.61	**0.06**
Conventional Lecture	0.75	0.32	0.30	0.55	0.65	0.57	0.77	0.55	0.47	**0.07**

Experimental Design

Once all components needed to conduct the study had been refined through the design-based research process across iterations 1-3, they were used in the official round of data collection and analysis which was used for publication. The pretest/posttest comparison group design, as described by Campbell & Stanley (1963), was used to investigate the utility of a case study versus a conventional lecture when presenting ill-structured concepts to students in an undergraduate physiology course (Figure 2.8). This is considered one of the most robust experimental designs in educational research because it controls for many internal threats to validity (Figure 2.9).

R1	O	X_1	O
R2	O	X_2	O

Figure 2.8. **Pretest/Posttest Control Group Design.**

The R represents the random assignment of students into treatments. randomly assigned groups. The O represents the pretest and posttest given before and after treatments. The X_1 represents

the case study treatment, and the X_2 represents the conventional lecture treatment. The R1 is the case study group and R2 is the conventional lecture group.

History	Maturation	Testing	Instrumentation	Regression	Selection	Mortality	Interaction of selection, maturation, etc.
+	+	+	+	+	+	+	+

Figure 2.9. Internal Threats to Validity.

Threats to internal validity can weaken the meaning or value of the data if not controlled through randomization, control groups, and the data collection time frame. The pretest/posttest control group design eliminates these threats to validity.

Using a convenience sampling method, 122 students from an undergraduate physiology course completed all components of the study (Figure 2.10). Because the study included multiple components that needed to be completed in class, various parts of the study were administered over a range of several months to prevent participant fatigue. The PKA and SAL were given on the first day of class while the pretest measuring the background knowledge of students for the ill-structured concepts presented within each comparison treatment was given approximately 6 weeks later, 28 February of 2017. Participants were given the case study or lecture treatment on either 1 or 2 March of 2017 during their lab times.

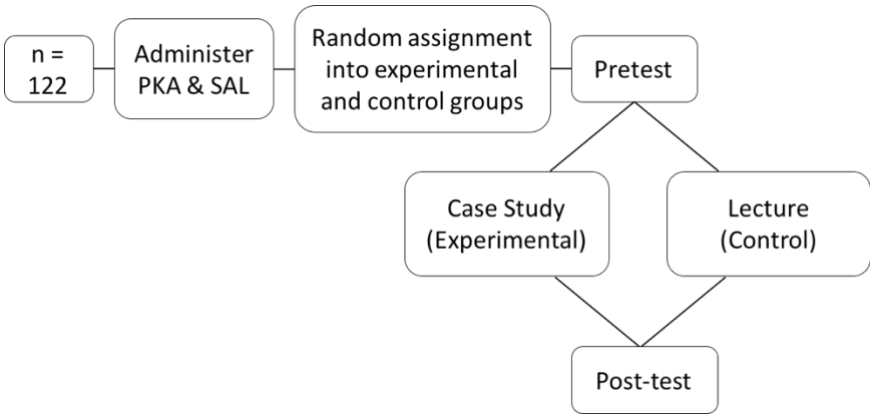

Figure 2.10. **Experimental Design of the Insulin Resistance and Type II Diabetes Case Study.**

Components of a pretest and posttest true experimental comparison group design were used in this study. The PKA and SAL were given on the first day of class, then a topic-specific pretest was given right before randomizing participants into either the case study or conventional lecture treatment. A posttest was given immediately after each treatment.

Administration and Randomization of Students into Treatments

The course used for this study consisted of a lecture and four lab sections. Participants were randomized within each lab section using a random number generator in Microsoft Excel (Microsoft Corporation, Redmond, VA). Students were given a number and randomly placed into treatment groups: case study or conventional lecture. Once students were separated, they were administered their treatments at the same time in different classrooms. Students in the case study treatment took between 60-90min to complete the case study and posttest. It took approximately 30-40min to deliver the conventional lecture and for students to complete the posttest.

Academic Readiness Assessments

After collecting the pretest and posttest data, academic readiness assessment data were collected. Two outside components were used for data comparison and analysis including ACT scores and academic credit hours completed.

ACT College Readiness Assessment

The ACT is a standardized college entrance exam given to high school students in the U.S. and Canada to assess college readiness and is often used as a prerequisite when applying to American colleges and universities. After gaining permission from student participants, ACT scores were collected from the university's online information system.

Academic Credit Hours

Academic credit hours were used as a measure of academic maturity. The total number of undergraduate academic credit hours completed by each student, including transfer credits, were collected from the university's online system. These data were used to examine whether academic experience affected student learning from a case study or conventional lecture.

Qualitative Methods

The qualitative component of this mixed-methods study was included to gather student opinions about the insulin resistance and type II diabetes case study specifically, as well as the overall use of case studies as part of active learning in a more general sense. Qualitative research involves identifying thoughts or feelings that have been constructed by someone through an experience with the goal to understand a meaning or process (Merriam & Tisdell, 2015). Qualitative research is usually conducted through observations or interviews. Data collection and analysis within qualitative research is inductive, rather than deductive, and the researcher extrapolates meaning from the data (Merriam & Tisdell, 2015).

For the purposes of this study, semi-structured interviews were used as a qualitative research approach. This type of interview allows the researchers to use a list of broad questions that promotes detailed and in-depth answers. After the questions were written, they were reviewed and approved by the IRB. Each interview question had a follow up question, such that if the student answered yes or no to a question, there was a follow up by asking why. This approach allowed for the collection of additional information from each interviewee about his/her feelings towards the case study. The following features specific to the case study were investigated: the story line, flow charts, interactive graphics, questions, or bolded words. How students preferred to learn this information was also explored, whether that be from a case study, lecture, YouTube video, or multimedia.

To carry out these interviews, fourteen students from the case study treatment volunteered to participate in a brief interview the week following the case study experiment. To randomly select volunteers, every fifth student who completed and submitted the case study and posttest was asked to voluntarily participate in an interview. After voluntary consent was

attained and the random sampling method completed, students were emailed to schedule individual interviews and a total of ten students agreed to participate. Each interview consisted of 9 open ended questions (Appendix Table B.5). All interviews were video recorded and transcribed using oTranscribe (http://otranscribe.com/). If more than 50% of students had a similar answers or comments, those were identified as emerging themes. The following research questions were investigated:

1. Did the students who received the case study find it to be helpful? Why or why not?
2. Would the students who received the case study prefer another format of learning this information? If so, what format would they suggest?

Methodology II: Lactate Threshold Case Study

Another case study about the concept of lactate threshold was developed to validate the results of the insulin resistance and type II diabetes case study. This case study was implemented using another population of students within the same course, but during a different semester as this case study was given to students during the fall 2017 semester. The case study was then published in the National Center for Case Study Teaching in Science (NCCSTS) in spring semester of 2018 (Rhodes, Rozell, Wilson, 2018). The lactate threshold case study was used to confirm the correlation results from the insulin resistance and type II diabetes case study. Only one research question was investigated:

1. Is there a relationship between learning gains from the use of a case study and student characteristics including prior knowledge levels, approaches to learning, ACT scores, or college credit hours completed?

Topic Selection

The lactate threshold case study topic included information regarding the production of lactate within eukaryotic cells, specifically skeletal muscle cells, as well as lactic acid production, the difference between these two cellular products, how these products are used, and specifics about the accumulation of lactate in the blood and its relationship to lactate threshold. This case study also incorporated the top five core concepts in physiology (Michael et al., 2017) through the production and utilization of lactate within cells and lactate shuttling. Information within this case study could also pose a high intrinsic load on students because the fate of lactate varies depending on the needs of the cell or other cells within the body.

Design-Based Research

The lactate threshold (LT) case study was designed using the design-based research process (Figure 2.3). The first case study iteration was designed in the spring 2017 and implemented at the University of Buffalo case study teaching conference in May of 2017. Emerging themes from the first iteration were identified and addressed to create the second iteration, which was submitted and reviewed by the NCCSTS. After reviewers from NCCSTS revised on the second iteration, the third and final iteration was implemented into an intermediate physiology course at a large Midwestern university in the fall 2017 semester and accepted for publication in the NCCSTS.

Experimental Design

A one-group pretest, posttest experimental design (Campbell & Stanley, 1963) was used to assess relationships between learning gains from a case study and student characteristics (Figure 2.11). Using a convenience sampling method, 120 participants completed both the pretest and posttest over the case study. The Prior Knowledge Assessment (PKA) and Student Approach to Learning (SAL) survey were given on the first day of class. Students completed the case study, including the pretest and posttest, during their lab time approximately 10 weeks later. Participants completed the pretest in class on a Thursday, 26 October of 2017, and then completed the case study and posttest on either Monday, 30 October of 2017, or Tuesday, 13 October of 2017 depending on the participant's lab time. Immediately after completing the case study, participants completed the posttest.

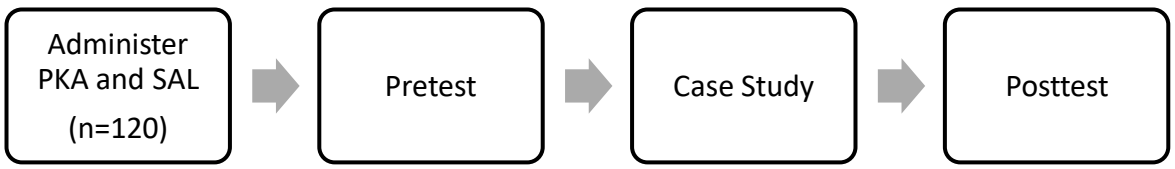

Figure 2.11. **Experimental Design of the Lactate Threshold Case Study.**
Participants completed a Prior Knowledge Assessment (PKA) and Student Approach to Learning (SAL) survey on the first day of the semester. Approximately ten weeks later, participants completed a pretest, the LT case study, and then the posttest.

Lactate Threshold Case Study Components

The pretest used for this case study, which went through three rounds of review and consisted of 10 multiple choice questions that could have multiple answers correct for each question, was presented to all students prior to administration of the case study (Appendix Table B.6). The day after the pretest was given, participants were asked to do a wall sit around the classroom for as long as they could. The wall sit was used as a form of experiential learning, prompting students to consider what causes muscle soreness. Right before the case study was administered, as a method of the interrupted case, students were asked if they were sore from the wall sit and why they think some people were sore and some people were not. Without providing students with the answer to the cause of muscle soreness, the case study was administered to all participants and the posttest, which was identical to the pretest, was given immediately after completing the case study.

With permission from participants, the PKA, SAL, ACT score, and academic credit hours completed were collected as described in the first methods section and used for correlations with learning gains. To verify the correlation between academic credit hours completed and learning gains, a t-test was carried out to compare learning gains between students who had more or less credit hours, defined as above or below the average credit hours completed.

Results I: Insulin Resistance and Type II Diabetes Case Study

RQ1: When presenting an ill-structured concept, is there a difference in overall student learning gains between using an active learning tool such as a case study compared to a conventional lecture?

Results from pretest, posttest, and learning gains are shown in Figure 2.12. There were no differences in pretest scores between the case study (n=62, M=1.84, SD=1.26) and conventional lecture (n=60, M=1.82, SD=1.39) treatments. Posttest scores were not different between the case study (n=62, M=5.11, SD=2.43) and conventional lecture (n=60, M=4.98, SD=2.15) treatments. Lastly, there were no differences in learning gains (posttest-pretest score) between the case study (n=62, M=3.27, SD=2.66) and conventional lecture (n=60, M=3.17, SD=2.20) treatments. To confirm learning gain results, a normalized learning gain equation was used as described by Hake (1998), where (posttest-prestest)/(10-pretest). The normalized gain equation allows learning gains to be compared between different instructional approaches, like the case study and conventional lecture, by comparing the ratio between actual learning gains: maximum learning gains. The average normalized learning gain for the case study treatment was 0.39 and 0.38 for the conventional lecture treatment with a p-value of 0.914, this verified that learning gains were not different between treatments.

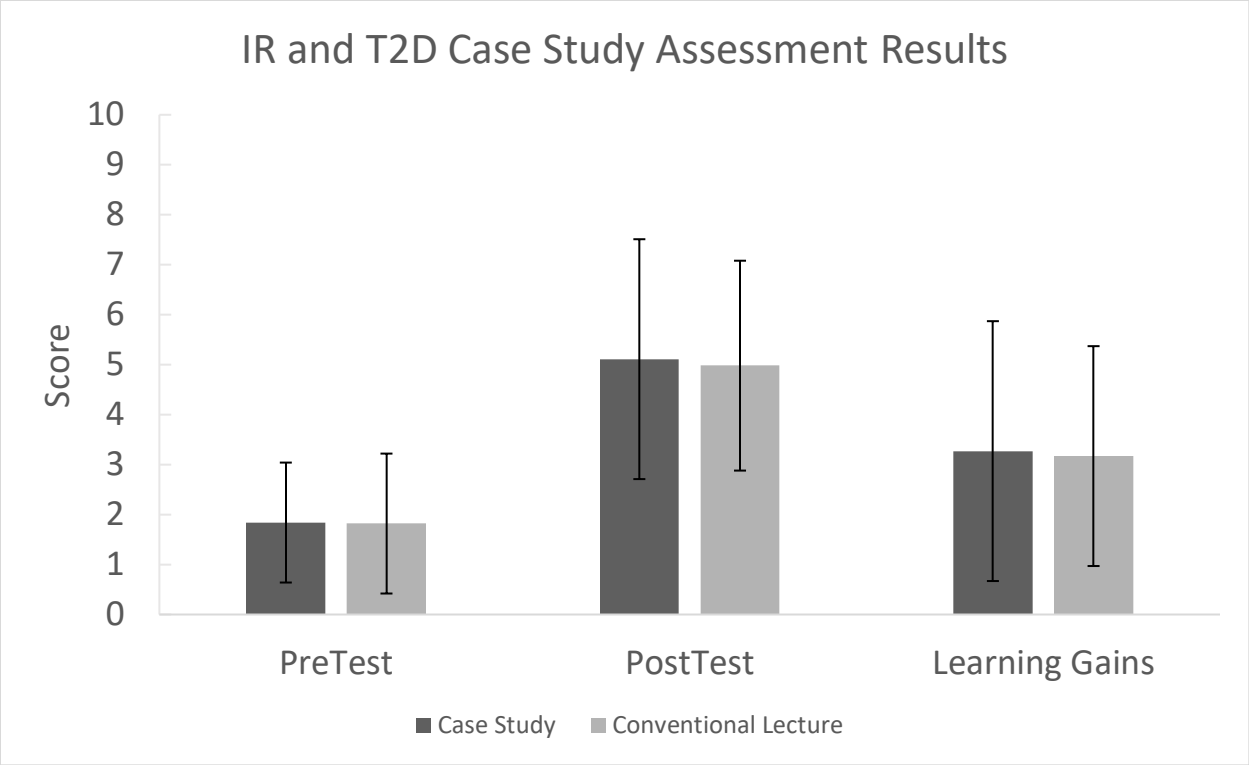

Figure 2.12. **Assessment Results.**

The pretest, posttest, and learning gains for the case study and conventional lecture treatments are shown on the y-axis. There were 10 questions on the pretest and posttest, each worth one point. To receive full credit for each question on the pretest and posttest, every correct answer had to be selected and none of the incorrect answers selected. This was an all or nothing grading scheme. Data are presented as mean +/- SD.

RQ2: Is there a relationship between a student's approach to learning, prior knowledge levels, ACT scores or credit hours completed and the amount learned from an active learning tool such as a case study?

Relationships between student characteristics and learning gains between treatments were assessed by examining correlations. There were positive and significant relationships between learning gains from a case study and prior knowledge (PKA), r(58)=.261, *p*=.046; approach to learning (SAL metric), r(58)=.270, *p*=.039; ACT score, r(51)=.370, *p*=.007; and academic credit hours completed, r(57)=.271, *p*=.039 (Figure 2.14, Figure 2.15, Figure 2.16, Figure 2.16, respectively). Alternatively, there was no statistical relationship between student learning gains from the conventional lecture treatment and PKA, r(59)=.021, *p*=.872; SAL metric, r(57)=-.039, *p*=.769; nor academic credit hours completed, r(57)=.011, *p*=.936 (Figure 2.14, Figure 2.14, Figure 2.16, respectively). However, there was a positive and significant relationship between student learning gains from the conventional lecture and ACT score, r(54)=. 303, *p*=.025 (Figure 2.15).

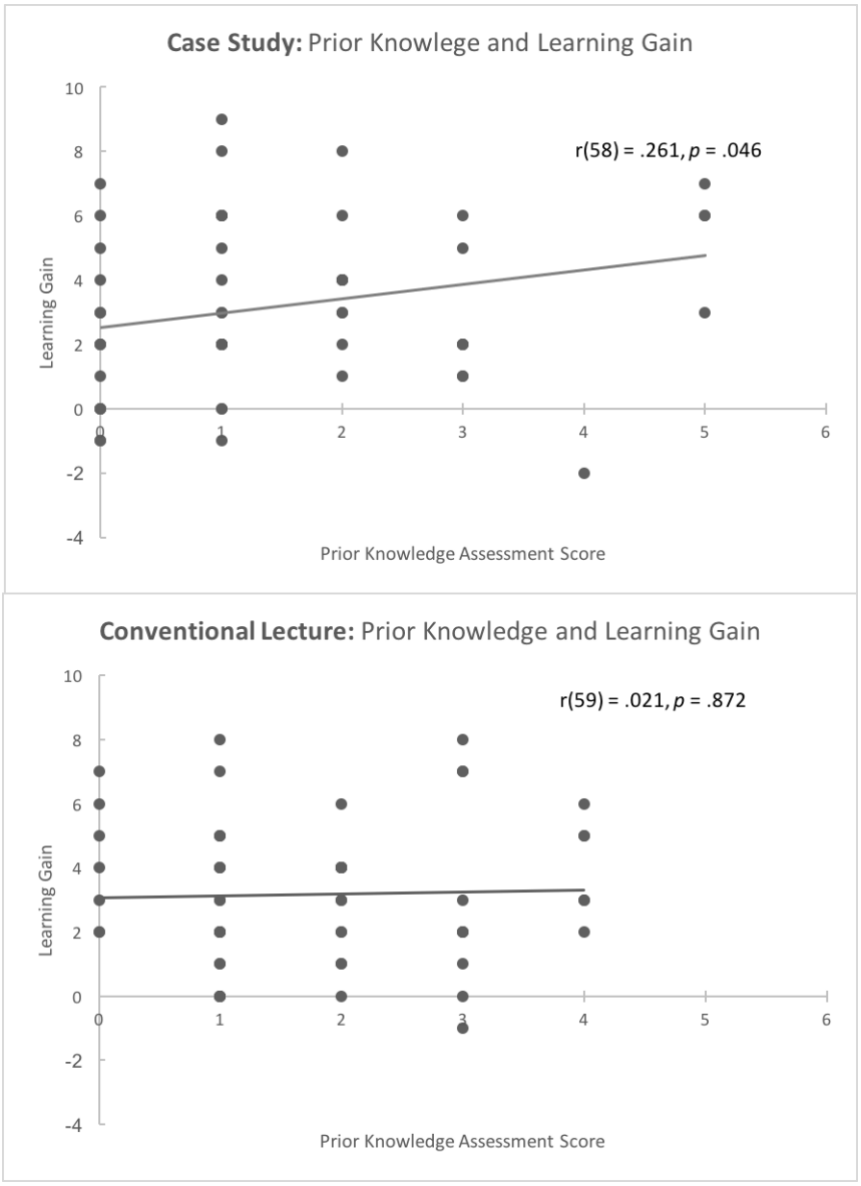

Figure 2.13. **Correlations between Prior Knowledge Assessment (PKA) Score and Learning Gains.**

Case study and conventional lecture treatment data are presented individually. The PKA was graded using an all-or-none grading scheme to eliminate guessing and thus resulted in whole numbers from 0 to 10. The PKA scores in this dataset ranged from 0 to 6. Discrete variables were used and thus each data point may represent the PKA and learning gain score for multiple students. Learning gains are displayed on the x-axis. Some learning gain scores were negative,

which represents that some students performed worse on the posttest than the pretest. Outliers were removed from the case study treatment and thus *n* was reduced.

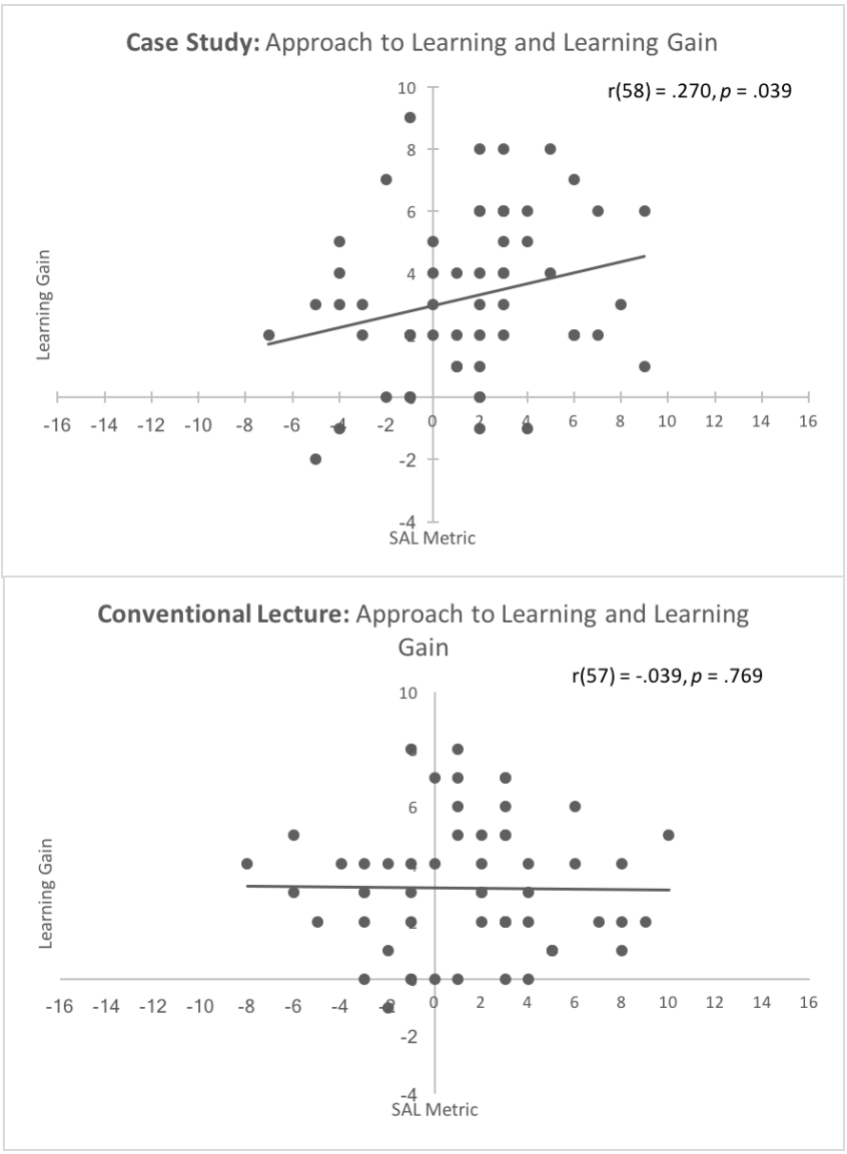

Figure 2.14. **Student Approach to Learning (SAL) metric and Learning Gains Correlations** Case study and conventional lecture treatment data are presented individually. The SAL metric, designed by Rhodes and Rozell (2017), ranges from -16 to +16 and was calculated by subtracting student's memorization score from their elaboration score. An SAL metric score of -16 indicates a student has a strong preference to memorize information, and a score of +16 indicates a student

has a strong preference to elaborate on information. The SAL metric scores in this dataset range from -8 to +10. Learning gains are displayed on the x-axis. Some learning gain scores were negative, which represents that some students performed worse on the posttest than the pretest. Outliers were removed from the both treatments and thus *n* is reduced.

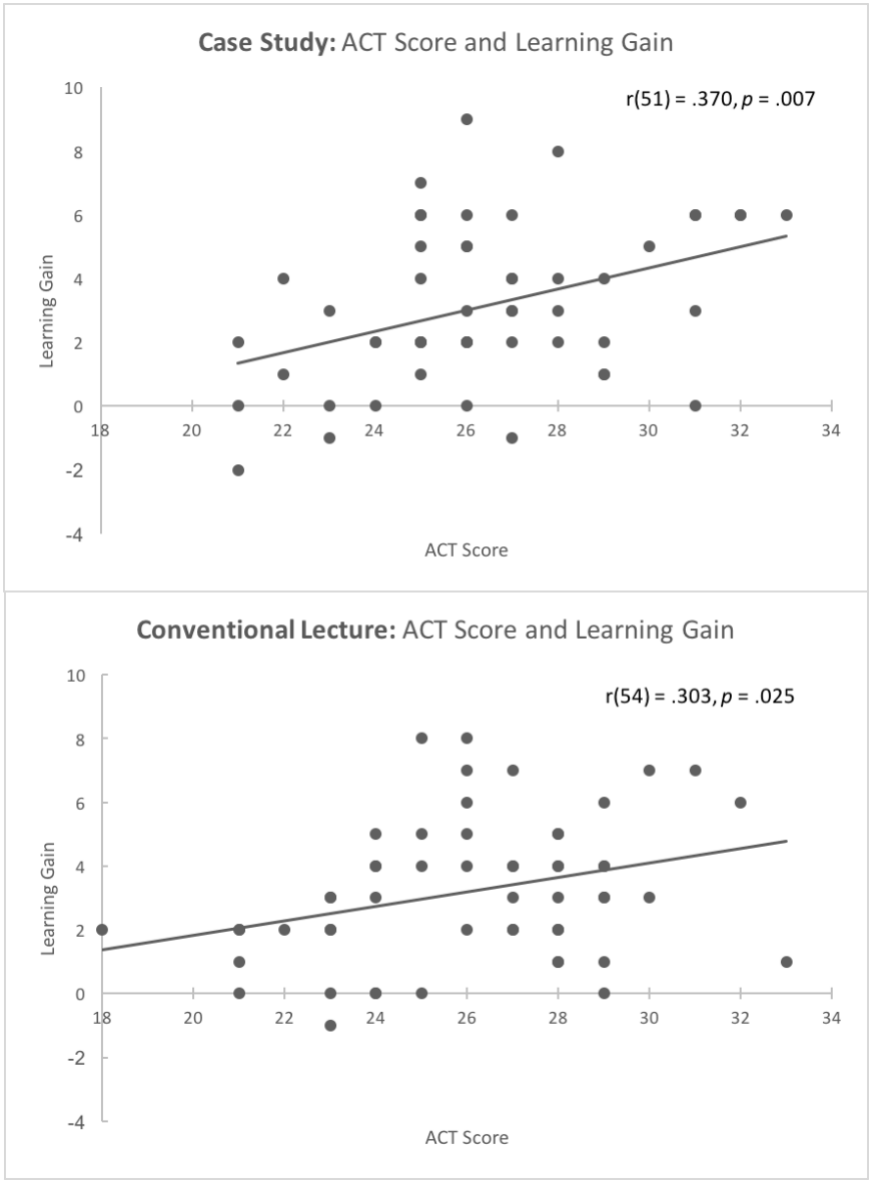

Figure 2.15. ACT Scores and Learning Gains Correlations.

Case study and conventional lecture treatment data are presented individually. Scores for the ACT can range from 0 to 36, however scores in this dataset ranged from 18 to 33 as indicated by

the y-axis. Learning gains are displayed on the x-axis. Some learning gain scores were negative, which represents that some students performed worse on the posttest than the pretest. It should be noted that not all participants had ACT scores, and outliers were removed from these data and thus *n* is reduced.

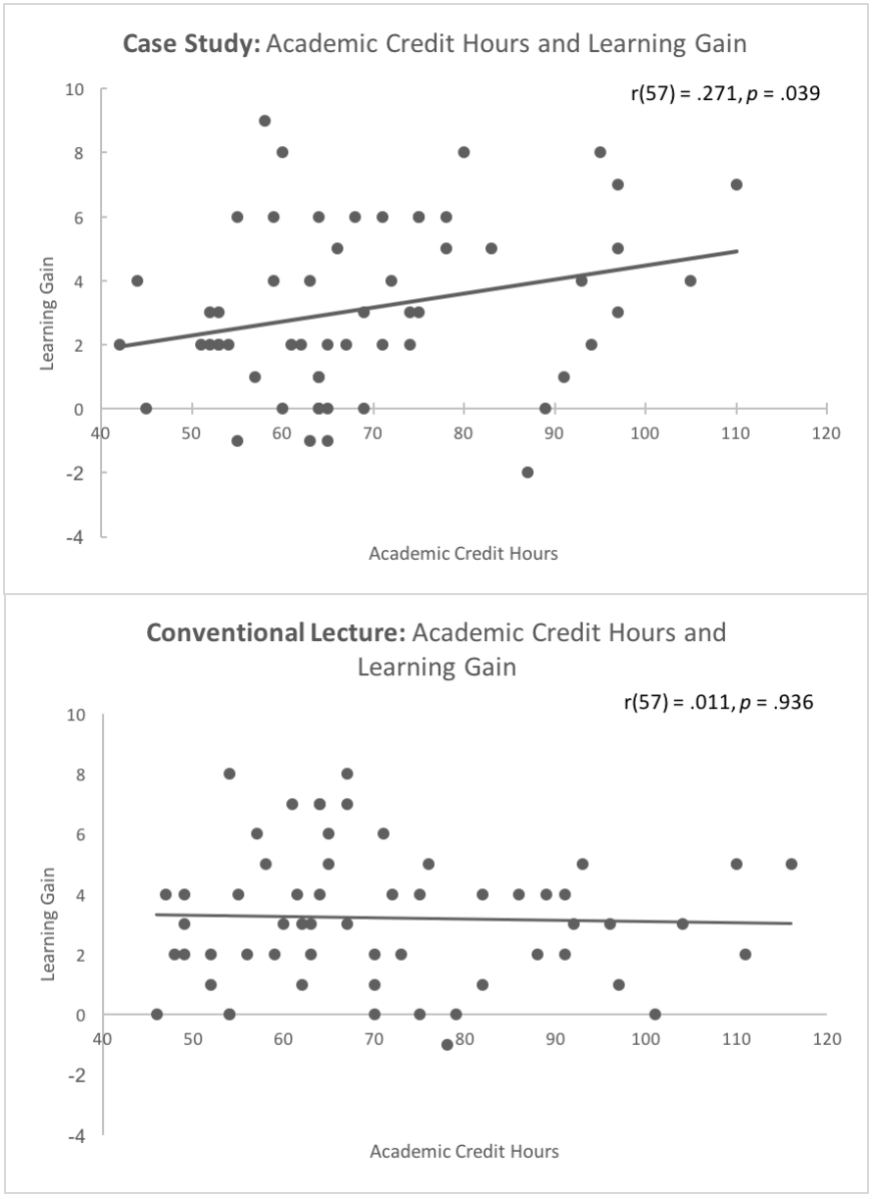

Figure 2.16. **Academic Credit Hours Completed and Learning Gains Correlations.** Case study and conventional lecture treatment data are presented individually. Academic credit hours may vary among students, however in this data set they ranged from 42 to 120 credits.

Learning gains are displayed on the x-axis. Some learning gain scores were negative, which represents that some students performed worse on the posttest than the pretest. Outliers were removed from the both treatments and thus *n* is reduced.

Qualitative Interviews

Emerging themes from each question asked during the interviews are diagrammed in Figure 2.17. Most students had used a case study in another class before this one and found the case study to be a helpful tool with an interesting story. The story within the case study was consistently reported to be interesting because it seemed relevant to their life or field they intended to pursue. All students found value in the interactive graphics, flowcharts, and bolded terms; these were the most helpful part of learning the material. The most documented complaint of the case study was that it was too lengthy, although most students did not think the material could be taught within a shorter time frame. Overall, students enjoyed the case study and would prefer to use the case study over a conventional lecture to learn this material.

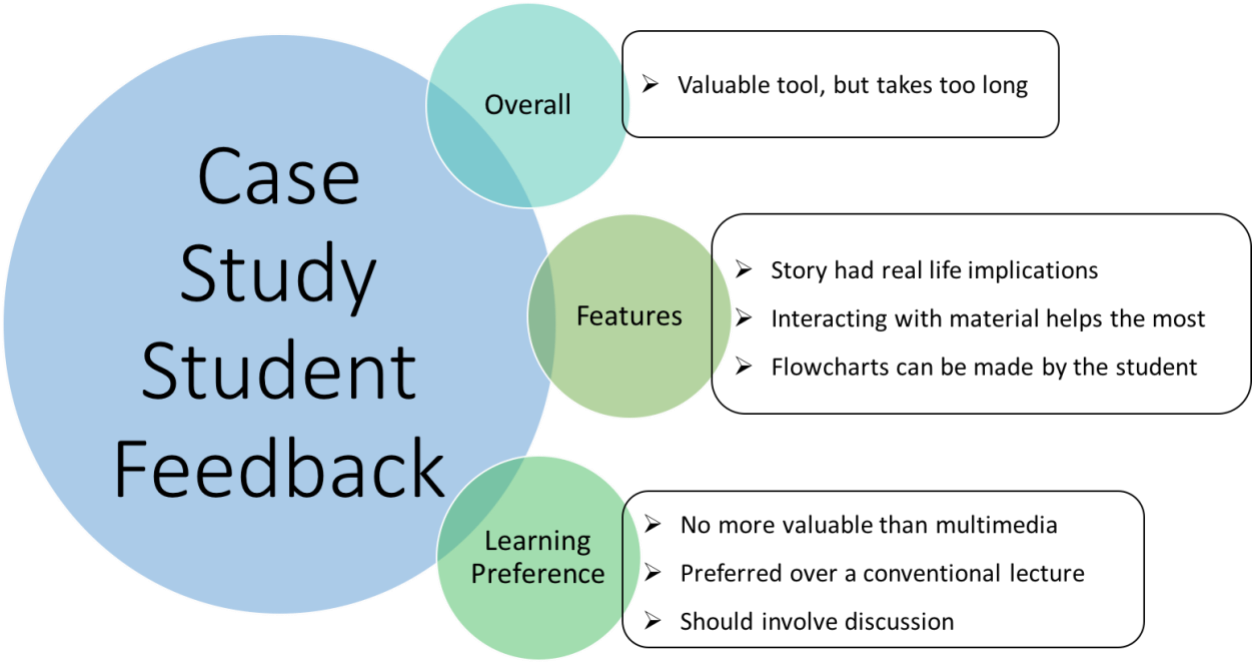

Figure 2.17. Emerging Themes of Insulin Resistance and Type II Diabetes Case Study Student Feedback.

Three main themes were identified from the interviews including overall opinion of the case study, features within the case study, and learning preferences. Overall, students found the case study to be a valuable tool. This emerging theme was supported by other emerging themes, including helpful features within the case study and student preference to learn from a case study.

Results II: Lactate Threshold Case Study

RQ1: Is there a relationship between learning gains from the use of a case study and student characteristics including prior knowledge levels, approaches to learning, ACT scores, or college credit hours completed?

Using the lactate threshold case study to validate results from the insulin resistance and type II diabetes case study, there were no relationships between learning gains from the case study and prior knowledge, approach to learning, or ACT scores. However, there was a positive and significant relationship between learning gains and academic credit hours completed, r(109) = .253**, p = .008 (Figure 2.18). To verify this correlation, a t-test was carried out to compare upper and lower tier students based on academic credit hours completed. The average number of credit hours among all students was 56. Students in the upper tier were defined as those who had 56 credit hours or more while lower tier students were defined as those who had less than 56 credit hours. As shown in Figure 2.18, the upper tier students had greater learning gains (4.19 +/- 1.77) from the use of a case study than lower tier students (3.32 +/- 1.90), t(108) = 2.42, p = .017.

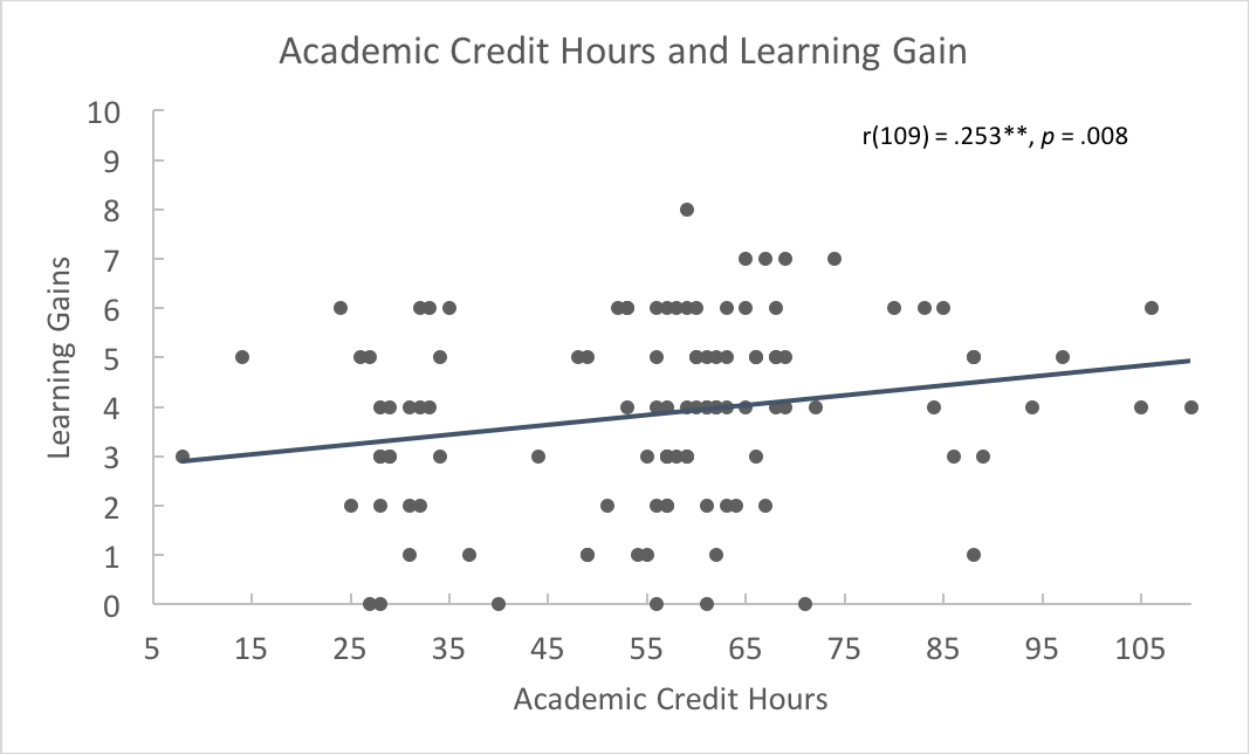

Figure 2.18 Academic Credit Hours Completed and Learning Gains Correlations. Academic credit hours are shown on the x-axis. Although academic credit hours can vary among students, these data ranged from 8 to 110 credit hours. Learning gains are displayed on the x-axis. Outliers were removed from the both treatments and thus *n* is reduced.

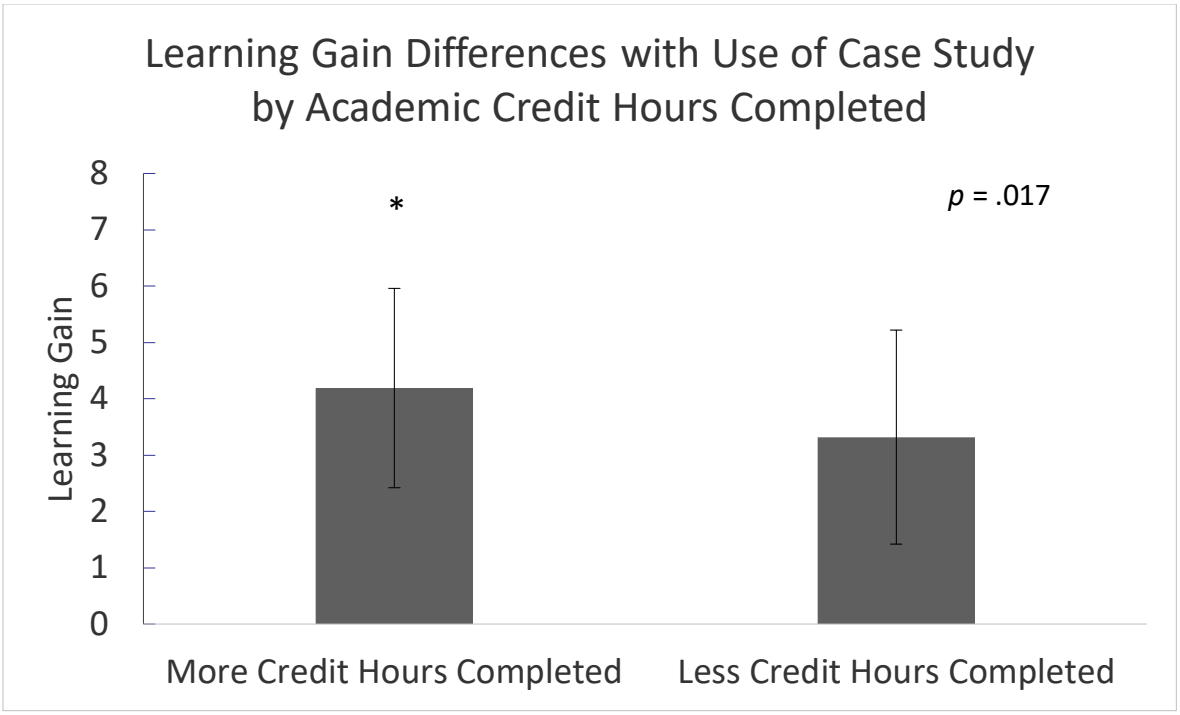

Figure 2.19 Learning Gain Differences with Use of Case Study by Academic Credit Hours Completed

Mean differences in learning gains between lower and upper tier students. Average number of academic credit hours completed was 56. The upper tier students were defined as those with 56 academic credit hours completed or more while lower tier students were those with less than 56 academic credit hours completed. Data are presented as means +/- SD.

Discussion

In this study the relationship between student characteristics and learning gains from the use of different pedagogical approaches were highlighted. Instructional approach is not black and white. As revealed in this study, selecting the proper instructional approach is quite complex. Despite being very different pedagogical tools, there were no significant differences in student learning gains between the case study and conventional lecture treatments. Although active learning has been a highly encouraged approach within all classrooms, especially in STEM, results from this study and a growing number of others do not support the notion that active learning will benefit all students, particularly those in introductory and intermediate science courses (Andrews et al., 2011; Hundley, 2007; Stull & Mayer, 2007; Wilson, 1999). For example, Andrews et al. (2011) found that student learning gains in various undergraduate biology courses were not associated with active learning. In addition, Hundley (2007) did not find any significant difference between a traditional and an interactive classroom in an introductory geology course. While some results from this study complement others, further exploration of these data highlighted the significance of student characteristics.

On the surface it may appear that active learning and conventional lectures provide the same benefit for students, however, a deeper investigation revealed that the benefit provided by each approach depends more on the characteristics of the students. There were positive and significant relationships found between PKA, SAL, academic credit hours completed and learning gains from the insulin resistance and type II diabetes case study. Based on these results, it appears that student characteristics have a large impact on learning gains from case studies. Conversely, there were no relationships between PKA, SAL, and academic credit hours completed and learning gains from a conventional lecture. Thus, students with more academic experience, higher prior knowledge, and a greater propensity to elaborate on information were

more successful with an active learning tool in the form of a case study. Students with less academic experience, lower prior knowledge, and a greater propensity to memorize information had greater learning gains from the use of a conventional lecture. There were, however, positive and significant correlations between learning gains from both treatments and ACT scores. College students with high ACT scores, therefore, appear to have greater success with either instructional approach.

The use of active learning tools in the form of a case studies resulted in greater learning gains from students with higher prior knowledge; whereas, the use of a conventional lecture resulted in greater learning gains for students with lower prior knowledge. These results are consistent with other studies in the literature that have reported the significant impact of prior knowledge on learning gains (Jensen, Kummer, & Godoy, 2015; Kirschner et al., 2006; Stull & Mayer, 2007; Thompson & Zamboanga, 2004; Tobias, 1994; Westrick et al., 2015). Stull and Mayer (2007) found that younger students with less prior knowledge had greater success creating their own graphics when provided with instructional guidance rather than extrapolating ideas on their own through reading text only. This likely occurred because the information was scaffolded, such that support was provided by the instructor or an educational tool, for students who did not have well developed schemata and were unfamiliar with the domain. Similar to that study, students in this study with low prior knowledge performed worse from the use of a case study. This was likely because the case study required students to interact with graphics and think about questions that asked them to apply the information they learned, rather than the graphics that were scaffolded for them in the conventional lecture. High prior knowledge learners do not need the same amount of scaffolded information because they have more developed schemata. This allows them to incorporate and apply incoming information to novel

scenarios and allows them a greater ability to accurately create and interact with graphics under minimal guidance. High prior knowledge learners with a well-developed cognitive architecture and a vast network of schemata allows for more efficient processing within the working memory system, as opposed to low prior knowledge learners who may experience cognitive overload (Clarke, Ayres, & Sweller, 2005; Cook, 2006; Kirschner et al., 2006; Leppink & van den Heuvel, 2015; Paas et al., 2004; Sweller, 1988; Van Merriënboer & Sweller, 2010; Young et al., 2014) The appropriate amount of instructional guidance depends on the learner's prior knowledge level, or schemata volume (Clarke et al., 2005; Cook, 2006; Kirschner et al., 2006; Thompson & Zamboanga, 2004).

In addition to prior knowledge, the approach a student takes to learning may influence the benefit derived from the use of active learning tools. Students who self-identified as preferring an elaborative rather than a memorization approach to learning had greater learning gains after completing the insulin resistance and type II diabetes case study. In contrast, students who self-identified as preferring a memorization rather than an elaborative approach to learning had reduced learning gains after the case study. Therefore, smaller learning gains will likely occur for students who have the propensity to approach learning through memorization. This finding agrees with the results reported by Rhodes and Rozell (2017) who found that using a more elaborative approach to learning information resulted in higher exam averages in an undergraduate physiology course. Students who take an elaborative approach to learning are more likely to make connections among schemata and think flexibly. Elaborative learners assimilate new information with previous experiences and previously acquired knowledge and try to use these to solve a novel problem. Although an elaborative approach to learning is much preferred, especially for students in physiology who may be pursuing a career in animal or

human medicine, many students have trained themselves to memorize information by habit. These habits are likely developed from success with memorization in other academic courses; however, this pattern of memorization becomes problematic within ill-structured topics like physiology. Furthermore, past academic experiences may have unknowingly encouraged and cemented this habit such that it becomes very hard to correct as students mature. Memorizing results in the formation of inert knowledge structures that are not synthesized in relation to or connected with other knowledge structures and therefore do not provide as much benefit for knowledge application in real world scenarios (Rhodes & Rozell, 2017). It is important to note that active learning tools will not necessarily encourage students to approach information via elaboration. Using active learning techniques could actually discourage some students and decrease motivation and persistence in STEM, leading to an increased propensity to memorize. This type of behavior is supported by the CLT. As intrinsic and extraneous loads increase, which could occur through the use of active learning tools within an ill-structured domain, germane load will be compromised (Kalyuga et al., 1998; Paas et al., 2004; Van Merriënboer & Sweller, 2010; Young et al., 2014). Thus, it is crucial to encourage students to approach learning via elaboration and carefully design learning tools that improve study skills. This must be considered first and foremost; continuing to use active learning strategies without first considering student characteristics will likely result in similar issues described in this study.

While prior knowledge and approach to learning are important indicators of the best teaching approach, ACT scores appear to have similar impacts on learning gains regardless of the approach. The ACT score was the only variable that positively correlated with learning gains from the conventional lecture and the insulin resistance and type II diabetes case study; therefore, students from both treatments had higher learning gains with higher ACT scores.

These findings are consistent with others that have found ACT scores to be a reputable prediction of academic success in undergraduate education (Thompson & Zamboanga, 2004; Westrick et al., 2015). According to these results, a student's ACT score is a good prediction of academic success using active learning or conventional lectures.

In addition to relationships between learning gains and ACT scores, other measures of academic preparedness such as undergraduate credit hours completed appear to be important indicators of success. The number of academic credit hours completed was positively correlated with learning gains from both case studies and negatively correlated with learning gains from a conventional lecture. Based on these findings, it is likely that active learning tools such as case studies will be more effective in advanced, more specialized classes taken later in the undergraduate curriculum or even in professional schools. This is reasonable based on results from others who have investigated the efficacy of active learning in professional programs including medical school, pharmacy school, nursing school, and veterinary school (Gleason et al., 2011; Hoke & Robbins, 2005; Koh, Khoo, Wong, & Koh, 2008; Newman, 2005; Schmidt et al., 2009). Conversely, conventional lectures may actually be better for learning gains in introductory and intermediate courses taken earlier in the curriculum. Other studies have reported that undergraduates who fell within the average range of 18-20 years of age, first or second year undergraduate students, did not benefit from active learning activities (Stull & Mayer, 2007; Wilson, 1999). Interestingly, there was a negative correlation between students with more academic credit hours and learning gains from a lecture, which is also supported by the CLT; specifically the phenomenon known as the expertise reversal effect (Young et al., 2014). This effect posits that students who have taken more undergraduate courses perform worse when taught complex concepts via conventional lectures because the information may

seem overly simple or redundant, and thus could result in the overthinking of concepts causing students to make additional mistakes. Given these results and those of others, the effectiveness of active learning tools, such as a case study, largely depends upon academic preparation of students.

Qualitative Interviews

Interviews conducted from participants in the insulin resistance and type II diabetes case study treatment provided insight about the utility of the case study as a learning tool. All students liked the case study overall and would prefer this form of learning over a conventional lecture. These results are consistent with other studies that have found that students enjoy active learning approaches (Gauci et al., 2009; Smith & Cardaciotto, 2011). However, many of these studies only evaluated the efficacy of active learning through a survey or interview without any qualitative data to support learning gains.

Three major emerging themes were identified after transcribing all interviews: features within the case study, learning preferences, and general opinion of the case study. As for features in the case study, students found the bolded terms, flowcharts, and interactive graphics especially helpful. Many of them stated that they were visual learners and that the features throughout the case study softened the text. In addition, most students mentioned that they would have drawn out a flowchart on their own. Flowcharts can be beneficial for students to consider applying physiological concepts to a novel scenario by "breaking" or missing a piece of the flowchart.

Student learning preferences were another emerging theme, as most students reported that they preferred a case study or form of multimedia to learn complex information like insulin resistance and type two diabetes. Many students said they cannot focus well when listening to a

lecture or watching a YouTube video. Students also mentioned that they prefer learning information alone first, and would prefer discussing the material in groups or as a class after. These data support the CLT, such that working in groups could be distracting in comparison to working alone as is adds extraneous load.

Overall, students reported that they enjoyed the story within the case study and many reported that it was relatable to a personal experience or to their future careers. While most students liked the case study, the quantitative data did not show that case studies provided equal benefits for everyone. It is important to consider qualitative and quantitative results when selecting a teaching approach. Although most students found the case study to be helpful and relevant, almost all of them reported that the case study was too long. While this was a big concern, most students agreed that, given the detail of information, there was no possible way to shorten the case study. While students may enjoy active learning, this does not guarantee it is the most effective way for all students to learn. Thus, drawing conclusions from data that were only collected from an opinion-based survey can be precarious without investigating the data more comprehensively.

Do Case Studies Really Work?

To validate the results from the insulin resistance and type II diabetes case study and confirm it was not simply an anomaly, relationships were examined between learning gains from the lactate threshold (LT) case study and student characteristics. Although, there was no relationship between learning gains from the LT case study and PKA, SAL, or ACT scores, there was a strong and positive relationship between learning gains from the LT case study and academic credit hours completed. After verifying this with t-tests, the number of academic credit

hours completed was shown to be a strong indicator of student success with the use of an active learning tool like a case study. Students who have had more experience in college courses likely have a more schemata and a well-developed cognitive architecture, and thus have the ability to think flexibly or transfer information into novel scenarios like those found in a case study. Additionally, the LT case study was presented approximately 10 weeks into the semester, whereas the insulin resistance and type II diabetes case study was given 6 weeks into the semester. Therefore, it is plausible that students from the LT case study were better prepared to solve abstract physiological problems, like those presented in the LT case study.

The LT case study validated the efficacy of case studies for certain types of students. Although there are many forms of active learning, case studies were selected for this study because they have been thought to engage students within various courses and class sizes (Dewprashad, 2013; Dori et al., 2003; Herreid, 1994, 2006, 2006; Herreid & Schiller, 2013; Raju & Sankar, 1999; Rybarczyk et al. , 2007; Smith & Murphy, 2016). Other active learning approaches, such as small group discussion, debate, collaborative or cooperative learning, can be influenced by the internet or other students which makes it difficult to evaluate individual student learning gains. A common concern about implementing active learning into large college classrooms is the lack of time for professors to design a tool or an idea (Bonwell & Eison, 1991; Faust & Paulson, 1998; Prince, 2004). However, science teachers have access to case studies that have been per reviewed from the National Center for Case Study Teaching in Science (http://sciencecases.lib.buffalo.edu/cs/). This website provides teaching notes, class management suggestions, and additional information that instructors may need to use the case study. Additionally, adopting active learning methods has been thought to take away time in class. This is a serious concern for some courses that are required to cover copious amounts of information

in a semester and cannot designate time for alternative, yet effective, teaching approaches. While small group or whole class discussion, pause lectures, or debates might disrupt allotted class time, case studies are an adaptable active learning instrument such that they can be used inside or outside of class time. Lack of funding for active learning activities or classroom environment are also common concerns, especially for activities that require props or excessive printing. However, case studies can be uploaded to the university portal system for student access as an individual or group assignment. Finally, many courses might have a very diverse population of students with varying levels of prior knowledge which could limit the type of active learning tool used. Although, if time permits, instructors could design case studies that are gaged towards specific types of students.

Conclusions and Future Directions

In conclusion, active learning is not always better than a conventional lecture approach. Additionally, student characteristics such as prior knowledge, approach to learning, ACT score, and academic credit hours completed all contribute to learning gains from an active learning tool like a case study. Based on these findings, an instructional approach that presents information with a high intrinsic load in the form of an active learning tool like a case study may present an overwhelming extraneous load for some students. This is especially true of students with less academic credit hours completed, as well as those with lower prior knowledge levels, lower ACT scores, and those who are more likely to memorize information. Thus, it is possible that a conventional lecture could be more beneficial for novice learners, especially if ill-structured domains are the focus as is the case in physiology and many other STEM disciplines. Conversely, students who have more academic credit hours completed and higher levels of prior

knowledge, higher ACT scores and those who prefer to learn through an elaborative approach will likely have greater learning success with active learning tools such as case studies. Thus, when intrinsic load is high, extraneous load becomes more problematic; this is especially true for lower tier students with less academic background and preparation. Active learning can increase the extraneous load for some students, therefore a conventional lecture may be more beneficial to make complex information easier to understand for less prepared students. To design effective instructional approaches for certain student populations, the causes of extraneous load for students must be considered.

In order to engage and encourage students to remain in STEM, the population of students must first be considered to select the proper instructional approach. Student characteristics, such as prior knowledge, approach to learning, ACT scores, and academic credit hours completed should be considered before utilizing active learning tools in college classrooms; otherwise, active learning tools may not be as effective as teachers may have hoped.

General Conclusion

This unique project allowed me to integrate scientific and educational research. Through this study, I learned many novel and ill-structured physiological concepts that required me to apply science in a way that I never had before. The educational portion of this project required me to build a case study, which prompted me to learn how to conduct a literature review, cite articles, and present information from scientific research to diverse groups of students. I learned how to take the information from my graduate research field work and apply those skills to teaching undergraduate students. Graduate students are rarely taught how to teach but are usually expected to do so. Through conducting a scientific and educational research project, however, I was able to grow and understand the role of graduate student in terms of research, writing, communicating, and teaching.

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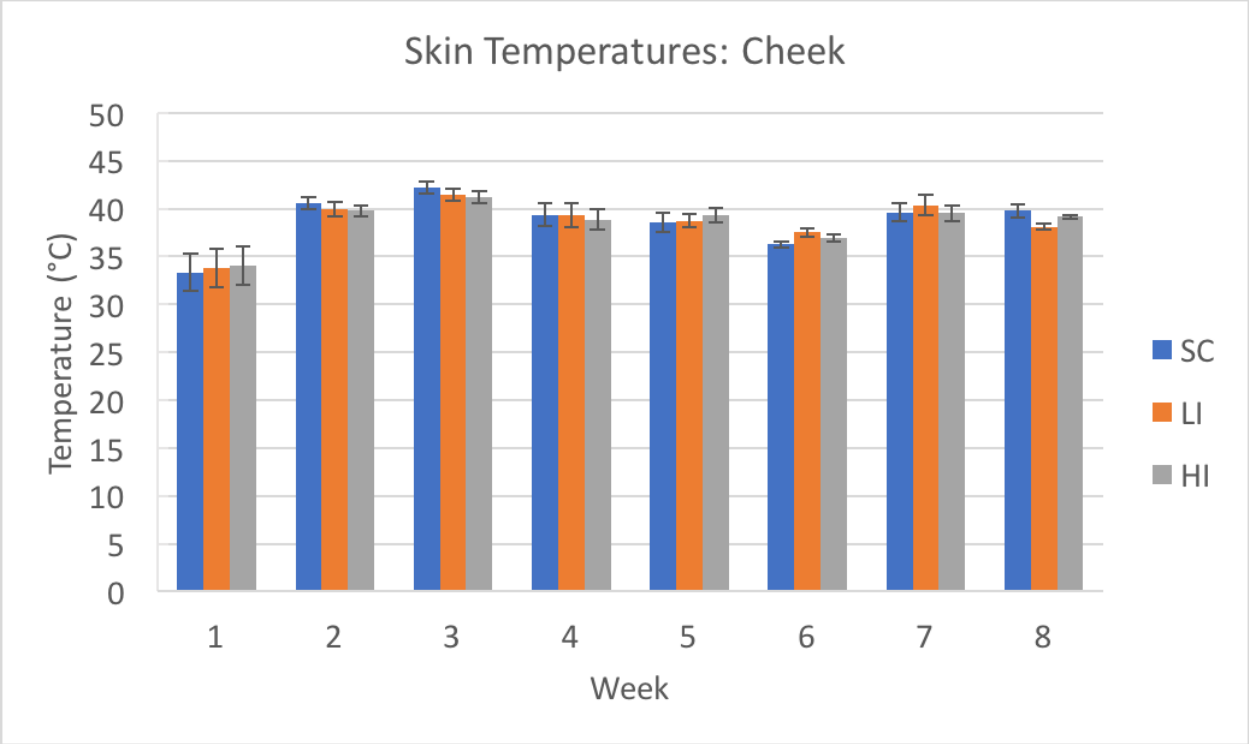
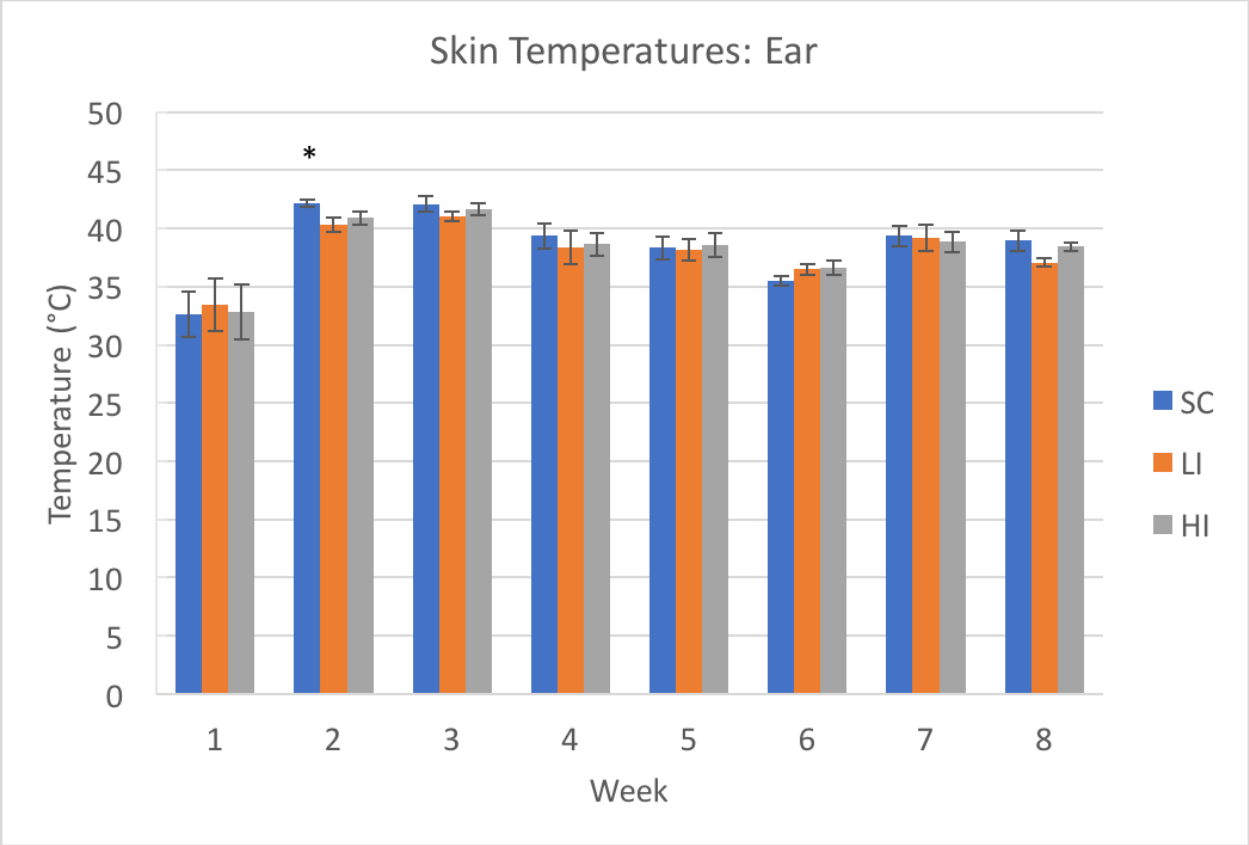
Appendix A - Appendix for Chapter 1



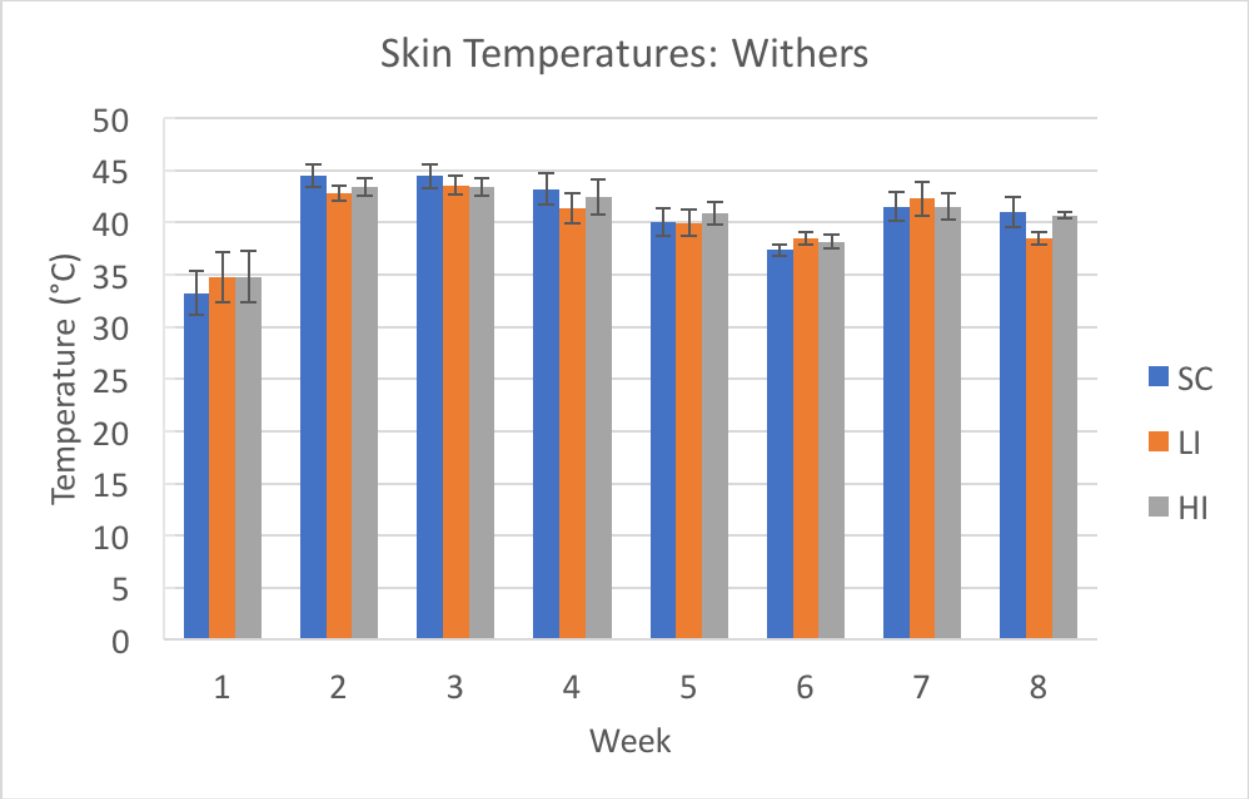
Appendix Figure A.1. Skin Temperatures of the Cheek.

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 and 2 weekly skin temperatures taken from the cheek were combined. The x-axis represents the week of data collection. The y-axis displays the mean skin temperature of the check for each treatment.



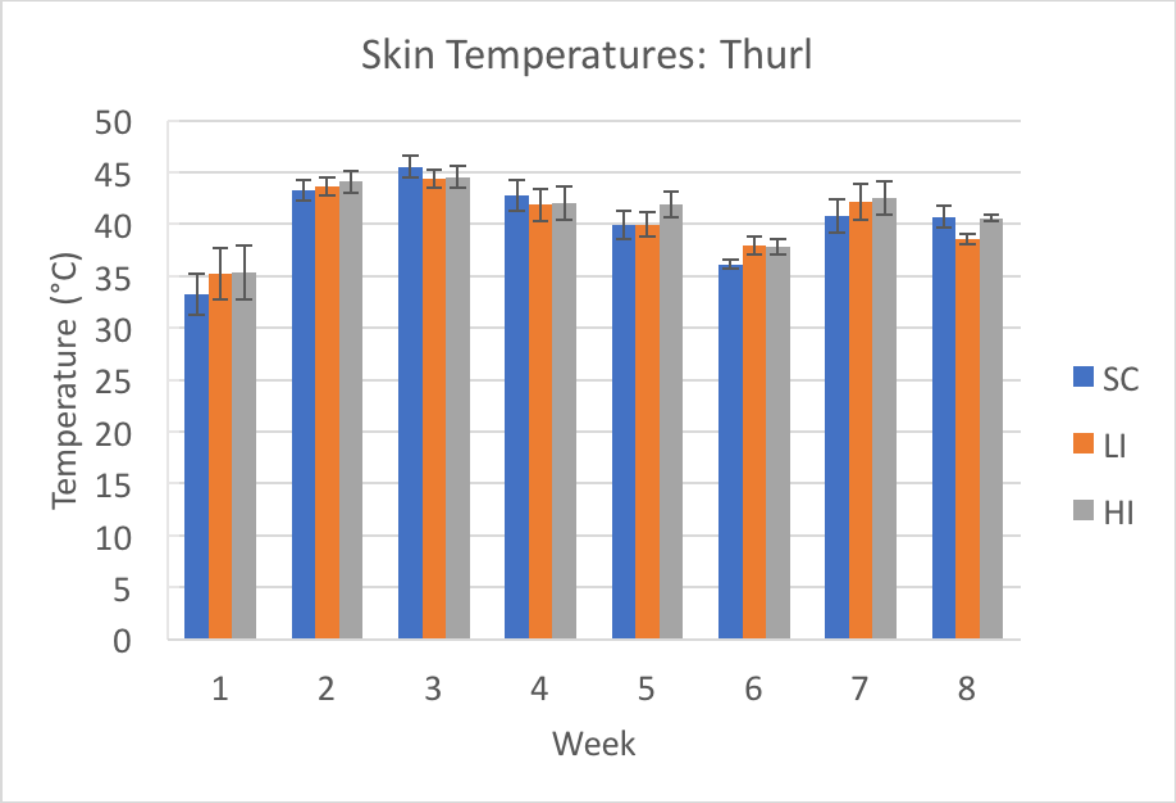
Appendix Figure A.2. Skin Temperatures of the Ear.

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 and 2 weekly skin temperatures taken from the ear were combined. The x-axis represents the week of data collection. The y-axis displays the mean skin temperature of the ear for each treatment. The sedentary control treatment had significantly higher skin temperatures on the ear on week two of exercise (p <0.05).



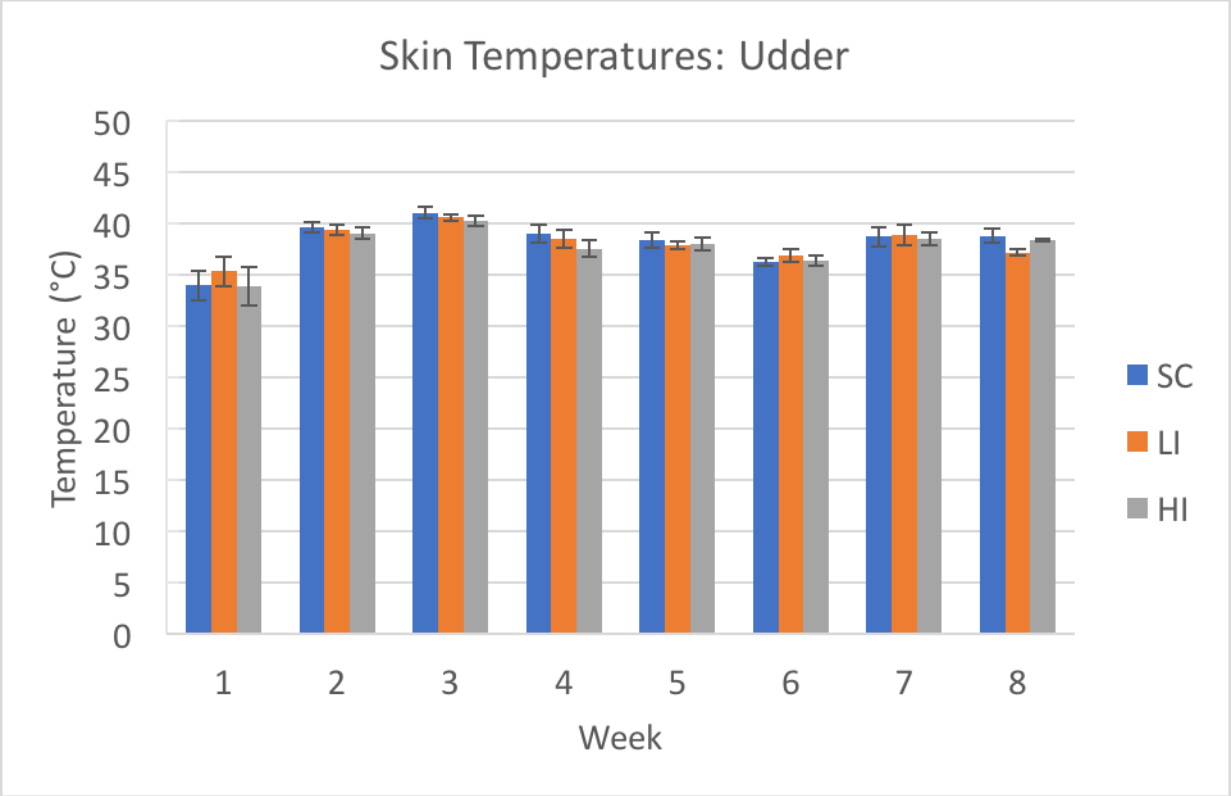
Appendix Figure A.3. Skin Temperatures of the Withers.

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 and 2 weekly skin temperatures taken from the withers were combined. The x-axis represents the week of data collection. The y-axis displays the mean skin temperature of the withers for each treatment.



Appendix Figure A.4. Skin Temperatures of the Thurl.

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 and 2 weekly skin temperatures taken from the thurl were combined. The x-axis represents the week of data collection. The y-axis displays the mean skin temperature of the thurl for each treatment.



Appendix Figure A.5. Skin Temperatures of the Udder.

Treatments: SC = sedentary control, LI = low-intensity exercise, and HI = high-intensity exercise. Trial 1 and 2 weekly skin temperatures taken from the udder were combined. The x-axis represents the week of data collection. The y-axis displays the mean skin temperature of the udder for each treatment.

Appendix Table A.1. SAS output for LDHa mRNA expression.

Effect	Week	Treatment	_Week	_Treatment	Estimate	Error	DF	t Value	|t|
Week	W0		W8		0.1678	0.06900	38.8	2.43	0.0197
Treatment		Control		High	-0.03901	0.1463	41.6	-0.27	0.7911
Treatment		Control		Low	0.1035	0.1482	41.6	0.70	0.4889
Treatment		High		Low	0.1425	0.1461	41.6	0.98	0.3349
Week*Treatment	W0	Control	W0	High	-0.06857	0.1643	65.1	-0.42	0.6777
Week*Treatment	W0	Control	W0	Low	0.2001	0.1666	65	1.20	0.2342
Week*Treatment	W0	Control	W8	Control	0.2125	0.1210	38.9	1.76	0.0869
Week*Treatment	W0	Control	W8	High	0.2031	0.1682	68	1.21	0.2316
Week*Treatment	W0	Control	W8	Low	0.2194	0.1710	68.2	1.28	0.2039
Week*Treatment	W0	High	W0	Low	0.2687	0.1643	65.1	1.64	0.1068
Week*Treatment	W0	High	W8	Control	0.2811	0.1692	68.5	1.66	0.1013
Week*Treatment	W0	High	W8	High	0.2716	0.1168	38.7	2.33	0.0253
Week*Treatment	W0	High	W8	Low	0.2880	0.1688	68.3	1.71	0.0924
Week*Treatment	W0	Low	W8	Control	0.01242	0.1712	68.3	0.07	0.9424
Week*Treatment	W0	Low	W8	High	0.002981	0.1682	68	0.02	0.9859
Week*Treatment	W0	Low	W8	Low	0.01933	0.1208	38.9	0.16	0.8736
Week*Treatment	W8	Control	W8	High	-0.00944	0.1731	71.1	-0.05	0.9567
Week*Treatment	W8	Control	W8	Low	0.006915	0.1755	71.1	0.04	0.9687
Week*Treatment	W8	High	W8	Low	0.01635	0.1726	70.9	0.09	0.9248

Appendix Table A.2. SAS output for LDHb mRNA expression.

Effect	Week	Treatment	_Week	_Treatment	Estimate	Standard Error	DF	t Value	Pr > |t|
Week	W0		W8		0.5272	0.1813	42.5	2.91	0.0058
Treatment		Control		High	0.3976	0.4691	45.8	0.85	0.4010
Treatment		Control		Low	0.4621	0.4758	45.8	0.97	0.3366
Treatment		High		Low	0.06448	0.4688	45.7	0.14	0.8912
Week*Treatment	W0	Control	W0	High	0.4024	0.5082	61	0.79	0.4315
Week*Treatment	W0	Control	W0	Low	0.7850	0.5156	61	1.52	0.1330
Week*Treatment	W0	Control	W8	Control	0.7457	0.3177	42.5	2.35	0.0236
Week*Treatment	W0	Control	W8	High	1.1386	0.5176	63.6	2.20	0.0315
Week*Treatment	W0	Control	W8	Low	0.8849	0.5261	63.8	1.68	0.0975
Week*Treatment	W0	High	W0	Low	0.3826	0.5082	61	0.75	0.4544
Week*Treatment	W0	High	W8	Control	0.3433	0.5193	64	0.66	0.5110
Week*Treatment	W0	High	W8	High	0.7361	0.3069	42.4	2.40	0.0209
Week*Treatment	W0	High	W8	Low	0.4825	0.5188	63.9	0.93	0.3559
Week*Treatment	W0	Low	W8	Control	-0.03931	0.5262	63.9	-0.07	0.9407
Week*Treatment	W0	Low	W8	High	0.3535	0.5176	63.6	0.68	0.4971
Week*Treatment	W0	Low	W8	Low	0.09987	0.3175	42.5	0.31	0.7546
Week*Treatment	W8	Control	W8	High	0.3928	0.5285	66.4	0.74	0.4600
Week*Treatment	W8	Control	W8	Low	0.1392	0.5365	66.5	0.26	0.7961
Week*Treatment	W8	High	W8	Low	-0.2536	0.5280	66.3	-0.48	0.6326

Appendix Table A.3. Regression equations for core body temperature (Tb), mean skin temperature (MST), and MST:Tb.

Body Temp	Control	Low	High
Tb	Tb = 35.11 + (0.047*THI)	Tb = 36.32 + (0.033*THI)	Tb = 35.26 + (0.046*THI)
MST	MST= 10.48 + (0.353*THI)	MST= 2.96 + (0.439*THI)	MST= 10.92 + (0.35*THI)
MST:Tb	MST:Tb= 0.32 + (0.008*THI)	MST:Tb= 0.14 + (0.010*THI)	MST:Tb= 0.33 + (0.008*THI)

Appendix Table A.4. Treatment order through the chute for trial 1, week 6 data collection. Treatments: 1 = sedentary control, 2 = low-intensity, 3 = high-intensity. Half of the heifers from treatment 1 were the *first* ten heifers to enter the chute, while 4/7 (57%) heifers from treatment 3 were the *last* ten heifers to enter the chute. Heifers from the treatment 3 had significantly higher MST than treatment 1 ($p = 0.029$).

Treatment	Tb	MST	MST:Tb
1	101.4	36.4	0.9
2	102.4	41.0	1.0
2	102.4	40.1	1.0
3	102.1	39.2	1.0
3	102.8	39.5	1.0
2	102.2	38.7	1.0
2	102.2	38.6	1.0
1	102.7	37.4	1.0
1	101.6	36.4	0.9
1	102.5	37.6	1.0
3	102.4	37.9	1.0
1	103.3	36.4	0.9
1	103.1	37.6	1.0
2	102.2	36.8	0.9
3	102.3	36.4	0.9
2	101.1	36.4	0.9
1	102.6	38.3	1.0
1	101.8	36.7	0.9
3	101.7	38.6	1.0
2	102.3	39.9	1.0
3	102.2	39.9	1.0
3	101.3	41.4	1.1

Appendix Table A.5. Treatment order through the chute for trial 2, week 1 data collection. Treatments: 1 = sedentary control, 2 = low-intensity, 3 = high-intensity. Four out of the seven heifers (57%) from treatment 3 were among the first 10 to enter the chute, while 4/7 (57%) of the heifers from treatment 2 were among the last 10 to enter the chute. Heifers from the treatment 2 had significantly higher Tb than treatment 1 ($p = 0.013$) and 3 ($p = 0.033$).

Treatment	Tb	MST	MST:Tb
3	103.7	44.1	1.1
1	103.3	41.3	1.0
3	102.9	41.6	1.1
2	104.2	40.4	1.0
1	103.4	41.5	1.0
3	102.8	42.2	1.1
2	104.1	42.3	1.1
3	103.1	42.5	1.1
2	104.2	43.1	1.1
1	103.3	39.9	1.0
1	102.6	40.8	1.0
1	103.6	40.4	1.0
2	102.9	41.2	1.0
1	102.6	40.9	1.0
3	102.5	39.7	1.0
3	103.9	40.3	1.0
3	104.0	43.6	1.1
2	105.0	42.8	1.1
2	104.4	41.7	1.0
2	104.0	40.9	1.0
3	103.8	40.2	1.0
3	103.7	44.1	1.1

Appendix B - Appendix for Chapter 2

Appendix Table B.1. **Core Concepts in Physiology.** These data were collected using a prior knowledge assessment that was given on the first day of class in an intermediate physiology course at a large Midwestern university. The bolded concepts are those that less than 3% of students answered correctly, these were considered the core concepts in physiology and used to design the insulin resistance and type II diabetes case study.

PKA question concept	(% Correct)	Case Study Implications
1. Diffusion and Transport	**0%**	**Understanding how glucose is transported throughout the body using many different types of active transport**
2. Diffusion	3.3%	
3. Diffusion and Osmosis	**2.7 %**	**Importance of movement and concentration gradient of glucose and other ions**
4. Glucose	**0%**	**Blood glucose regulation and utilization in insulin-dependent cells**
5. Insulin	**0 %**	**Role of insulin, glucose uptake, and insulin resistance leading to T2D**
6. ATP	**2.7%**	**Importance of glucose for ATP production**
7. ATP related	5.3%	
8. Cell membrane proteins	28.7%	
9. Negative feedback	29.3%	
10. Negative feedback	3.3%	

Appendix Table B.2. **Prior Knowledge Assessment.**

Prior Knowledge Assessment (PKA) questions and answers.

Multiple Choice Question/Statement	Multiple Choice Answer
ATP:	**a) Stands for Adenosine Triphosphate** **b) Is broken down by enzymes such that ATP → ADP + Pi, with Pi representing an energy source** c) Is produced primarily in the liver **d) Is used to provide energy for muscle contractions** e) Is used by the brain in large quantities in order to absorb glucose
The production of ATP:	a) Can only occur in the presence of oxygen **b) Requires catabolic and anabolic reactions** c) Can only occur if a cell has adequate amounts of glucose d) Always yields byproducts such as CO^2 and H^+ **e) Can occur via a process known as substrate level phosphorylation**
Choose any of the following that are correctly matched:	a) Facilitated diffusion – Process whereby molecules can pass through a cell membrane; the only thing required for this movement is a concentration gradient b) Simple diffusion – Process whereby molecules pass through a cell membrane in order to create a concentration gradient from one side of the cell membrane to the other **c) Primary active transport – Process whereby molecules are moved through a cell membrane against their concentration gradient, using ATP directly as a source of energy** d) Osmosis – Process whereby water diffuses from an area where there are more dissolved particles to an area where there are fewer dissolved particles

	e) Secondary active transport – Process whereby molecules are moved through a cell membrane against their concentration gradient, using the concentration gradient of another molecule as a source of energy
If a red blood cell is placed into a hypertonic solution of sodium chloride it could:	**a) Shrink due to osmosis** b) Swell due to osmosis c) Shrink due to diffusion d) Swell due to diffusion **e) Die due to inability to regulate metabolic processes**
The movement of water from the blood and into a cell:	**a) Is dictated by the tonicity of the cell** **b) Occurs more efficiently if membrane proteins such as aquaporins are present** c) Will slow if too much Na+ is present within the cell's cytoplasm **d) Could be disrupted if edema developed in the interstitial space** e) Is dependent upon the number of pumps within the cell's membrane that actively transport water into the cell
Glucose:	**a) Is regulated by insulin** b) Cannot be used by the brain for production of ATP **c) Is covalently linked to fructose to form sucrose** **d) Is composed of a six-carbon ring structure that also includes oxygen as a component of the ring** e) Cannot be synthesized by the body so must be obtained from the diet
Insulin:	a) Is secreted by the pituitary gland in response to high blood glucose **b) Should cause a decrease in blood glucose if all negative feedback pathways are working normally** **c) Could cause excess glucose in the blood to be converted to fat and stored within adipose tissue**

	d) Should increase the amount of circulating rather than stored energy, and thus would be elevated in those people who are undergoing high stress conditions **e) Is normally required by skeletal muscle cells in order to absorb glucose from the surroundings**
Cell membrane proteins:	**a) Can be upregulated or downregulated by the cell** b) Are fixed or set for the duration of the cell's life **c) Require transcription and translation before being inserted into the membrane** **d) Allow the cell to receive and respond to messages from other cells** e) Are the same within every cell of an organism's body
Negative feedback is a crucial component of homeostasis because:	a) It allows the body to anticipate changes and prepare for what is about to happen b) It amplifies or increases the deviation of a regulated physiological variable away from its set point **c) It returns a regulated physiological variable back to an acceptable range after its deviation has been detected** d) It prevents regulated physiological variables from ever deviating outside of their strict set point e) It requires conscious regulation by the brain and thus keeps the organism aware of its physiological condition at all times
Which of the following is a physiologically correct example of negative feedback to help keep body temperature at its normal set point when environmental conditions are much colder than body temperature?	a) Increased activity of insulin to catabolize fat tissues **b) Increased skeletal muscle activity to liberate energy in the form of heat as ATP is broken down** c) Increased blood flow to the skin **d) Increased piloerection to improve insulation at the surface of the skin** e) Decreased release of hormones that stimulate hunger sensations in order to save energy by reducing digestive processes

Appendix Table B.3. **Questions to Student Approach to Learning (SAL) Survey.**

Memorization	5 Item Likert-type scale
When I study, I try to memorize everything that might be covered.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree
When I study, I memorize as much as possible.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree
When I study, I memorize all new material so that I can recite it.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree
When I study, I practice by saying the material to myself over and over.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree
Elaboration	5 Item Likert-type scale
When I study, I try to relate new material to things I learned in other subjects.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree
When I study, I figure out how the information might be useful in the real world.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree
When I study, I try to understand the material better by relating it to things I already know.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree

When I study, I figure out how the material fits in with what I have already learned.	1) Strongly disagree 2) Mildly disagree 3) Neutral 4) Mildly agree 5) Strongly agree

Appendix Table B.4. Insulin Resistance and Type II Diabetes Pretest/Posttest Questions and Answers.

This was used to assess students understanding of glucose metabolism, insulin resistance, and type II diabetes. Correct answers to each question are shown in bold text.

Multiple Choice Question/Statement	Multiple Choice Answer
1. Glucose can enter an enterocyte via:	**a) GLUT2 transporter** **b) SGLT-1 co-transporter** c) Simple diffusion d) GLUT4 transporter
2. In order for glucose to exit an enterocyte and enter a capillary:	a) Insulin must be present and bound to its receptor on the enterocyte's basolateral membrane **b) Facilitated diffusion must occur** **c) Glucose levels inside the enterocyte must be higher than those in the blood** d) **A transporter is required to move the glucose across the basolateral membrane of the enterocyte**
3. Certain African plants contain a poison substance that blocks the activity of the Na+/K+ pump in enterocytes. How might ingestion of this plant affect glucose metabolism in a person?	a) No glucose could be moved from the lumen of the small intestine into the enterocyte b) More glucose than usual would be absorbed into the enterocyte **c) Less glucose than usual would enter the blood** d) D. Insulin would increase to help meet the increased demand for glucose
4. Assuming the person in the previous question survives, how might ingestion of this poison impact insulin resistance?	**a) It should decrease release of insulin and prevent down-regulation of the insulin receptor** b) It should ultimately result in an increase in Glut-4 on the cell membranes of skeletal muscle cells, and therefore help to reduce blood glucose c) It should increase insulin resistance in the person due to increased insulin secretion d) D. There is no physiological connection between Na+/K+ pumps in enterocytes and insulin resistance

5. In a healthy person, when glucose enters a skeletal muscle cell:	a) sodium must also enter the skeletal muscle cell, moving downhill with its concentration gradient. **b) blood glucose levels drop, even if only by a tiny bit.** **c) it can be broken down for energy production.** d) **it can be used to produce glycogen.**
6. Exercise can reduce or even reverse the symptoms of insulin resistance. Which of the following describes a potential benefit of exercise?	**a) Contracting skeletal muscles can move GLUT-4 to the cell membrane without insulin** **b) Exercise should increase utilization of glucose by cells and thus lower circulating glucose and insulin** c) Exercise should increase the secretion of insulin, which then improves the utilization of glucose by cells d) People with insulin resistance should not exercise because they do not have enough glucose for muscle contractions
7. Insulin resistance is:	**a) A disease in which certain cells become less responsive to insulin.** b) A disease in which the pancreas can no longer secrete insulin. c) A disease in which glucose can no longer be absorbed from the small intestine. d) A disease that requires people to inject insulin after each meal
8. How would insulin resistance impact a skeletal muscle cell?	**a) Insulin resistance would reduce the expression of certain cell membrane proteins** **b) Insulin resistance would decrease facilitated diffusion of glucose into the cell** **c) Insulin resistance would lead to a decrease in glycogen replenishment** **d) Insulin resistance would lead to a decrease in ATP production**

9. If you took a blood sample from a person with uncontrolled or untreated insulin resistance several hours after they ate a meal containing only complex carbohydrates, which of the following would you likely find?	**a) Elevated insulin levels that were well above the normal range** **b) Elevated blood glucose levels that were well above the normal range** c) Elevated levels of complex carbohydrates in their blood, well above the normal range d) None of the above; complex carbohydrates do not contribute to issues associated with insulin resistance
10. Which of the following drugs would help reduce blood glucose levels in a person already suffering from insulin resistance?	**a) A drug that downregulated the number of SGLT-1 co-transporters on the apical membrane of enterocytes within the small intestine** **b) A drug that was able to cause expression of SGLT-1 co-transporters on the basolateral membranes of enterocytes with a simultaneous increase in GLUT2 transporters expression on the apical membrane** c) A drug that increased the production and release of insulin by beta cells within the pancreas d) A drug that reduced the expression of GLUT4 transporters within skeletal muscle cells, causing them to be retained within the cytosol of these cells

Appendix Table B.5. **Insulin Resistance Case Study Interviews.**

The questions asked to each interviewee are on the left side if the table. Follow up questions to the interviewee's answer (bolded) are on the right side of the table. Which of the following is true of lactate?

Open Ended Question	Answers/Follow up questions
1. Have you ever used a case study in a class before? Did you find the case study to be a helpful or detrimental learning tool?	**Yes**: What class? Can you describe the case study that you used? **No**: What did you think of this case study? Did you find it to be helpful?
2. Did you find the information in the case study to be interesting or dull?	**Interesting:** What did you find to be interesting about this info? **Dull:** Why didn't you find this to be interesting?
3. Did you find the story that guided the case study to be helpful or not helpful to understand this information and why it matters?	**Helpful:** How was it helpful? **Not helpful:** Why did you not find this story to be helpful?
4. Did you find the flowcharts to be helpful or not so helpful? (Provide the flow charts with the answers filled in for the student to recall interacting with this)	**Helpful:** How was it helpful? **Not helpful:** Why did you not find this story to be helpful?
5. Did you find the interactive figures in the case study to be helpful or not so helpful to understand this information? (Provide the interactive figures with the answers filled in for the student to recall interacting with this)	**Helpful**: How were these helpful? ➢ **They helped me understand the material.** o Was it the interacting portion that helped or the final product? **Not helpful**: Why not? ➢ **They just confused me more**. o Were you not able to visualize what you had just read? o **Yes, exactly.** Would it have been more helpful if you were

	working together with someone else? ○ **No, not necessarily.** Why not? Would something else have been more helpful for you?
6. Did you find the bolded text to be helpful or distracting?	**Helpful:** How was it helpful? **Distracting:** Can you describe how these bolded terms were distracting?
7. What did you think about the length of the case study; was it too long, too short, or just right?	**Too long.** Would you rather have a shorter case study, or broken the case study up into several parts? **Too short.** Would you rather have a longer case study? **Just right.**
8. Would you have liked to discuss this in a group or with the class; or did you like completing this alone?	**In a group:** Why? Why not as a class or alone? **As a class:** Why? Why not in a group or alone? **I liked it the way it was.** Why?
9. Do you think you would have rather learned this information through watching a YouTube video, listening to a lecture, or multimedia (define this as: animation with narration that explains the information after reading it)?	**Youtube:** Why? **Lecture:** Why? **Multimedia:** Why? **No, I really liked the case study.** Why?

Appendix Table B.6. **Lactate Threshold Pretest and Posttest questions and answers.**

Correct answers to each question are bolded.

Questions	Multiple Choice Answers
1. Which of the following is true of lactate?	a) Lactate is only produced when a skeletal muscle cell is low or lacking oxygen b) Lactate and lactic acid are the same molecular compound, the only difference between these two is their location with lactate found inside the skeletal muscle cell and lactic acid found outside of the cell **c) Lactate can be converted to pyruvate within a skeletal muscle cell** d) **Lactate that spills into the blood from skeletal muscle cells can be used as a fuel source by different tissues within the body**
2. Lactate dehydrogenase:	**a) Is an enzyme, and thus a protein, and is produced in a skeletal muscle cell via transcription and translation** b) Can only convert pyruvate to lactate within the sarcoplasm **c) Can be produced in varying amounts and even in different forms within a skeletal muscle cell based on needs of the cell** d) **Allows the skeletal muscle cell to create a lot of lactate, even if oxygen is present in sufficient amounts**
3. Lactate produced in a skeletal muscle cell can:	**a) Help balance the pH of the sarcoplasm** **b) Exit the cell through a monocarboxylate transporter (MCT)** **c) Enter the mitochondria** d) Enter or exit the cell via simple diffusion
4. Skeletal muscle soreness after exercise is due to:	a) The accumulation of lactic acid within skeletal muscle cells b) The accumulation of lactate in the sarcoplasm of cells and in the extracellular fluid (ECF) c) An increase in pH within muscle cells **d) Accumulation of H+ in the extracellular fluid (ECF) surrounding skeletal muscle cells**
5. Lactate threshold refers to:	a) The greatest amount of lactate skeletal muscle cells can produce at any one time

	during exercise and excrete into the blood for use by other cells b) The point at which skeletal muscle cells have become so fatigued they can no longer contract and begin to release their stored lactate into the blood c) The amount of lactate a cell can produce and store before it stops functioning due to a severe decrease in pH **d) The point at which blood lactate levels substantially increase because the amount of lactate produced by skeletal muscle cells exceeds the amount that can be used**
6. Suppose an untrained, sedentary individual begins an exercise training program that exclusively focuses on jogging. What changes would likely occur in this individual's skeletal muscle cells after a period of about 4-6 weeks of dedicated training?	**a) An increase in the number of monocarboxylate transporters (MCT) within the sarcolemma of their skeletal muscle cells** b) **An increase in the amount of lactate dehydrogenase produced within their skeletal muscle cells** c) **An improvement in their lactate threshold such that less lactate would be present in their blood during moderate intensity exercise** d) **An increase in the number and/or size of mitochondria in their skeletal muscle cells.**
7. Which of the following would decrease the amount of glucose that could diffuse into the sarcoplasm of an actively contracting skeletal muscle cell within a 30 minute period of exercise at a moderate intensity?	**a) A decrease in the availability of glucose transporters, specifically GLUT4, within the sarcolemma** **b) Inability to efficiently convert glucose to glucose-6-phosphate within the skeletal muscle cell** **c) A decrease in the amount of O2 present in the sarcoplasm and thus the inability to continue the process of glycolysis** d) **Lack of adequate blood flow to the actively contracting skeletal muscle cell**
8. Suppose you are monitoring a person's blood in real time while they are jogging on a treadmill and you notice that while they have been at it for least 30 minutes, their blood lactate levels haven't changed much at all. How could this be?	**a) Less lactate is diffusing out of their skeletal muscle cells because it's being used within the cells producing it** b) The person had not entered a period of anaerobic respiration yet meaning their skeletal muscle cells still had access to

	sufficient amounts of O2 and weren't producing lactate c) Additional mitochondria have been produced within existing skeletal muscle cells during this bout of exercise so the cells are more efficient at aerobic respiration and are not producing as much lactate d) This person was very fit and didn't produce lactate to begin with
9. If you were asked to design a drug or therapy that helped alleviate skeletal muscle soreness in a person who had exercised too heavily given their current level of fitness, which of the following would you choose?	a) A drug that quickly eliminated any lactate that had accumulated outside of the sarcolemma as lactate is responsible for causing skeletal muscle soreness **b) A drug that helped buffer the H+ that had accumulated outside of the sarcolemma as the accumulation of H+ activates action potentials in pain neurons** c) A drug that blocked the channels responsible for the efflux of H+ and lactate from the skeletal muscle cells as neither of these are beneficial and instead just represent cellular waste products that could be broken down in the cell **d) A drug that reduced the ability of sensory neurons to detect H+ in the extracellular fluid**
10. Skeletal muscle cells are beneficial for the body in many ways. For example, skeletal muscle cells provide other cells in the body with beneficial compounds or molecules such as:	**a) Glucose via GLUT4 transporters** **b) Lactate via monocarboxylate transporters (MCTs)** c) Hydroxide ions (OH-) via simple diffusion through the sarcolemma d) ATP via simple diffusion through the sarcolemma

Appendix Figure B.1. Insulin Resistance and Type II Diabetes Case Study

NATIONAL CENTER FOR CASE STUDY TEACHING IN SCIENCE

From Twiggy to Tubby:

The Progression of Insulin Resistance and Type II Diabetes

by
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Part I – Glucose Digestion and Absorbtion

Timmy and His Fast Metabolism

Timmy had been way too skinny for as long as he could remember; he'd had a growth spurt early in high school but in spite of stopping at 6′2″ he was teased unmercifully by his friends and family for being a twig, only weighing 160 lb. Although he didn't realize it at the time, his lean physique contributed to his achievement of the highest vertical leap of all the athletes in his high school district and he was extremely well coordinated. Timmy made the varsity basketball team as a freshman in high school and was a starter all four years, making first-team all-conference his junior and senior year.

In spite of his athletic success, mostly he just heard the name calling and jokes about being so thin, and he packed in as many calories as he possibly could. In fact, all through high school Timmy chose his foods based on the amount of carbohydrates they contained. He had at least four large energy drinks per day, ate a large pizza and a box of donuts for lunch, snacked on one or two family size bags of potato chips, and his nightly routine consisted of chocolate milk and Oreo cookies. While he was the all-star basketball player in high school, his height left him with just a few scholarship offers from smaller schools and junior colleges. However, Timmy's ultimate dream was to become a surgeon so he chose to turn down all the scholarship offers and attend a large university with an excellent pre-med program.

Glucose Digestion

At this point in Timmy's life, he is able to metabolize glucose efficiently. Due to the fact that Timmy is extremely active, the cells in his body use glucose to make energy at a relatively fast rate to keep up with his active lifestyle. Let's take a closer look at what happens to the glucose Timmy consumes while he's still young, active, and healthy.

While carbohydrates are important for energy production in the body, many of us consume far too many carbohydrates just like Timmy. There are two basic types of carbohydrates: simple carbohydrates, or monosaccharides and disaccharides, and complex carbohydrates, or polysaccharides.

The process by which carbohydrates are made available to cells in the body begins with the movement of food from the *mouth* through the *esophagus*, into the *stomach*, and lastly into the *small intestine* where the final steps for chemical digestion and absorption occur. *Digestion* is the chemical or mechanical breakdown of nutrients whereas *absorption* is movement of nutrients in their simplest form into the body, this primarily occurs in the small intestine but some absorption can also occur through oral mucosa.

Abigail Wilson is a graduate student and Ashley Rhodes is an associate professor in the Division of Biology; Timothy Rozell is a professor in the Department of Animal Sciences and Industry.

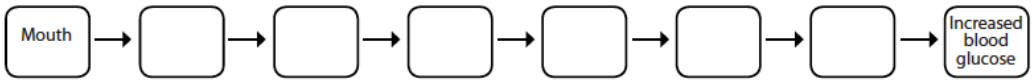
Mechanical breakdown is the physical breakdown of food, which involves grinding food with teeth or the muscular layers of the stomach. *Chemical breakdown* requires enzymes to break bonds, such as the breakdown of polysaccharides into monosaccharides and disaccharides. While most carbohydrates are chemically digested in the small intestine via enzymes secreted by the pancreas and intestinal mucosa, small amounts of chemical digestion also occur within the oral mucosa via salivary amylase. Effective carbohydrate digestion allows simple carbohydrates, such as the monosaccharide glucose, to be absorbed into intestinal epithelial cells called *enterocytes*.

Glucose is found in many, many foods, even those that do not taste sweet. Glucose is a large, polar, water-soluble molecule and therefore cannot passively diffuse across a cell membrane without transport membrane proteins. Once monosaccharides, like glucose, have passed through enterocytes, they will diffuse into the *extracellular fluid (ECF)* and finally into the blood via facilitated diffusion. Lastly, they will travel through the blood to the cells throughout the body, such as skeletal muscle cells to produce energy or energy stores, called *ATP* and *glycogen* respectively.

Activity 1

Fill in the flowchart (Figure 1) indicating where a polysaccharide travels through the GI tract beginning with oral ingestion and ending with monosaccharide and disaccharide absorption into the blood.

Figure 1. Movement of glucose from ingestion into systemic circulation.



Questions

1. Compare and contrast the processes of mechanical and chemical digestion in relation to the breakdown of polysaccharides. Where in the GI tract do each of these processes occur and what is involved in these processes?

2. How would a diet that includes more simple carbohydrates, rather than complex carbohydrates, impact glucose absorption from the GI and into enterocytes?

3. What might happen to glucose levels in the blood if carbohydrates could not be efficiently digested within the GI tract?

Cool Down and Recovery

After Timmy would play a hard game of basketball, he liked to cool down with a large chocolate milkshake. Every time he gulped down his post-workout milkshake he noticed that he soon felt ready for more basketball action, almost as though he had a renewed level of energy. His muscles felt good and he was ready for more. Considering Timmy's burst of energy after his ice cream ritual, we can conclude that he was able to ingest, absorb, and uptake glucose efficiently and utilize this glucose for energy within his skeletal muscle cells that provided him with the ability to run, dribble, and shoot basketball.

Glucose Absorption

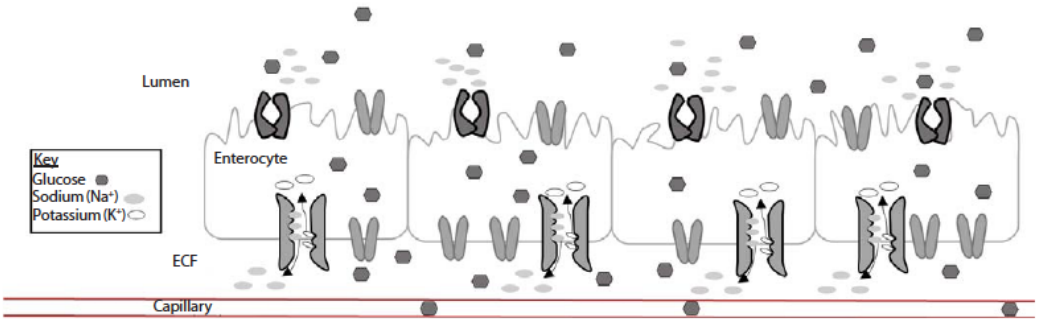
Once carbohydrates are broken down into their monomers, the process of absorption can begin. The *sodium glucose-linked transporter (SGLT-1)* is the primary transporter used for glucose absorption from the lumen of the small intestine into the intestinal epithelial cells called enterocytes. As indicated by its name, the SGLT-1 moves both sodium and glucose at the same time, making it a symporter. Like most symporters, the SGLT-1 relies on the work of Na+/K+ pumps to keep intracellular sodium levels low. These pumps are located along the basolateral membrane of enterocytes and use ATP to continuously pump sodium out of the cell while pumping K+ into the cell. Once glucose enters the enterocyte via the SGLT-1 it will diffuse into the extracellular fluid (ECF) through a uniporter transport protein called *GLUT2* located on the basolateral membrane. GLUT2 also permits facilitated diffusion, as glucose moves from a high concentration to a low concentration.

When luminal glucose levels are very high, for example, after finishing a meal, GLUT2 can also assist the SGLT-1 transporters on the apical side of the enterocytes. The *translocation*, or movement between cellular compartments, of some GLUT2 transporters from the *basolateral* membrane to the *apical* membrane of the enterocyte helps increase glucose absorption. When glucose levels within the enterocyte are low, glucose will still diffuse into the cell because the Na+/K+ ATPase pumps continually pump sodium out of the enterocyte to drive sodium into the cell, and thus pull glucose and sodium through the SGLT-1.

Activity 2

In the figure below, use arrows to label the *Na+/K+ pumps, SGLT-1 transporters,* and *GLUT2 transporters.* Then, indicate the *direction* in which glucose and sodium are moving through the SGLT-1 and GLUT2 transporters using arrows.

Figure 2. Glucose absorption in the jejunum of the small intestine.



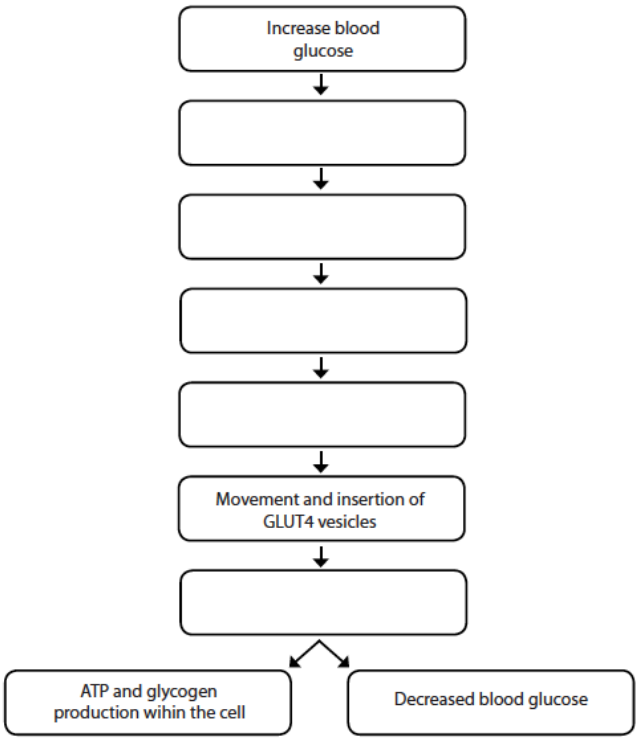
Recall that after Timmy absorbs the glucose from his chocolate milkshake, he gets a burst of energy and is ready to play another round of basketball. As long as Timmy is able to absorb glucose from his diet efficiently it will enter his blood stream and will travel to his body's cells, including *pancreatic beta cells.* When pancreatic beta cells receive glucose from the blood they respond by releasing *insulin*. This hormone travels in the blood and signals any cell possessing an *insulin receptor* to take up glucose from the blood. Insulin is required for glucose uptake in some cells; these are referred to as *insulin dependent* cells and include resting skeletal muscle cells and adipocytes, or fat cells. *Insulin independent* tissues can absorb glucose without insulin; these include exercising skeletal muscle cells, neurons and glial cells, and hepatocytes, or liver cells. We will use a *skeletal muscle cell* as our model for understanding how this process occurs.

In skeletal muscle cells, insulin receptors are located on the sarcolemma and t-tubules. Once insulin binds to its receptor, a *second messenger pathway* is activated which involves intracellular signaling that results in the translocation of *GLUT4* to the *sarcolemma* or *t-tubules*. GLUT4 is a transport protein that is stored and moves within the cell via *storage vesicles*. The storage vesicle will *fuse* to the sarcolemma or t-tubule to provide GLUT4 transporters and will allow a way for glucose to enter the skeletal muscle cell. The expression of GLUT4 within the sarcolemma or t-tubule is triggered by both insulin and muscle contraction. Thus, skeletal muscle cells can be considered as both insulin dependent and insulin independent. Regardless of what activates GLUT4 translocation to the sarcolemma and t-tubules, glucose will enter the skeletal muscle cell and be used for ATP or glycogen production.

Activity 3

Fill in the flow chart below indicating what will happen to resting skeletal muscle cells when insulin is released from the pancreatic beta cells.

Figure 3. Insulin release and glucose uptake.



Questions

4. The flow chart you have just filled out (Figure 3) represents a resting skeletal muscle cell. Will the rate of glucose uptake change in an exercising skeletal muscle cell? If so, how?

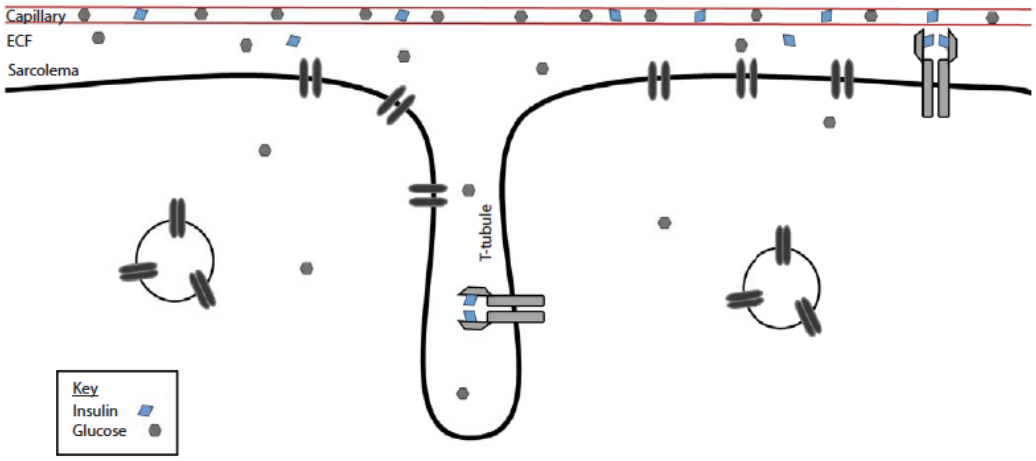
5. If insulin cannot properly interact with its receptor, how might this affect glucose uptake into the resting skeletal muscle?

6. Suppose glucose in the blood cannot be detected by pancreatic beta cells. Refer to your flowchart above and explain how this will affect glucose levels in the blood. Will this influence energy production among insulin dependent cells?

Activity 4

In Figure 4 below, use arrows to label the following: insulin in the extracellular fluid (ECF), glucose in the ECF, insulin receptor, GLUT4 storage vesicle, GLUT4, and GLUT4 fusion into the sarcolemma and t-tubule. Then indicate the direction in which glucose is moving through the GLUT4 transporter.

Figure 4. Glucose uptake into skeletal muscle cell.



Part II – Insulin Resistance

Testing Timmy

"Mom! You're washing my jeans in hot water and they're getting too tight!" said Timmy as he was preparing to return to college after spring break. In spite of being several pounds overweight herself, Timmy's mom replied, "Nope, the water heater is broken. Timmy, I'm afraid you've gained some weight. It looks like we'll be doing some clothes shopping before you go back to school!"

When Timmy began college he soon realized it would be the best time of his life. And while he lost all interest in playing sports, he thoroughly enjoyed attending all of the university sporting events. His favorite activity was tailgating at college football games. Over lunch with his best friend Ashlyn at the college dining center, Timmy sat down with his usual pizza, potato chips, Coke, and his favorite self-serve ice cream.

Ashlyn took a deep sigh and said, "Timmy, do you ever eat anything green? You know, food with nutrient value…"

Timmy replied, "HA! I hate anything green, all healthy food tastes horrible."

Ashlyn looked at him disturbed. "Timmy, I'm only concerned about your eating habits because you've gained quite a bit of weight since we met the first week of school in the dorms." Timmy looked at his plate and sighed; he suddenly started to feel anxious.

The following summer, Timmy had a yearly check-up appointment at his family doctor. Dr. Wilson noticed that Timmy's weight had increased to 203 lb and immediately began to question Timmy's weight gain and lifestyle. After evaluating Timmy's dietary lifestyle, she decided that he would need to return the next day at 8:00AM to complete a number of blood tests. Timmy's eyes widened and his mouth dropped when Dr. Wilson informed him that he had to fast before the test and could neither eat nor drink anything but water after midnight.

The first test performed on Timmy was an A1C, which measures average blood glucose for the past two to three months. The second test was a fasting plasma glucose (FPG) test that includes drawing a sample of blood after fasting for at least eight hours. Blood is drawn again every 30-60 minutes for up to three hours, this test is often repeated on a different day.

Once Dr. Wilson reviewed Timmy's lab results, she empathetically approached Timmy with serious concerns about his health.

Table 1 below includes cut-off ranges and diagnoses for average blood glucose percentages (A1C) and fasting plasma glucose (FPG). The A1C test is based on the attachment of glucose to hemoglobin proteins located on red blood cells (RBC). Considering the average lifespan of a RBC is about three months, this test indicates an average of blood glucose levels over time. FPG tests requires a patient to be fasted for at least eight hours which will indicate the amount of glucose in the blood without food consumption. These tests are both helpful in diagnosing diabetes because if glucose levels in the blood are elevated on average over the course of three months and/or after an eight hour fast, we can conclude that glucose is not being taken up effectively by the cells in the body, such as skeletal muscle cells.

Table 1. Blood glucose tests.

Test	*Normal/Average*	*Prediabetes*	*Diabetes*
A1C (%)	Less than 5.7%	5.7-6.4%	6.5% or greater
FPG (mg/dL)	100 mg/dL	100-125 mg/dL	126 mg/dL or greater

Question

7. Predict Timmy's A1C and FPG values as an inactive college student. Explain your reasoning for these values and how you believe Timmy has been placed in this category.

   A1C: *FPG:*

In healthy people, such as high school Timmy, blood glucose is a tightly regulated physiological variable requiring constant communication, interaction, and feedback between many body systems. If any of these components are disrupted or fail, blood glucose levels will become difficult to control and wide fluctuations begin to occur.

As we have seen with Timmy, the inability to regulate blood glucose has led to chronically elevated blood glucose levels, a condition known as hyperglycemia. If allowed to persist, hyperglycemia can lead to elevated levels of insulin in the blood, a condition known as hyperinsulinemia. However, despite the fact that additional insulin is continuously being released, the body's cells become less and less able to respond. This decreased responsiveness to insulin is termed insulin resistance (IR). IR further contributes to hyperglycemia as the signal for blood glucose uptake by insulin-dependent cells becomes less effective perhaps due to changes in the conformation of the insulin receptor or changes in the second messenger pathway leading to a decrease in expression of GLUT4 transporters within cell membranes. Regardless of the cause, the result is often the same: insulin regulated glucose uptake into cells drastically declines while blood glucose levels increase. If this problem persists, the beta cells in the pancreas that produce insulin begin to fail, resulting in hypoinsulinemia although hyperglycemia remains. At this point, insulin resistance is diagnosed as prediabetes or diabetes, depending on the severity of the hyperglycemia.

Let's take a moment to recap the sequence of events that occurs over time with insulin resistance, prediabetes, and type II diabetes: chronically elevated blood glucose levels leading to hyperglycemia, increased insulin release from beta cells of the pancreas, hyperinsulinemia, downregulation or changes to insulin receptors present within cell membranes, decreased glucose diffusion into cells, continued hyperglycemia, exhaustion of beta cells.

Part III – Type II Diabetes

Twenty-Five Years Later

Timmy graduated college, got married, and had two kids. Although he originally was interested in becoming a surgeon, he switched his major to teaching high school science. He now taught biology and coached basketball at the high school in his home town. His lifestyle had not changed much since college; he still refused to eat his greens, and consumed large amounts of soda pop and potato chips. Timmy was required to have blood tests every one to two years since being diagnosed with prediabetes back in college.

"Hello Timmy! According to your chart, it appears that you've gained some weight since your last visit with us," Dr. Wilson stated as she walked into the patient room where Timmy had plopped down on the examination table.

"Yeah, I'm *still* struggling with this whole dieting thing Dr. Wilson," Timmy said with his head down in embarrassment. "I just love the food I eat so much, it's like I'm addicted to it!"

Dr. Wilson contemplated the situation and asked how often he exercised. Timmy replied, "Dr. Wilson, I couldn't tell you the last time I participated in any exercise. I don't even enjoy playing basketball anymore! I'm dog-tired every day and don't have the energy to keep up with my two kids."

Dr. Wilson replied, "It appears you have also been experiencing aches all over your body and loss of sensation in your feet?"

"Yes doctor; I didn't even feel the giant splinter I got in my foot last weekend and my entire body just seems to ache every day."

Dr. Wilson looked down at Timmy's blood results from earlier that morning and took a deep breath. "Timmy, it's time to make some serious lifestyle changes. According to your results this morning, you have officially developed type II diabetes. Previously we diagnosed you with prediabetes as your body became resistant to insulin; that means your body was not able to utilize glucose efficiently. At that point your body was still able to make sufficient amounts of insulin, which is a hormone that permits glucose to enter into your body's cells to make energy. But now your body is not making enough insulin to support your energy needs. Now, all we can do is consider disease management options. You *must* start paying close attention to your diet, stay away from fast foods and processed carbohydrates. Also, you absolutely *need* to exercise for at least 30 minutes every day. Ask your wife or kids to hold you accountable for these things. We should also consider the use of metformin, a diabetic medication that will assist in lowering your blood glucose levels, or insulin injections. Also, we have a diabetes educator here at the clinic I encourage you to make an appointment with."

While Timmy's case involves the development of type II diabetes through poor diet and lack of exercise, lifestyle choices are one of many causes linked to this multifaceted disease. Type II diabetes can also result from a genetic predisposition, fat distribution throughout the body, age, sex, hormones, infection, and/or race/ethnicity. Insulin resistance and type II diabetes is still not entirely understood and is an active area of research and investigation.

Questions

8. During the early stages of IR, Timmy did not need insulin injections, however Dr. Wilson explains that at this point in Timmy's diagnosis, insulin injections may be necessary for his health. Why do you think Timmy did not need insulin injections when he was insulin resistant, but now that he has type II diabetes he does?

9. Explain how the ingestion of fast food, such as French fries, can cause a more dramatic change in the blood glucose concentrations in a type II diabetic as compared to a normal individual. Be sure to include any transporters necessary for glucose uptake in enterocytes and absorption into resting skeletal muscle cells, or lack thereof.

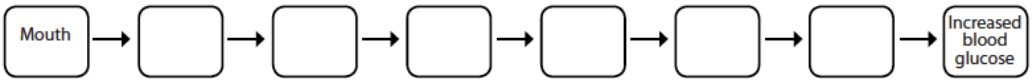
10. If you were working in a lab that studied IR and type II diabetes and needed to develop a drug to help treat type II diabetes, what would you want your drug to do and where would it be useful for it to interact? Explain why a drug with this mechanism of action would be helpful.

In this last section, we'll begin to pull together many of the concepts investigated in this case study and gain a more in-depth understanding of the complex web that exists between lifestyle choices and the development of IR.

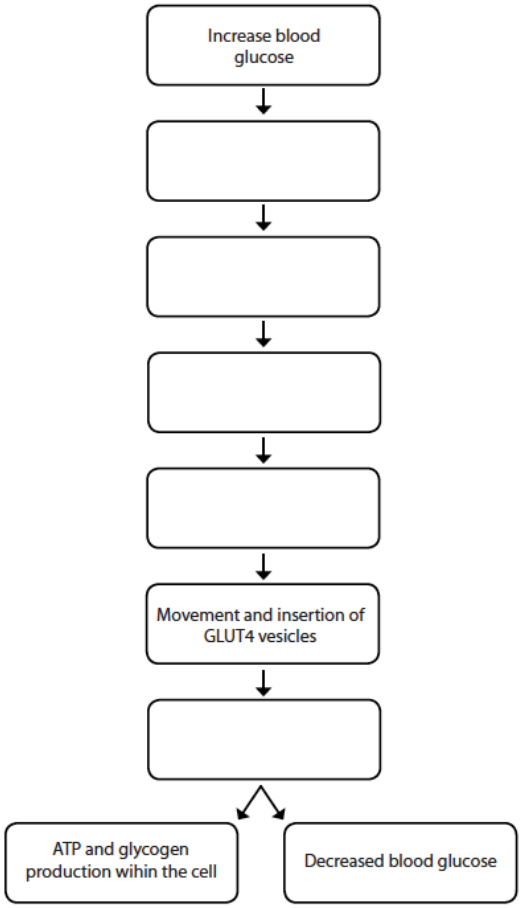
Activity 5

Copy your previous flow charts (Figures 1 and 3) into the combined graphic on this page (Figure 5). When shown together, these flow charts illustrate the sequence of events that should occur after the consumption of carbohydrates and ending with the use of glucose by a cell. However, in a person who is suffering from IR and on their way to becoming diabetic, these sequences break down and in doing so begin to reveal the complex web of events that exist between the digestive system and the cells which rely on insulin for glucose uptake. Keep in mind that the sequences you have created so far represent the ideal. Now, let's see how IR and diabetes impact these by working through the following.

Figure 5. Linking diet, exercise, and insulin resistance together: impacts on a skeletal muscle cell.



- During the end stages of IR and the beginning of diabetes, pancreatic beta cells no longer produce adequate amounts of insulin due to exhaustion. Put an *X* over the one box in your flow chart that best represents this specific disruption and note how many subsequent steps are negatively impacted.
- *Draw a line* between the box you just put an *X* over to the box describing the resulting effects on blood glucose levels.
- Suppose Timmy continued to eat simple carbohydrates despite his diagnosis of IR or even diabetes. *Draw lines* indicating areas along the digestive tract where glucose, as a simple carbohydrate, can be absorbed from his food to the box describing the effects on blood glucose levels.
- While changes are occurring within the blood, changes are also occurring within resting skeletal muscle cells that leaves them without adequate fuel for contracting. *Shade in the box* that identifies these fuel sources to indicate a diminishing supply.
- If these fuel sources within skeletal muscle cells cannot be created, what impact would this have on blood glucose levels? *Draw a line* from the box you were just directed to shade in above to the resulting impact on blood glucose levels.
- Suppose you were asked to design a drug to help reduce blood glucose levels in a person with insulin resistance after they had eaten simple carbohydrates. *Using asterisks, mark the boxes* within your flowcharts that would be good targets for this blood glucose-reducing drug. Keep in mind there are several ways to reduce blood glucose levels.



Appendix Figure B.2. Lactate Threshold Case Study

"I hate running."

This was Shelby's first thought upon hearing the announcement in class about an exercise challenge where students in two different courses competed to see which course could log the most miles during a semester. Her second thought was that she was sorry she even came to class this day; her friends were excited about starting the exercise challenge right away and wouldn't stop talking about it but Shelby was skeptical and pnot at all enthused. For as long as she could remember running was always difficult, she always felt out of breath and her muscles burned painfully whenever she would attempt to run more than a mile. The next day would be even worse due to muscle soreness. Because of this, Shelby never stuck to any sort of training program even though she had heard the benefits of doing so in many kinesiology courses she had taken in college. For her, it was always much easier to tell someone else what they needed to do to get in shape but she found it hard to follow her own advice.

"So, what do you think about this exercise challenge?" Patrick asked after class. Shelby just rolled her eyes and said, "I think you'll excel, like you do with everything, but I'm not interested." Patrick just giggled. "But what if we exercised together? We could use the time to not only get in shape but also discuss the concepts we are learning in class. Maybe this would even help our grades a little." Shelby definitely needed help with the course material and Patrick talked a lot about how he had in-depth experience in many areas of anatomy and physiology. "Ok," Shelby said, "but don't expect me to be able to keep up with you. I can tell from your physique that you exercise a lot." Patrick just smiled and said, "How about we start first thing tomorrow morning with just a two mile run? Maybe we could even do a few sprints for fun!"

Day 1:

As Shelby had predicted, the run was physically hard. Her muscles burned, she was short of breath, and she couldn't wait for it to just end. "Wow, that was awful." Shelby said to Patrick as they sat in the grass, stretching after their run. "I never want to do that again! I'm just not made for running, especially endurance running. I'm sore already thanks to all the lactic acid in my muscles." Patrick shot her an inquisitive look and said, "I don't think it's the lactic acid that's making you sore, but I could be wrong. I remember hearing something about this in class." Shelby paused for a moment and then said, "Maybe it's lactate then? Isn't that produced in skeletal muscle during exercise? Or maybe lactic acid and lactate are the same thing? My track coach in high school used these terms a lot so I think they might be interchangeable. He even made us do drills that he called 'lactates' just to make us sore so it's got to be one of these."

"Well, regardless," Patrick said, "I know lactic acid and lactate are only produced in muscle when it's being heavily used and runs out of oxygen. Maybe with training you won't produce these compounds at all and won't be sore after you run."

The conversation between Shelby and Patrick highlights several common misconceptions about skeletal muscle cells, the effects of training, and what causes muscle soreness. See if you can identify these misconceptions using the chart below:

Shelby's Misconceptions	Patrick's Misconceptions

To untangle the misconceptions in the conversation above, let's begin by taking a close look at the structures within skeletal muscle cells that are involved in energy production at a submaximal level of exercise in an untrained person. This would be the case for Shelby, who is just beginning an aerobic exercise program that includes mostly endurance training occurring in 20-30 minute bouts. In addition, let's examine some common metrics such as lactate threshold and blood pH to assess what is happening in an untrained person during exercise.

1.1 Blood, Skeletal Muscle Cells, and the Process of Glucose Uptake during Exercise

During the first 30-40 minutes of Shelby and Patrick's run at a submaximal level (40-70% VO2 max), blood glucose levels increase due to glucose release from the liver (Zinker, 1990), providing skeletal muscle cells with a concentration gradient of glucose for uptake and subsequent ATP production. However, glucose is a large polar molecule and cannot simply diffuse unaided through the phospholipid bilayer comprising the sarcolemma of skeletal muscle cells and into the sarcoplasm. Instead, to take up blood glucose, skeletal muscle cells must produce and insert glucose transporters, commonly referred to as GLUTs, into their sarcolemma. GLUTs are a diverse family of large, integral membrane proteins that permit facilitated diffusion of glucose down its concentration gradient. Skeletal muscle cells can express several different forms of GLUTs, but the most common form is GLUT4 (Gaster, Handberg, Beck, & Schroder, 2000; Goodwin, 2010; Richter, 2013). Interestingly, the insertion of GLUT4 into the

sarcolemma can be insulin-stimulated, meaning it relies on the receipt of insulin produced and released from the pancreas and activation of the insulin receptor on the sarcolemma, or simply contraction-stimulated requiring no insulin at all (Richter, 2013).

Activity:

1. Using the information in the text above, label all structures shown below in Figure 1.1, which represents a portion of a skeletal muscle cell's sarcolemma during exercise. Also label the extracellular fluid compartment, blood, and sarcoplasm. Indicate where in the body glucose and insulin would have come from and how they are transported to the skeletal muscle cell.

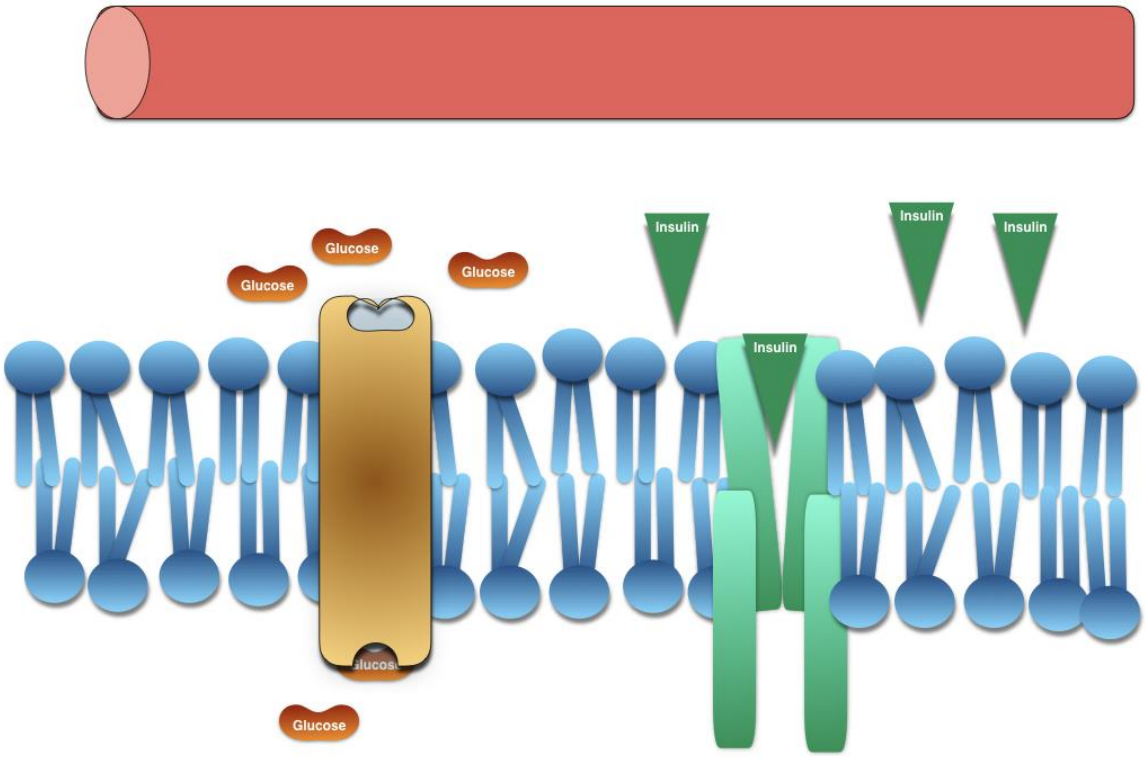


Fig. 1.1

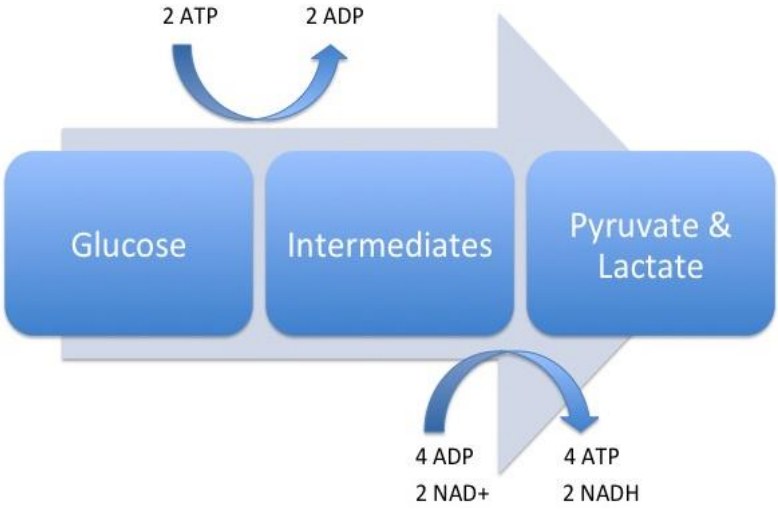
Questions:

1. Why does glucose uptake by a skeletal muscle cell require a transporter such as GLUT4? In other words, why isn't simple diffusion possible?

2. What stimulates the insertion of GLUT4s into the sarcolemma?

3. The existence of GLUT4s in the sarcolemma does not guarantee glucose uptake into the cell. Why? What else is required?

4. Exercise helps reduce blood glucose levels in people, even if they are insulin resistant, meaning their cells can no longer respond to insulin efficiently. How is this possible?



1.2 Glucose Utilization within Skeletal Muscle Cells

After glucose enters an active skeletal muscle cell it is first converted to an intermediate called glucose-6-phosphate by the addition of a phosphate from ATP. Not only does the creation of this new compound kick-start the process of glycolysis (breakdown of glucose), but it also prevents glucose from accumulating in the cell's sarcoplasm, which could slow or even stop subsequent diffusion of additional glucose molecules from the blood and extracellular fluid by reducing the concentration gradient that drives the diffusion of glucose *into* the cell. As the

Fig. 1.2

process of glycolysis continues, another phosphate from an additional ATP molecule is donated resulting in energized, yet unstable intermediates that will eventually yield useful products such as 4 ATP, 2 NADH, and 2 molecules of pyruvate. Many of the pyruvate molecules will ultimately be converted to lactate, the very compound that Shelby was most worried about when she began her running program. A simplified schematic of glycolysis is shown in figure 1.2. It is interesting to note that during glycolysis no carbons are lost from the cell and no CO2 is produced.

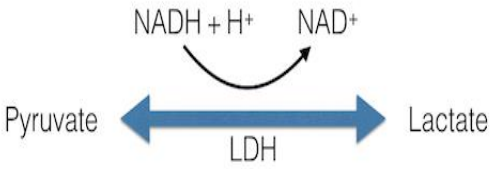


Fig. 1.3

The driver or catalyst converting pyruvate into lactate is an enzyme called lactate dehydrogenase, or simply LDH, which combines pyruvate with NADH and a proton (H+) to produce lactate while also regenerating NAD+. Figure 1.3 shows a simple representation of this reaction.

It is crucial to note that the processes described above occur in the sarcoplasm of muscle cells and do not require oxygen, even if oxygen is present in sufficient amounts. Furthermore, lactate is always an end product of glycolysis due to the presence of LDH, even if oxygen is present in sufficient amounts (Rogatzki, 2015). In fact, lactate is constantly created in fully oxygenated cells and potentially ten times more lactate than pyruvate is ultimately produced from glycolysis, even when the skeletal muscle cell has adequate access to oxygen and is at rest (Brooks, 2000). At one time, lactate was thought to simply be a cellular waste product or a dead

end to anaerobic respiration. We now know that isn't true at all and that lactate serves many useful functions for the cell in which it was produced as well as in other cells of the body. For example, as illustrated in figure 1.3, the production of lactate helps prevent pyruvate from accumulating in the sarcoplasm which could actually impede the production of pyruvate in the future, regenerates NAD+ so that glycolysis can continue, and reduces the number of free protons (H+) that could decrease intracellular pH. Furthermore, lactate can be converted back to pyruvate in mitochondria, which can then be used to fuel the Kreb's cycle. In fact, even lactate that 'spills' out of muscle cells and enters the blood is useful to cells of the heart, kidneys, and liver, which can use lactate from the blood for ATP production! Thus, the production of lactate should not be seen as a waste product, but instead as both a useful product and substrate.

Activity:

1. Using the detailed equation given below (Fig. 1.4), identify which compounds are substrates and which are products in the reaction catalyzed by LDH and the meaning of the bi-directional arrow. Explain why the conversion of pyruvate to lactate is beneficial to a cell and when this direction of reaction would most likely occur. Also, circle the two items that lactate "accepted" that makes its chemical structure different from that of pyruvate.

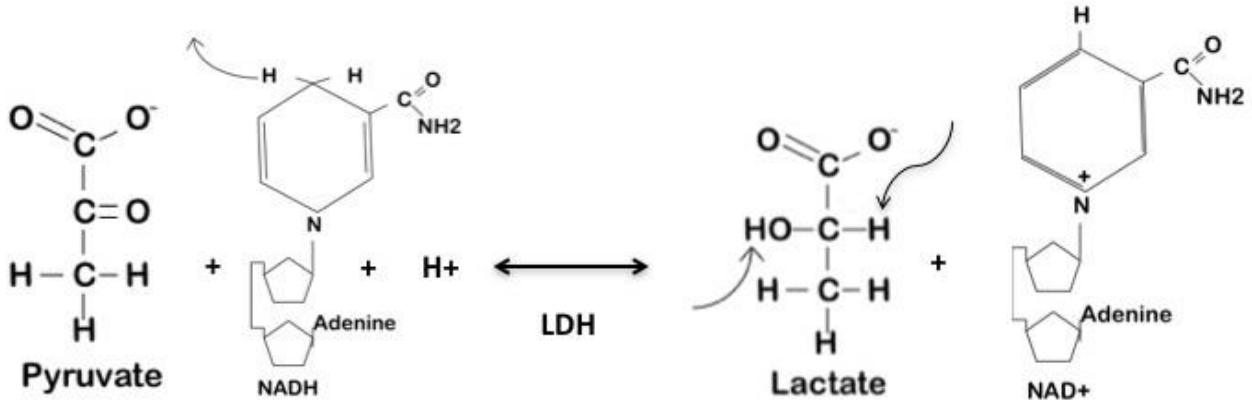


Fig. 1.4

Questions:

1. During glycolysis a molecule of glucose, which has six carbons, is split into two molecules of pyruvate, which have three carbons each (fig. 1.4). Each pyruvate molecule can then be converted into a molecule of lactate, which also has three carbons. Oxygen is not required for any of these processes to occur. Why?

2. After lactate is produced in the sarcoplasm, where might it go and how might it be used?

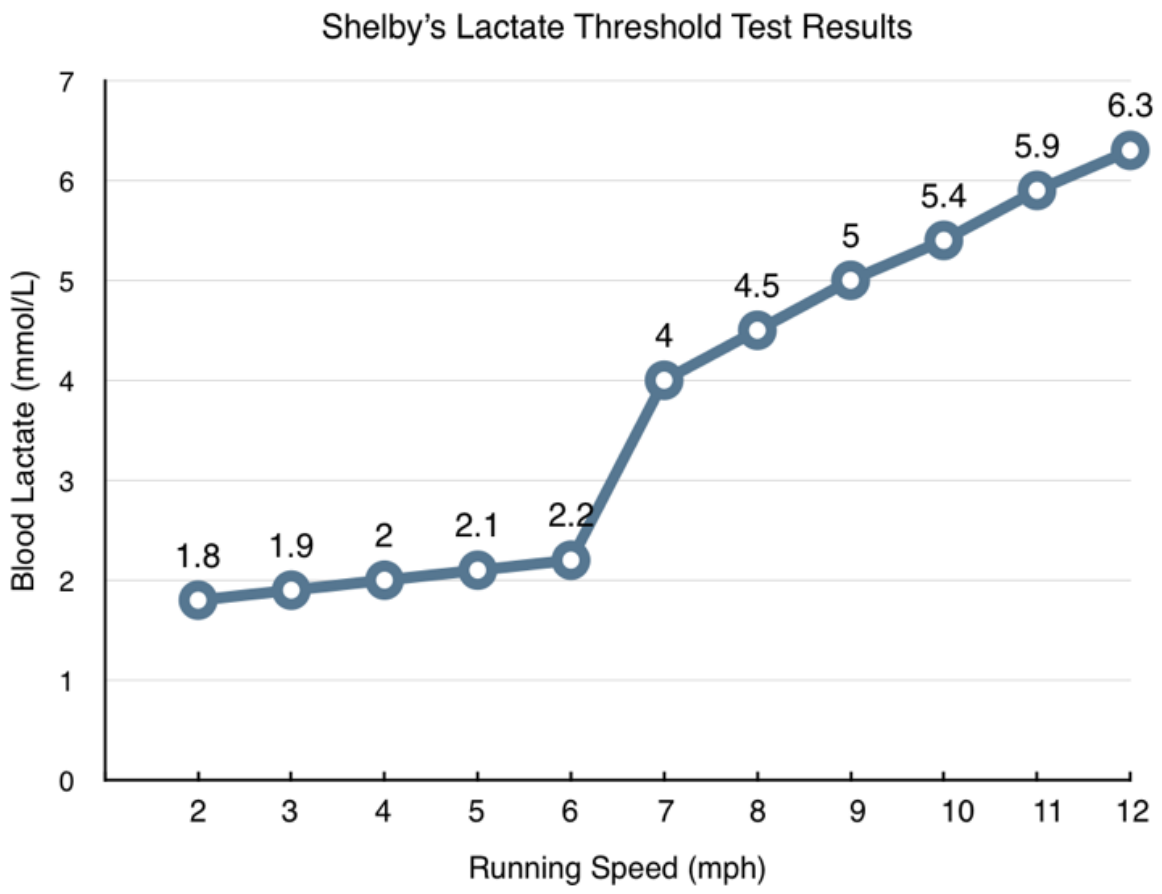
3. How does the production of lactate protect or buffer the cell from acidosis, which is defined as the accumulation of H+ in a fluid filled compartment?

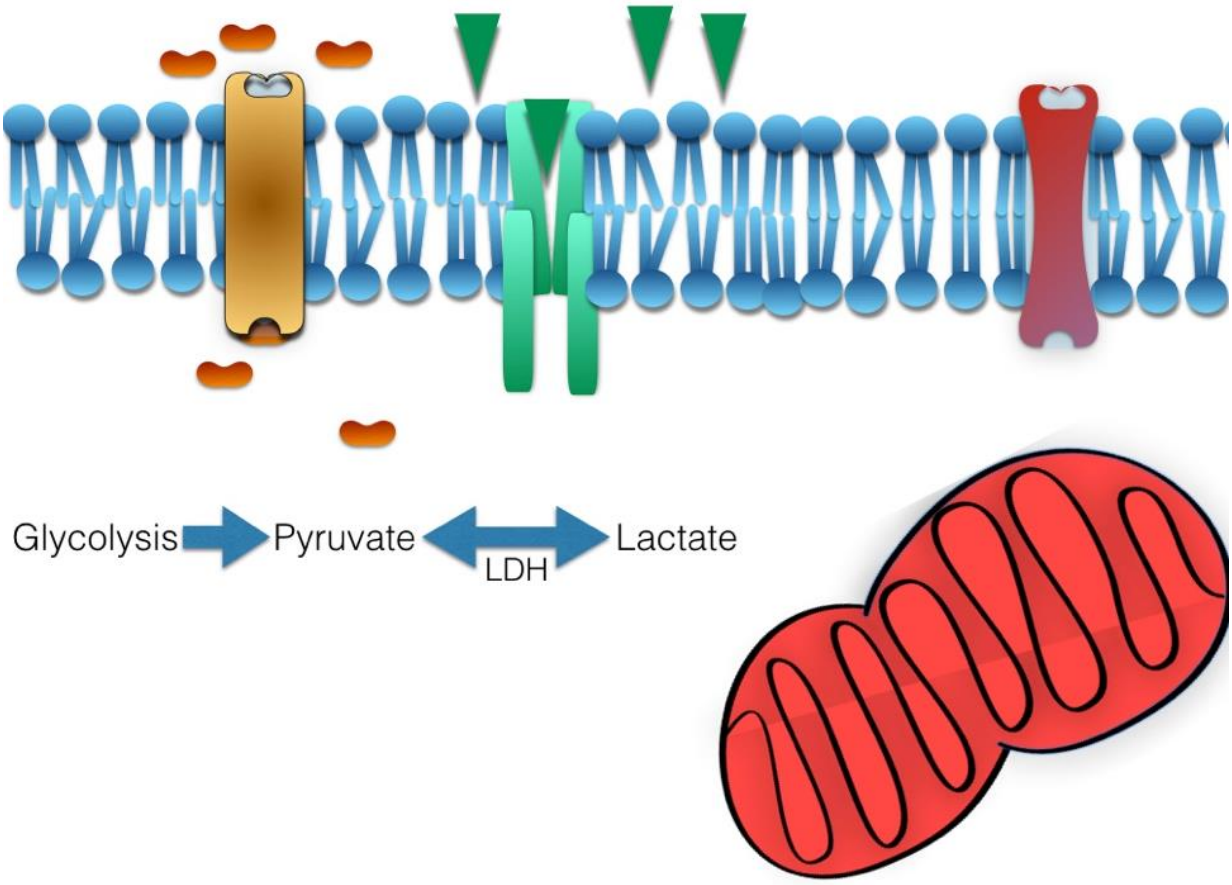
4. Look at the structure of lactate (fig. 1.4). Do you think it can leave the cell via simple diffusion? Why or why not?

1.3 Use of Lactate within Skeletal Muscle Cells

If you look closely at the reaction catalyzed by LDH (figs. 1.3 & 1.4) you will notice it has an arrow pointing in two different directions meaning the reaction is reversible and can occur in either direction; however, the characteristics of the LDH strongly favor the production of lactate from the substrate pyruvate. As a result lactate production can outpace lactate use in some cells, for example, untrained skeletal muscle cells used during submaximal endurance activities like jogging. In these active, untrained cells, the production of lactate far exceeds use; consequently, the lactate accumulates and diffuses or 'spills' out of the cell via a transporter and into the blood. One of the reasons untrained skeletal muscle cells cannot efficiently use the large amount of lactate they produce is that they do not have sufficient numbers of mitochondria to sustain them during an endurance or aerobic activities. Interestingly, it is the mitochondria that ultimately serve as the 'sink' for much of the lactate produced from glycolysis. Known as the Intracellular Lactate Shuttle mechanism (Brooks, 1999; Cruz, 2012), lactate produced by LDH in the sarcoplasm can enter the mitochondria where another isoform of LDH converts lactate back to pyruvate for use in the Krebs cycle, a process which ultimately results in at least 10 times more ATP production from glucose than that produced by glycolysis in the sarcoplasm. Thus, the more lactate produced and the more LDH available both in the sarcoplasm and within the mitochondria, the more ATP can ultimately be produced via aerobic respiration. In other words, lactate is ultimately used to fuel ATP production within the mitochondria aerobically, or in the presence of oxygen.

A common way of measuring the point at which lactate 'spills' out of working skeletal muscle cells and into the blood is the lactate threshold test. This test indicates the level of work (speed, for example) or exertion the amount of lactate produced in cells exceeds the amount of lactate that can be used either by the cells producing it, or by other cells in the body that have the ability to pull lactate from the blood for use as an energy source. As mentioned before, cells of the liver, heart, and kidney can use lactate for ATP production (Robergs, 2004). At rest, blood lactate levels remain fairly steady with production matching use. But when an untrained person like Shelby begins jogging, the production of lactate within skeletal muscle cells increases faster than it can be used causing it to spill into the blood, in turn causing blood lactate levels spike.


Shelby's Lactate Threshold Test Results
Blood Lactate (mmol/L)
0
1
2
3
4
5
6
7
1.8
1.9
2
2.1
2.2
4
4.5
5
5.4
5.9
6.3
2
3
4
5
6
7
8
9
10
11
12
Running Speed (mph)


Glycolysis
Pyruvate
LDH
Lactate

Activity:

1. Using the information above, indicate three possible fates of lactate in a skeletal muscle cell by labeling the structures and processes in fig. 1.5.

2. Examine the results of a lactate threshold test from an untrained person like Shelby who is just starting an endurance training regimen (Fig 1.6). Identify the area on the graph where the amount of lactate produced by skeletal muscle cells has overcome the ability of many of the body's cells to use it, thus causing it to accumulate in the blood.

Questions:

1. If lactate is a useful energy source for the cell in which it is produced, why does it 'spill' into the blood; in other words, why do blood lactate levels increase during exercise?

2. Through dedicated training, the speed or level of effort coinciding with a person's lactate threshold improves. Within the skeletal muscle cell, what changes might have occurred to permit this improved lactate threshold, resulting in less lactate spilling out into the blood?

3. LDH is an enzyme and thus a protein. In order for a skeletal muscle cell to increase the amount of LDH within the sarcoplasm, what cellular processes must be completed? What organelles need to be added to figure 1.5 in order for LDH to be produced?

1.4 Extracellular Fluid and Blood Composition Changes with Skeletal Muscle Cell Activity

As Shelby and Patrick run together during their first day of training, Shelby's legs begin to burn and her breathing becomes difficult while Patrick, who is in far better shape, doesn't feel the burning in his legs in spite of going at the same pace as Shelby. During exercise, extracellular fluid and blood composition changes due to reactions occurring within the body's cells. For example the amount of lactate diffusing out of skeletal muscle cells and accumulating in the extracellular fluid and blood increases, with lactate efflux and accumulation occurring more rapidly in untrained people like Shelby compared to trained athletes like Patrick. Blood pH can also change during exercise as H+ accumulates in skeletal muscle cells during extended periods of rapid contractions, eventually spilling into the extracellular fluid and blood, causing the pH to decrease in both of these areas if not adequately buffered. The increase of lactate and H+ in the extracellular fluid and blood occurs simultaneously as lactate produced within skeletal muscle cells is removed by a transporter, specifically the monocarboxylate transporter (MCT), which is embedded in the sarcolemma and was introduced to you in figure 1.5. This transporter functions as a symporter, permitting the diffusion of both lactate and H+ at the same time (Robergs, 2004).

For a century or more this simultaneous increase in blood lactate and H+ concentrations during exercise has perplexed scientists and it was originally believed that lactate production within skeletal muscle cells was the *cause* for increasing H+ concentrations because both lactate and H+ leave the cell at the same time and accumulate in the blood at the same time. As a result, it was also believed that the sensation of discomfort perceived by Shelby was due to accumulation of lactate. But as we've just seen, the production of lactate is not at all the source of increased H+ levels within cells or blood as the production of lactate is alkinilizing. So why

has lactate received such a bad reputation?

To answer this question, a very brief history is warranted for the terms lactate and lactic acid. In 1780 a Swedish chemist named Carl Wilhelm Scheele first discovered lactic acid in samples of soured milk (thus the term 'lactic' meaning or pertaining to milk) and isolated the compound using impure conditions. Despite some initial criticism of Scheele's work, other chemists of that period had also verified the presence of lactic acid in additional organic tissues such as fresh milk, meat, and blood (Robergs, 2004). While we now know that the presence of lactic acid - *and not lactate* - in many of these experiments was due to impure samples and bacterial fermentation, the term 'lactate' stuck and has been associated with the increased presence of H+ and resulting acidosis in working human skeletal muscle cells, extracellular fluid, and even in the blood ever since, regardless of the true cause. And once these terms became interchanged, correcting the issue has proven very difficult. In fact, even recent studies have mistakenly interpreted correlations illustrating the simultaneous increases of lactate and H+ in the blood during exercise as a cause and effect (Juel, 2004). But according to Robergs (2004) no experimental evidence has ever revealed a cause-effect relationship between lactate production and acidosis. In summary, this large and enduring misconception in the field of skeletal muscle physiology is due to improper scientific techniques that occurred centuries ago, continued misinterpretations of correlations depicting simultaneous increases in blood lactate and H+ levels, and the sustained use of lactate and lactic acid interchangeably.

Activity:

1. Using the information in the text above and your work in figure 1.5, complete the following graphic (fig. 1.7) by labeling all structures, including the sarcoplasm, extracellular fluid, and blood. Identify the monocarboxylate transporter (MCT) and indicate what items diffuse through this protein. Then indicate what changes would occur in the extracellular fluid and blood as products leave the cell through this protein.

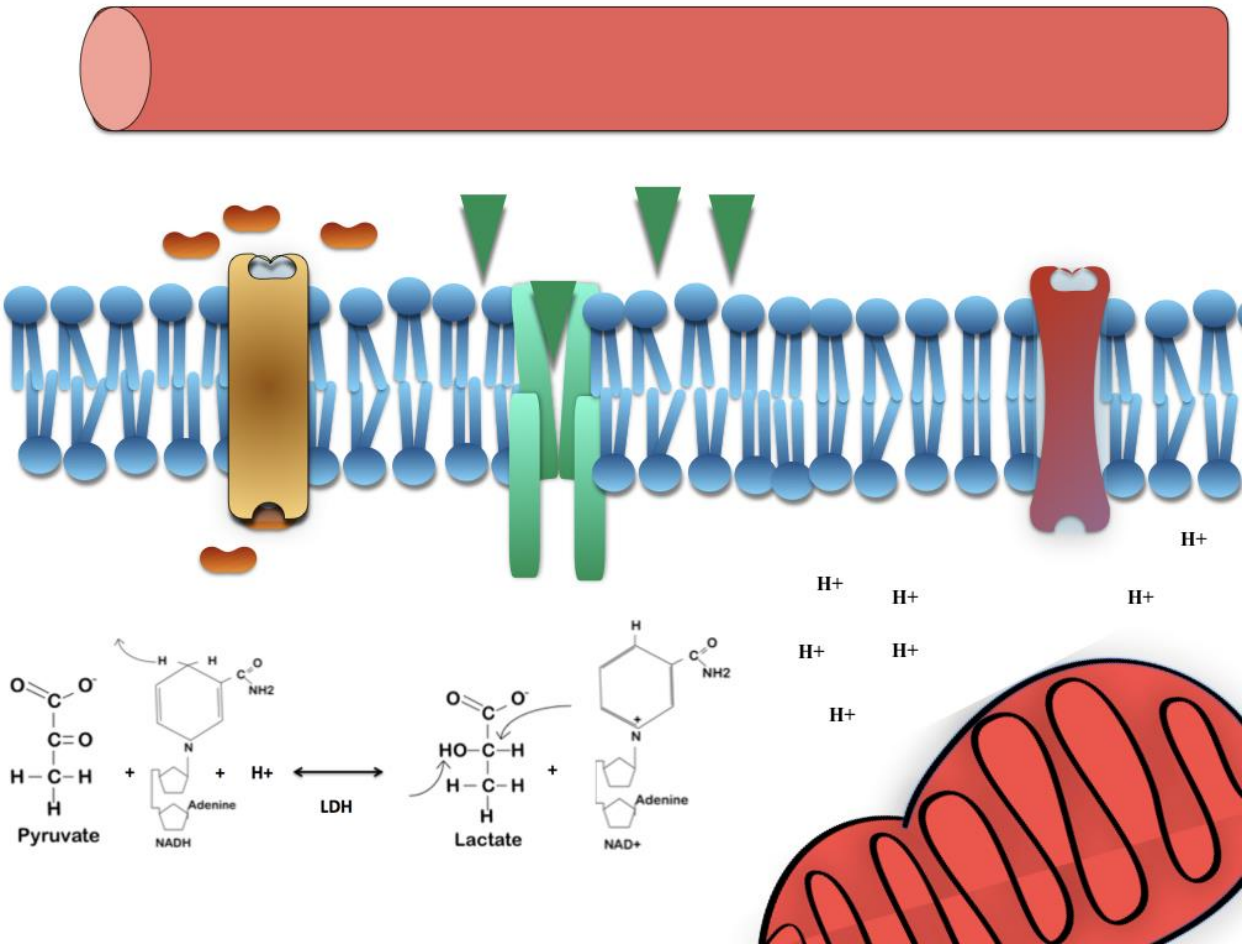


Fig. 1.7

Questions:

1. Suppose you are measuring changes in blood composition in real time in an untrained or unfit person like Shelby who is jogging at a moderate pace on a treadmill and you notice that as lactate levels increase in her blood, so do H+ levels. Why is this? Is lactate the original source of these additional H+ ions?

2. Hypothesize how Shelby's lactate threshold test would change with dedicated aerobic training. Specifically, describe the changes that should occur within skeletal muscle cells to delay the 'spilling' of lactate into the blood?

1.5 Muscle Soreness: The True Culprit

As you have probably noticed by now lactic acid and lactate, while often used interchangeably, are not the same. For example, lactic acid is a true acid, meaning it donates a proton (H+) while lactate is a proton acceptor, meaning it accepts H+ ions. Furthermore, lactic acid is most commonly produced during times of intense exercise when anaerobic metabolism is the only option for skeletal muscle cells and glycogen must be used as the fuel source. Lactate, on the other hand, is constantly produced by the cell during times of rest and during times of low to moderate intensity exercise, which is generally fueled by the diffusion of glucose from the blood and burned via aerobic metabolism (Westerblad & Lannergren, 2002).

But what about the painful after effects noted by many recreational runners like Shelby and even sometimes by trained athletes like Patrick after a hard training session? Interestingly, the cause of muscle soreness is likely caused by the accumulation of H+ in the extracellular fluid just outside of the sarcolemma. While the production of H+ can result from many different

reactions in the skeletal muscle cell, the production and hydrolysis of lactic acid to lactate yields a substantial amount of these ions. As H+ flows out of the cell via facilitated diffusion through MCTs in the sarcolemma it accumulates in the extracellular fluid and is thought to activate sensory neurons that convey information about the skeletal muscle cell to the brain (Westerblad & Lannergren, 2002). In other words, the accumulation of H+ in the extracellular fluid may cause extraneous afferent action potentials to be generated and sent to the brain where they are interpreted as pain.

Activity:

1. Using the information presented within this case study, complete the following graphic (fig. 1.8) comparing and contrasting lactic acid and lactate in skeletal muscle cells.

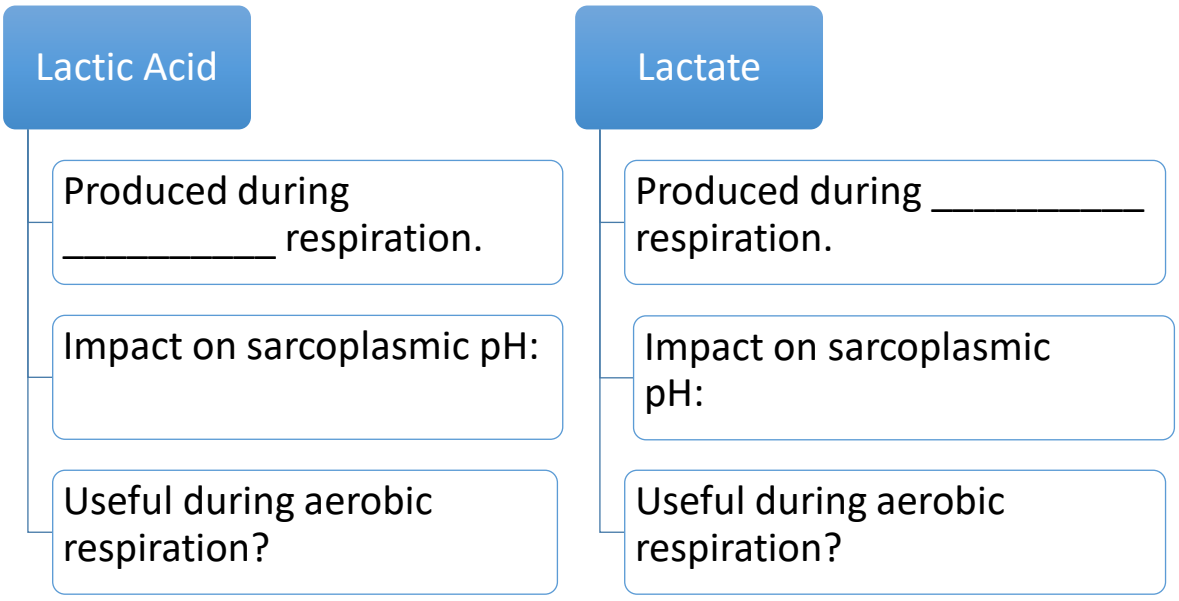


Fig 1.8

Questions:

1. Skim back through this case study and identify the causes of H+ production and accumulation in the sarcoplasm, extracellular fluid, and blood. Is lactate ever the original source of these ions or just guilty by association?

2. What is the relationship between the production of lactate and the burning sensation many people like Shelby describe after they begin a new exercise regime or activity? How is the brain made 'aware' of this situation?

3. Hypothesize what changes could occur within skeletal muscle cells that would help prevent the burning sensation after completing an aerobic exercise activity like jogging.

In spite of her initial discomfort, Shelby discovered that running actually was enjoyable and kept at it. Having Patrick to encourage her made a big difference as well, and she found herself thinking about metabolism of glucose and production of lactate within her muscles as she ran. As a result, she started getting better grades on exams and quizzes. After a few weeks she noticed that she could run with Patrick for the full 2 miles at a moderate pace without pain during the run or soreness the day after. Shelby called Patrick the day after they had ramped up their running distance to 3.5 miles, and she couldn't stop herself from excitedly gushing to her friend. "Patrick, this is amazing. I think what's happening is that I've gotten better at using lactate. I'm pretty sure it's because my cells have increased production of the LDH isoforms and have increased mitochondrial density, both of which allow me to use whatever lactate is produced much more efficiently so less spills out into my blood. What do you think?" After a long

awkward silence Patrick finally said, “Uh, I think I’ve created a monster, I was just hoping for a date.” Shelby gave a respectable pause and said, “Well thanks, Pat, but you’re a little slow for me now.”

Appendix Figure B.3. Individual Interviews

Interviewee #1:

A: Um, so yeah I'll ask you about 10 questions and we'll be done. Okay, so have you ever used a case study in a class before?

#1: Mhm.

A: What was it like?

#1: Um it was with human nutrition and basically like the whole class and all the tests were structurally like case studies, so I got really used to them with that.

A: Yeah.

#1: And I liked, I really liked learning like that way because it kinda like puts it into perspective like what I might be dealing with in the future because my major is dietetics and so that whole class is like well if this person has like this disorder then like how do you fix it and like what's wrong with them, so...

A: Right, right. So, what did you think about um this particular case study? Did you find it helpful or detrimental?

#1: I thought it was really helpful.

A: Did you?

#1: Yeah.

A: Cool, why?

#1: I think because I don't know (haha). I liked how it like gave us the uh, um boxes to write in like the steps of like...

A: You liked these flowcharts? (point to flow charts in case study)

#1: Yeah, mhm. The flowcharts were helpful I thought.

A: Okay. That's my next question, so cool you liked those. Um would you have made a flowchart on your own?

#1: I did actually.

A: You would have?

#1: Before yeah, like before...I think it, I like did that one....but then I like did one over here...(points to where she put the flowchart).

A: Um, cool! So you found the story to be helpful or not so helpful?

#1: Yeah, I thought it was helpful.

A: Really? Um, just because you could relate it back or why?

#1: I think yeah, it like made it like a story kind of, and so I was like looking at his past and then looking at him now it kind of like put everything into perspective I guess a little bit.

A: Yeah, into a real life...

#1: Mhm.

A: Cool! Um did you think that these like interactive figures were helpful? (point at interactive figures in case study)

#1: Yeah, I did. Especially like with the arrows I could see like these are going in and then out. And I could like draw arrows on my own and kind of, like the key was helpful to know like that was glucose and stuff.

A: Yeah, so do you think it was more like interacting portion, like you actually interacting with it or the finished product that helped you? Like...

#1: Interacting with it.

A: Yeah, the interaction?
#1: Mhm.

A: K, um did you find these like bolded words to be distracting or helpful?

#1: Um, I thought most of them were helpful...um, yeah I would say helpful. (hesitates)

A: Some of them...maybe...

#1: Yeah, like I didn't like know what t-tubules were. I feel like it was bolded and then...

A: Not explained?

#1: Yeah. (ha)

A: Good, good to know. Except it sucks because I've already published this, but that is a really good point.

(#1: laughs)

A: Um, yeah, um...do you know what they are now?

(#1: shakes her head no)

A: Okay, that's important. So, um the sarcolemma is just...um you know what that is right?

#1: Yeah.

A: The plasma membrane. Um so that's all that the t-tubule is, so t-tubule is the sarcolemma, it's just describing the invagination. So truly in a cell it would go do, do, do , do, do (moving finger up and down to show invaginations in the sarcolemma)...that's all it is.

#1: Okay.

A: Um, so what did you think about the length of the case study? Did you think it was too long, too short, or just right?

#1: Mmmm I kind of thought it was too long, but like in the end all of it was helpful I think so...I guess like just right but it took a long while to get through.

A: It seemed long, right.

#1: Yeah, mhm.

A: Um, so you would you have liked to discuss this in a group, or with the class, or did you like completing it alone?

#1: I like doing it along first, but then I would have liked going through it as a class at the end just to make sure I got it right.

A: Yeah I agree. I would like to discuss too, I like doing it alone and then seeing what other people got out of it.

#1: Yeah.

A: Um, would you have rather learned this information using this case study, using a YouTube video, watching a lecture, or um looking at like a multimedia; which is an animation with narration added to explain the information.

#1: Mhmm, not the lecture one.

A: K.

#1: Probably this still because I like like writing things out.

A: Really?

#1: Yeah (haha).

A: Good! This is so, I'm really surprised at how many people, almost every single student has said this.

#1: Really?

A: Mhm, and I was not expecting that.

(#1: laughs)

A: So that's, that's good. Do you have any questions for me?

#1: No.

A: No? Cool, well that was super short....

Interviewee #2:

A: Okay, so. Have you ever used a case study in a class before?

#2: I don't think so, no.

A: No? What did you think of this case study, did you find it to be helpful or detrimental to you?

#2: No I felt like it was definitely helpful, um, yeah it was kind of, it was a lot different than just like being lectured on something because like the case study actually like gave us the situation in which that could be applied to like real life.

A: And that helped too?

#2: Yeah I mean, IH'm a really visual learner so I feel like that kind of helped.

A: Really?

#2: Yeah.

A: So do you, you said you're a really visual learner, did you, uhm, would have preferred like Ashley's lecture or that?

#2: Uhm,

Since she's a pretty, you know...

#2: Well I mean, like Ashley's lecture is great because like with the picture like there were pictures on the case study and stuff but like when we go through Ashley's lecture like we're actually we're seeing it in real time and like yeah drawing it as the things happen so when we go back through and look at it we can kind of like, at least I remember better that like this happens first rather than if you just throw all that on the picture and like say this happens first time...I feel like as your drawing it you kind of learn at least I remember better like the sequential steps of which stuff works but

A: Yeah! Very cool. Good deal. Ummm. So did you find the information in the case study to be interesting or dull?

#2: Mmmmm so like what do you mean? Like like the actual

A: The science

#2: the insulin stuff? Ughhhh. I mean it's kind of dull to me

A: Yeah

#2: Like I'm a nutrition major, but I don't really like it.

A: Really?

#2: Yeah I'm in HN 400 right now and it's like all that type of stuff and I just...I don't know The stuff at the cellular level to me like I can learn it and its just ughhh it's not as interesting to me personally but

A: So the like cell signaling you're not interested in that but you're interested in nutrition in what way?

#2: I mean honestly I'm only a nutritional science major because I'm premed and that like covered a lot of my premed ughhh classes

A: Pre recs?

#2: Yea, yea, I don't know. I'm really into like fitness and eating healthy and stuff. So like in that aspect nutrition interests me but like at the cellular level, not so much...I mean I know I need to know it and it's good knowledge but to me it's just kind of dull

A: Yeah well good to know. Um did you find the story that guided the case study to helpful or not helpful to understand the information?

#2: Yeah definitely yeah

A: And why it matters

#2: Yeah the story, yeah for sure

A: How did that help you?

#2: Because if the story wasn't there I would have just gone to the questions and gone back to like the reading part of it like the information, not the story part but like after the story, we had like the paragraphs of information. I would have just gone through there and like picked out what I needed for that like to answer those questions but with the story it helped tie it all together

A: Good thats awesome. Did you find the flow charts to be helpful or not helpful?

#2: Yeah because I like when I study a lot I do like sequence events and I thought the flowcharts were awesome because I actually make those flowcharts on my own when I study

A: Really? Good, so do you think you would have written those out even if they weren't there?

#2: Yeah probably, yeah. I mean yea it just helps like when you have a big paragraph of information like if I go and take out the steps and write them out to the side like that in a flowchart or sequential steps like I don't know for me that helps me learn a lot better than like reading through a bunch of like crap.

A: Text?

#2: Yeah exactly

A: It's like this is blah blah blah what do I need to know

#2: Yeah I, I cause I only need to know the certain parts and I pick them out and then yeah, so I like the flow charts for sure

A: Good. Um did you find the interactive figures in the case studyto be helpful or not helpful to understand?

#2: Like with the are you talking about the channels and stuff?

A: Yeah the things you interacted with.

#2: For sure yea those are helpful.

A: And you mentioned you like drawing those in class anyways?

#2: Yeah yeah exactly and thats like just goes back to me being a visual learner because if you tell me like about the SGLT pathway and like what it does like that's great but unless I'm like drawing it out and seeing it, I won't remember it on the test. On tests I remember pictures really.

A: Right

#2: Yeah

A: Yeah

#2: I don't know I'm just like that's how I lean

A: Absolutely, I'm the exact same way, I can totally relate there.

A: Umm, did you find the bolded text to be helpful or distracting.

#2: Ummm I don't really remember which like what was in bold

A: Um so there were like certain bolded words like uh, uh esophagus and small intestine, did you find those to be distracting or did they sort of help you?

#2: Um those are for sure helpful once again because I'm a visual learner and like when I just see a paragraph of words

A: Woh woh woh

#2: Yeah it's hard to pick out stuff like even in like our lab manuals and stuff I have to go through and highlight those buzz words or like those important words. So like with them already being in bold, yeah that helped

A: K. Umm. What did you think about the length of the case study, did you find it to be too long, too short or just right?

#2: Ha, well I mean I wasn't really I had no idea what to expect going into that. Yeah, we don't usually, my group usually finishes up lab before the actual time is over so like I was in there for a while but I mean it it wasn't bad. Like I've definitely been through worse

A: So would you find it to be like just the right length for you to understand the information or do you think it should have been shorter and you still would have understood it or longer?

#2: I probably still could have understood it if it was a little bit shorter but I mean not everyone learns the same as me and not everyone is able to do that.

But, it was, it was okay.

A: Yeah, lots of people find it to be, that's the first thing they do is look through and they're like oh my gosh this is so long! How am I going to do this?

#2: Yeah it was it was a lot

A: Yea its pretty long so that's good to know

#2: It wasn't terrible

A: Um, would you have liked to discuss this in a group or with the class or did you like completing it alone?

#2: Umm a small group would probably have been more efficient at least.

A: You think?

#2: Yeah

A: Why?

#2: Just because like, I don't know, brain power. I mean put a bunch of heads together like and you can't I feel like it would have been more efficient in the fact that we would have gotten it done sooner, like we would have got through it sooner. Would I have learned it as in depth? Probably not. But it would have been faster to do it in a smaller group.

A: K

#2: But I feel like making me do it on my own and like going through and like actually learning that stuff as I go it definitely was more beneficial for like the posttest

A: So when you study do you study alone or in a group

#2:I actually study in a group

A: You do?

#2: But I do a lot on my own too. Ah eh, I kind do both

Both? you need both?

#2: I need both, yeah

A: Umm, do you think you would have rather learned this information through watching a Youtube video, listening to a lecture, or some sort of multimedia which is like animation with narration added to it

#2: Animation with narration probably just because like I'm a visual learner and like I yeah...

A: Yep

#2: I can, I can yea like I can like see when stuff...yeah probably either that or a youtube video but I like animations and stuff like that

A: So I guess uhh more specifically would you have rather heard what you were receiving or would you have rather read it like you did in the case study?

#2: Ah man, if I'm just hearing it without seeing anything I would I would honestly rather read it because if I'm hearing stuff I'm gonna have to write it down to remember it anyways so

A: But you would rather have both, hearing and seeing?

#2: Yes absolutely

A: K, well that's all my questions for you

Interviewee #3:

A: So, um have you ever used a case study uh in a class before?

#3: I don't think so.

A: No, never prior to this? Um, so what did you think of this case study, did you find it to be helpful or a detrimental tool?

#3: No, it was pretty good like you know understanding like with the help of someone else's example I think that helped

A: Did it? Okay, cool! Um did you find the information in the case study to be interesting or dull?

#3: I mean, ha, some of it was like really interesting, but then some of it was just like stuff I was just like what, what's happening?!

A: What's going on?!

#3: Yeah, ha

A: So, can you explain that a little more for me, what, what do you mean? Like some of it you found...

#3: Like some of it that was like related to like that person and everything was like interesting but there were bits of it that was like completely off. It was like: information, information, information! I was just like, STOP!

A: So much more! So, did you just think the information was just overwhelming or was it boring to learn about?

#3: I mean it was just a lot. But it wasn't boring cause like you know it was interesting, but it was like a lot of information all together, I was like "uhhhh"

A: So much to take in! So did you find the story that guided the case study to be helpful or not helpful to understand the information and why it matters?

#3: I think it was pretty helpful just like to understand like I said like the example like I know what this is, but like the fact that it's like relating it to like a person like how it works, I think that was like

A: That was helpful?

#3: Yeah

A: Cool! Um, did you find, uh, these flowcharts here, like this right here, did you find that to be helpful or not so helpful? This one too: (showed flowcharts)

#3: It was, it was pretty good I think, like just like getting together like everything that I read like just like making sure like I understand everything just like writing down everything

A: Kind of double checked everything for you?

#3: Yeah, yeah

A: So would you have made those flow charts if they weren't there? Do you learn this way? Do you, do you write things down in a sequence like this?

#3: Oh yeah!

A: You do?

#3: Yeah, yeah

A: Okay. K, um, did you find the interactive figures in the case study to be helpful or not so helpful to understand?

#3: Oh they were yeah, they were really helpful.

A: Really?

#3: Cause like yeah, I was like what side is that gonna be and what side...and then when I saw I was like okay yeah that makes so much more sense.

A: Oh good! Good! Okay, great. So, do you think that the interacting portion of this figure and the other figure, these graphics right here, (showed graphics) did you think it was the interacting part that was helpful or did you think it was the final product that was helpful?

#3: I think just like looking at it like okay so this opens here, this goes from here, like that was...

A: That was the most helpful?

#3: Yeah

A: K. Um, did you find the bolded text to be helpful or distracting? And so you know these like bolded words throughout the paragraphs, did you find that to be helpful or distracting?

#3: No, I mean they were pretty good like they were just...

A: Yeah? You liked that?

#3: Yeah

A: K. Um, what did you think about the length of the case study? Did you find it to be too long, too short or just right?

#3: It was a little too long (giggle)

A: Too long? Okay. So do you think that uh that information could have been covered in a shorter...um a shorter amount of time or a shorter length?

#3: I mean, it would have been better, it could have been, but I mean...but like I already feel like that was like so much information so like I feel like if it was shorter it would be harder to understand, you know? Cause it will be the same information like condensed into little...and then like what...

A: So you think it would be better to be presented in several parts than...

#3: Yeah, yeah I think so. And like maybe like related like more to like what happens inside the person like you know like there was like all this information like you know like in the beginning it was related and then there was like a ton of information...

A: So you didn't necessarily like that it was a story and then information? You didn't like that?

#3: Like I liked it, but then there was like bits that that was just like a lot just like a lot of information without like what's happening, like you know?

A: So you thought there was more information than story?

#3: Yeah, yeah.

A: Okay, okay. So you think it would be more beneficial to have more story interaction? Okay, um, uh would you have like to discuss this in a group, or with the class, or did you like completing this alone?

#3: I mean I think I like completing it alone but then maybe later we could discuss it after

A: Discussion after? Yeah that's what I would have preferred as well. Um, do you think you would rather learn this information through watching a YouTube video, listening to a lecture, or using multimedia? And multimedia is like animations with narration. What, what would you be your preferred method of learning this material? Would have rather used the case study, watched a YouTube video, listened to a lecture, or use like a multimedia animation interaction narration?

#3: Mmm, I think the multimedia one.

A: Multimedia?

#3: Yeah.

A: Yeah, I, I would have to agree. Why do you think multimedia?

#3: Just cause it would like explain it better cause that, that would be like a visual like with this like you know?

A: And listening and you can stop and pause and replay, yeah. So would you say you're a more visual learner or more audial, audible learner?

#3: Like I don't like reading, ha, but like um I mean so yeah visual but like I like seeing stuff happening in front of my like an animation so I learn better

A: Okay, well that's good to know! Well...

Interviewee #4:

A: Okay, so have you ever had a case study in a class before?

#4: Mmmmm, yes I have.

A: Yeah? What was it like?

#4: Um it was different, it was um with medical ethics kind of.

A: Oh, okay, in a medical ethics class or...?

#4: Um no, a pre-health application class, but...

A: Oh okay, um but it was like uh ya know a med ethics based...

#4: Right, yes.

A: Was is like this or very different?

#4: No, it was different, there wasn't as much interaction throughout.

A: Okay. Um, so what did you think of this case study? Um, did you find it to be helpful or detrimental?

#4: Um I found it to be very helpful.

A: Really? Okay. Um, did you find that the, the uh information, the science in the case study, to be interesting or boring?

#4: Uh interesting because it showed the application of the science in the case study which made it more interesting.

A: Cool! Yeah, um so, with that I guess this kind of leads into the next one, but so did you find the actually story of Timmy to be helpful or not helpful to understand the information.

#4: Um helpful, cause like I said like seeing the real life application of it like, oh so that's why that happens.

A: This is what this means, yeah.

#4: Yeah.

A: Um, did you find these flowcharts to be helpful or not so helpful? (show flowcharts)

#4: Um I did, because I liked at the end how they all went together.

A: You liked this last one? (show the last figure)

#4: Yes and it made you like think throughout and not just like read over the information, made you like like okay this is...(moving hands in a circle) but it was nice to have how many boxes, cause then I knew like...

A: What needs to, yeah, yeah

#4: Yeah

A: Cool, um did you find these like interactive graphics (show graphics) to be helpful or not so helpful?

#4: Uh very helpful, just cause I like, personally, I like seeing it cause read it and you're like wait, what side is where?

A: Yeah

#4: And so it's like okay so this is like ECF and these are these two pumps type of thing.

A: Good, um did you think it was more of the interacting part that helped you or the finished product that helped you the most?

#4: Um do you mean like the end?

A: Yeah, so like once you had everything labeled, do you think that was most helpful for you to reflect and look back on it or do you think it was the actual interaction with it, like labeling?

#4: Um, I mean there's benefits to both, probably for me it was the end just because I could see the process type of thing, and I was like oh like the capillaries like it goes into enterocyte first and then type of thing. So just like looking at it and talking myself through it.

A: Yeah, final product. Okay, so um did you find these like bolded text words, did you think that was helpful or distracting?

#4: I always like that because I feel like when there's so much text you can just find yourself reading and then when you see a bold word your like oh I need to pay attention.

A: Oh this is important?

#4: Yeah (hah).

A: Okay, okay. Um, did you think the length of the case study was uh too long, too short or just right for the information?

#4: I mean for me it was long enough to where I still have retained the information, like in nutrition class we're going over that and I'm like, I know just what this is type of thing.

A: YAY!

#4: And so if it was shorter, I don't know if I would have as well. But it did take time like you had to keep yourself focused.

A: Yeah, um would you have liked to discuss this in a class, or in a smaller group, or did you like completing it alone?

#4: Um, smaller group might be nice, but I do like doing stuff on my own because I can work at my own pace and I don't feel rushed or like I'm having to wait on people.

A: Yeah, yeah. So you, do you generally study alone or in groups?

#4: It honestly, it honestly depends. Cause groups are nice to where if I have a question or you can talk stuff out with people. But I like, I like doing it alone first to get the information my own way and then talking it over with other people.

A: Very cool. Um, so do you think you would have rather learned this information using the case study, watching a YouTube video, listening to a lecture, or using like a multimedia, which is like animation with narration that explains the information?

#4: Um, for me definitely not YouTube video, cause those are so interesting to zone out.

A: Really? Good, interesting.

#4: Um, lectures aren't too bad, but I did like how the case study related it back to real life.

A: Yeah, okay.

#4: And it kind of was a break up in scientific material parts. And the multimedia, is that kinda like YouTube? Like...

A: Multimedia would be like this (point at graphic in case study), and then like a, a person in the background narrating what happens.

#4: Okay, um, might be helpful, but only in little, little parts. Cause it's easy like I said if you get confused or lost, I just stop listening. (haha)

A: Yeah. So you think a case study would have been the best one then for you?

#4: Yeah.

A: Very interesting, very interesting. Cool! Well, that's all I have. Um, I appreciate your time today. Oh, I have a cookie for you....

Interviewee #5:

A: Okay, so. Ummm, have you ever used a case study in a class before?

#5: Uhhhm I think we used some in 310

A: Yeah?

#5: And they would even like be on our exams, like we would read it

A: What were they like?

#5: Um, I guess kind of similar, like a paragraph or two and we'd have to determine something about it, I can't remember too specific; but Dr. Pettay would like try to apply it more through using a case study I think

A: Was it like the one that we did in class the other day?

#5: Shorter

A: Shorter?

#5: A lot shorter

A: But it just gave you a scenario and

#5: Mhmhm

A: And you had to relate the science to it

#5: Yep

A: K, cool. Did you find the case study to be a helpful or detrimental tool?

#5: Very helpful

A: How so?

#5: I had very little understanding I felt like at least going in and then took the post-test and I like I don't know how I did, but I felt like I did decent

A: That's awesome! Yeah! Um, did you find the information in the case study to be interesting or dull?

#5: Uhm, I thought it was interesting. I thought it was cool how like at the end we had to put everything like completely back together and kind of like tie in both like little flowcharts that we had done.

A: So you liked that last uh connecting tool, yeah?

#5: Mhmhm

A: Yeah?

#5: Yeah

A: Really? That's good to know.

#5: Ha, have other people hated that?

A: Yes

#5: Really?

A: Lots of other people say, "I don't, I don't like this; I don't understand this." And it's kind of good that they are uncomfortable with it. It shows that they...well it's not good, it's good data for me to see that they're uncomfortable with it cause it sort of, that last figure is what shows whether you can connect all of these things that we've just discussed. So, it's good that you

#5: Right. Yeah, I thought it it made that's what made it make sense to me I think.

A: Really?

#5: I think...

A: That's awesome! Yeah, that's great!

A: Ummm, did you find the story that guided the case study to be helpful or not so helpful to understand the information and why it matters?

#5: Yeah, I thought it was helpful. It was like, it made it simpler because it wasn't just science talk.

A: Cool! So it made it make more sense because it was a real life scenario?

#5: I think so.

A: Awesome. Did you find the flow charts to be helpful or not so helpful?

#5: I thought they were helpful. I don't know if I did them like since there were so many blanks in a row sometimes I was like what do I put where but like overall just to like see the order of things I thought it was good.

A: Yeah! Would you have made your own flowchart otherwise?

#5: Mhm, no I don't, not necessarily like a flow chart but I don't know, I just probably would have done like I'm more of a paragraph I'm just like write all my thoughts.

A: Really?

#5: Yeah. That's more of like a graphic that I could probably use more.

A: K

#5: Cause, yeah. Cause it organizes it, so.

A: Yeah. Did you find the interactive figures in the case study to be helpful or not so helpful to understand this information?

#5: The interactive figures?

A: So those like graphs with the pictures of the small intestine and also like the skeletal muscle.

#5: Oh, yeah. Like just the diagrams kind of?

A: Uh huh, those diagrams.

#5: Yeah, I thought that was good. Just like seeing a picture kind of, the general way that it would work.

A: So that helped you? It didn't confuse you or...?

#5: No, I think I needed that or else it would have been very abstract.

A: Okay, good! All good, all good. Um, did you find the bolded text to be helpful or distracting throughout the, the text? So some words were bolded and some

#5: I don't remember what was bolded, oh just the....mhmmm

A: So like uhh mouth, esophagus, stomach, small intestine, enterocytes; key words were bolded. Did you find that to be distracting or helpful?

#5: Uhhh, no. I mean I don't think I would have been distracting. Mhm, yeah I mean most texTbooks probably do that to help you learn the big words.

A: Yeah, yeah. Umm, what did you think about the length of the case study? Was it too long, too short, or just right?

#5: Umm I mean it felt long, but I don't know how you would condense that....

A: Much material

#5: And I, I mean relatively speaking it was not like ridiculous. You just had to kinda sit there and make sure you focus for the whole thing.

A: Right, right. Good. Umm, so would have liked to discuss this in a group or with the class or did you like completing it alone?

#5: Ummmm probably...I don't know cause the other one was a lecture, right?

A: Right

#5: I may have preferred that just because

A: A lecture over a case study?

#5: Yeah, yeah just cause like even in bio 198 like teaching yourself I don't know sometimes you feel more unsure probably then if somebody is like literally telling you how it works

A: Right

#5: And taking notes on that. I'm also more I'm like my learning style is auditory which is weird cause most people are either like visual or I don't know what the other ones are but like if I if I just like listen to stuff that's how I

A: So that sort of brings me to the next question is do you, would you have rather learned this information through watching a YouTube video, listening to a lecture, or a multimedia; which multimedia is animation with narration.

#5: Yeah maybe that.

A: Multimedia?

#5: Yeah cause I mean that would tie in I guess everything that would help you to get it

A: So you would have liked seeing it but also hearing it instead of just reading?

#5: Right, yes.

A: K. And also, would you have preferred to interact with other people or do you would you have learned it primarily better by yourself?

#5: Umm probably better by myself unless it was like somebody teaching me through a lecture.

A: K

#5: Like that kind of thing is good cause I like asking questions cause like hearing stuff again like helps me but then like yeah I'm not a group project person I guess I don't know....usually

A: That's okay. Yeah, so you study alone primarily?

#5: Umm until I get it like initially I study alone until I feel like I can teach it to somebody cause otherwise I just get distracted and just start talking to people probably

A: Yeah, yeah, yeah

#5: So I study by myself at first.

A: Yeah, very cool. So umm you said you're kinesiology, pre-pt?

#5: Mhmhm.

A: Yeah, so you like the way Ashley draws everything out?

#5: Yes

A: You, so you are visual? Well, relatively

#5: Yeah, yeah I mean it like...I don't know to put it from like an idea to like oh I can now see how that works

A: No, yeah, that makes sense. Well, that was it! So, thank you so much for taking time to come do this with me.

Interviewee #6:

NOTE: We went through the entire interview once and the camera was not on, so we had to repeat the interview.

A: So, have you ever done a case study before?

#6: Uh yes, in nutrition class I did case studies.

A: And those were like this one or different?

#6: They were a lot shorter but...kind of the same.

A: K and what did you think of this case study?

#6: Um, I thought it was very helpful I think I learned a lot I think I got a lot out of it um...I think like interacting with it helped me understand the context a lot more.

A: Good. Um, did you find the information in the case study to be interesting or dull?

#6: Uh, I thought it was really interesting. I think that, blah this is the next question but, I think that you using the person; like high school, college, and then after like his results, I think that really helped.

A: So you liked the story?

#6: Yes

A: K. Um, did you find (sorry I have two alarms going off, we have a meeting)...um did you find the flowcharts to be helpful or not so helpful?

#6: I think they were helpful because you didn't really like have to remember everything at the end like you'd section it out in portions and go through the line and like it'd make you go back and like make sure you had everything sorted out in order.

A: K, um, did you find the interactive figures in the case study to be helpful or not so helpful to understand?

#6: I think they were helpful, I think that it separated the uh transporters and like the different everything out into pictures.

A: Yeah. So was it the interacting portion that helped you or the final product?

#6: Um, probably with the interacting.

A: Really? K.

#6: Probably.

A: Um, why do you think you'd say that?

#6: Um, because like you didn't have to just like remember it and recall it all at the end; you like, as you were going through it, you like broke it into sections and then at the end it was very helpful to like put it all together.

A: That last page? Really?

#6: Yeah, the last page. Because it was kind of like, I mean you like understood the components rather than staring at this huge thing trying to understand it all, you knew like the different sections and like if this breaks, that will break this and this and this...

A: Good! That's awesome to hear, that that was helpful. Um, did you find the bolded text throughout the case study to be helpful or distracting?

#6: Probably helpful.

A: Yeah?

#6: Yeah.

A: Um, what did you think about the length of the case study; did you think it was too long, too short or just right?

#6: Um, I think for the like context and everything it was just right. If it would have been too short you probably would have like missed something. It took a while, but I mean it was like just right for the context.

A: K, ummm, would you have rather discussed this in a group or with the class or did you like completing this alone?

#6: Um, I liked completing this alone, but that's just, I work better when I'm by myself.

A: Alone?

#6: Yeah.

A: Mhm. Um, do you think you'd rather learn this information through the case study, or watching a YouTube video, listening to a lecture, or like a multimedia; which is animation with narration?

#6: Um, probably, I would pro...like if I was doing completely like I had to take a test after it I would choose the case study; but just personally I might choose the multimedia animation just because it'd be probably like yeah more like, you wouldn't have to do as much and you'd still get something out of it. But I think that I probably learned more from the case study from multimedia.

A: Because it made you do something?

#6: Yeah.

A: Well cool! So do you have any questions for me? (We both giggle) Now that you're an expert....

Interviewee #7:

A: So, um, have you ever used a case study in a class before?

#7: Um, yes. I think we did in biology...I think it was just kind of like random stuff in high school we did but it wasn't, wasn't like that.

A: How was it different?

#7: Um...it wasn't as in depth as that, I don' know, I want to say like it wasn't in that much detail. Like it was over stuff that like I knew already.

A: Okay, so what did you think of this?

#7: I thought it was interesting because, well it was funny because when we did the pretest I was like, I feel like I should know more, but I really didn't know anything about the diabetes stuff and that made more sense after I actually did it.

A: After you did it? Good! Um...did you find the information, so the science of the case study, to be interesting or boring?

#7: I thought it was interesting when they...I liked when they actually talked about the person.

A: Did you?

#7: Yes.

A: You liked the story?

#7: Yeah.

A: Well that answers the next question...(ha) ummm did you think the story was, um, did it help you uh guide through the information?

#7: Yeah

A: Did it make it easier?

#7: Yeah

A: Really?

#7: And it helps like connecting because I, like I know so many people that's happened to.

A: Really?

#7: Just stuff like that, like high schooler metabolisms insane and then they get here and it's like

A: Yeah and then they're like, why am I getting fat?

#7: Yeah (haha)

A: Yeah, lots of women I've noticed more than men which is frustrating, but...um, did you find these uh, these flowcharts to be helpful?

#7: Yes I did. I got confused on this last one...I never (ha), I didn't know if I got it right or not. But they did help.

A: So, so uh this last one I added actually because a lot of people would put ECF/blood.

#7: Yeah, okay so just put blood.

A: But it's, it's confusing because this last one says increase blood glucose which is the result, but um...

#7: Okay that's what I assumed...

A: It is, it is confusing and now that I'm looking, I mean it's too late now because it's published but I wish that I would have put this in like a different box.

#7: That made sense though because I was like I think that's just the, just the end, the last thing...okay (head nod).

A: Yeah, as long as it made sense. Um, good! Um, so did you find these interactive figures to be helpful?

#7: Yeah because we draw them in class kind of, like that's what I, for the last test that's how I studied basically.

A: Was drawing?

#7: Was yeah, doing all those things on the white bored

A: Yeah, so could you have, I mean if you were to have just read these paragraphs, would it have made sense?

#7: (nods no) No.

A: No? You do need to see it?

#7: Yeah, I'm more visual.

A: Yeah, me too. So do you think that it was like the actually interaction portion that helped you or do you think it was the finished product?

#7: I think it was...you mean like the steps going...I think it was yeah the whole thing together.

A: The interaction or the very end that really, when it all made sense?

#7: The interaction more.

A: Yeah. So you actually labeling is what made more sense?

#7: To break it down a little bit, yeah.

A: Yeah. So if it, if this were to sort of just been given to you with the answers you don't think you would have gotten as much out of it?

#7: (shakes her head no and smiles)

A: No? K, umm did the bolded text, so like these bolded words (show bolded words), was that distracting or helpful?

#7: Helpful.

A: Helpful?

#7: Mhm.

A: It was? Good! You think you probably would have highlighted or underlined if they weren't...

#7: Yeah, cause that helps me in lab too like how they, it's kind of like how they bullet, I mean not bullet, they bold certain words. That helps.

A: Words. Yeah. What do you think of the lab manual?

#7: I like it.

A: Do you?

#7: I like, I yeah. I like how it's not super long, but it's like all of the information you need to know. Like exactly everything you need to know.

A: Yeah.

#7: Like exactly everything you need to know. (Haha)

A: Yep. Mhm. If you memorize the book, you'll get a good grade.

#7: Literally, that's all you need to do. (Haha)

A: Yep, yep. Um, so what did you think about the length of the case study? Did you think it was too long too short or just right?

#7: I think it was just right. I was pretty tired that day (haha), but I think it was just right.

A: Yeah, for the amount of information?

#7: Yeah.

A: Yeah, so you wouldn't have changed it?

#7: I don't think so.

A: Would you have liked to discuss this with in a group, or with this class, or did you like completing it on your own?

#7: I think I would have have done it by myself and then gone with a group of....

A: Yeah, that's what I would have done too, because um then you can bounce ideas off but you get to interpret first.

#7: Yeah

A: Um, okay and then do you think you would have rather learned this information through the case study, or watching a YouTube video, or listening to a lecture, or uh like some sort of multimedia which is like animation with narration that explains the information.

#7: Um....(pause) can I pick two?

A: Yeah!

(haha)

#7: I was going to say, probably the case study and then the animation thing because I feel like that would make sense, kind of helping with the figures a little bit.

A: So like both?

#7: Yeah

A: So, you, if you had a choice, you would have like a story and then uh, a multimedia to explain things?

#7: Yeah, I think so. I don't think I would pick lecture.

A: K. No? Why not?

#7: I would get really side tracked I think.

A: Really?

#7: Yeah

A: Aaand side tracked, you mean you wouldn't understand the material because you would be like kind of squirrel mode or...?

#7: Yeah. Especially if it's not like right in front of me.

A: Woah! That was uh maybe a record timing. (haha)...

Interviewee #8:

A: Okay, so have you ever used a case study before this class?

#8: No, I don't think so.

A: No? And did you find the case study to be a helpful or detrimental tool?

#8: I thought it was helpful.

A: Why is that?

#8: Umm, it kind of applied the concepts to real life situations especially when we were learning about diabetes and stuff like that, it helps understand it.

A: Yeah, cool! Um, did you find the information in the case study to be interesting or dull?

#8: I thought it was interesting, there were parts that were kind of just like mehhh (rolled eyes and shrugged shoulders), but overall it was interesting.

A: So, when you say interesting did you find like the science interesting or the story or....

#8: I found the science interesting, the story kind of helped put it in perspective but...

A: But it was like the science that was actually like interesting? (Mackenzie shakes her head yes) Cool! Um, did you find the story that guided the case study to be helpful or not helpful to understand this information and why it matters?

#8: I thought it was helpful.

A: Helpful? You pretty much said that.

#8: Yeah, like I said just real world application helps a lot.

A: Right, right, um, okay, so did you find these flowcharts to be helpful or not so helpful? (Abi shows Mackenzie flow charts).

#8: I thought they were helpful and then when at the end we combined the two flow charts, I liked that.

A: You liked this? Really? So you liked the application here?

#8: Yeah.

A: Why did you like it?

#8: Uhh it helped tie, it kind of just helped me like concept check; cause I'm really bad about when I read stuff I'm like "oh yeah I got it, it makes sense" but then when you actually have to go back and like fill out the flowchart it makes me go, "I actually don't understand so let me go back and read this."

A: Good! That's good to hear. Um, did you find the interactive figures in the case study to be helpful or not so helpful in understanding the information? So by interactive figures or diagrams I mean like this guy: (show Mackenzie graphics)

#8: I thought those were helpful.

A: You did?

#8: Yeah cause when you're reading the paragraph it was a little confusing about like this is moving from the lumen into the cell and from into the cell you know to the other side so then that kind of helped

A: Yeah, totally agree. Um, so do you think it was the actual interacting portion that helped you the most or the finished product that helped you understand it?

#8: Probably the finished product.

A: Finished product?

#8: Yeah

A: Cool! Um, did you find these uh bolded words, bolded text (show Mackenzie bolded words on the case study), did you find that to be um distracting or helpful?

#8: Helpful.

A: Really? Good! Um, why do you think was helpful?

(Mackenzie shakes her head, looking sort of confused by the question)

#8: Just key...that kind of stuff

A: Terms? Would you have done that even if they weren't bolded? Would you have like highlighted those words or underlined them?

#8: Probably, but I'm sure if they weren't bolded, some of them wouldn't stand out as important. Like, I don't know if t-tubules, if that wasn't bolded I don't know if I would be like "oh t-tubules, they're important" that kind of thing.

A: Yeah, yeah. Um, what did you think about the length of the case study? Did you find it to be too long, too short or just right?

#8: It was a little long, it took me a while to get through, but...

A: So, um, would you have rather had a shorter case study or would you have rather had it broken down into you know several parts?

#8: Mhmm...I don't know, I don't know how much you could really shorten it because there's like a lot of information that you have to cover, so maybe just broken down into parts.

A: Yep, yep, I agree. Um, would you have liked to discuss this in a group, or with the class, or did you like completing this alone?

#8: I liked doing it by myself, I like to work individually.

A: Did you? So you, you do? Okay. So do you study alone?

#8: Yeah

A: Yeah, very cool. Um, do you find that other people sort of impact you in a negative manner when you study with them or...

#8: Yeah it just kind of depends who it is, some people they can better my understanding, but sometimes I feel like it I don't know you focus on different things and it turns into a distraction.

A: Yeah, um do you think you would have rather learned this information through the case study, watching a YouTube video, listening to a lecture, or watching a multimedia; which would be like animation with uh narration.

#8: Um...I liked the case study but I've, I don't know...when I was studying for the MCAT I used a lot kind of like that animation thing like Kahn academy where they walk you through it and kind of lecture it kind of video thing, and I really like those; but I did like the case study.

A: Did you? So you like both?

#8: Yeah

A: So you wouldn't say that you're particularly an audible or a visual learner? You're a mixture?

#8: Yeah, probably. A mixture.

A: Yeah? Cool! Well, that was my last question. That was super short and sweet, you only....

Interviewee #9:

A: Okay, so, have you ever had a case study in a class before? Have you ever used one?

#9: Um, I believe so.

A: Yeah? What was it like?

#9: Um, I think it was a...kind of a medical case study. It might of been like biology class and we just did something like that. I can't quite remember all of it.

A: That's okay. Um, well what did you think of this case study?

#9: I thought this one was way better.

A: Yeah?

#9: Especially because it kinda gave you a little synopsis and like explained it and then applied it and you could actually see it working. So it wasn't just like um like you're given the information and you have to go see how it works, it kind of gives you examples. I thought that was way more helpful.

A: Good! Good. Um, so did you find the information, so the science in the case study, did you find that to be interesting or boring?

#9: I actually thought it'd be kind of interesting because I never, I always had so much trouble understanding like that specific, like insulin resistance which leads to this to that, I think that case study actually like helped me understand it better.

A: Really?!

#9: And, um, like even I think when we took the pretest I went home and I kinda like googled a little bit of it... (smiled)

A: Researched it? Did you?

#9: Yeah, cause I just, that always confused me and I didn't know how it starts or what it can lead to and how it can like this part lead to that part (moving hands and directing a path), it was nice.

A: Yeah so when you say um it always confused you, did you mean like glucose regulation or you know disease states, like you didn't know what type II diabetes was?

#9: Like insulin, like the difference between insulin resistance and then like hyperglycemia and then like how those led to each like type I and type II.

A: Diabetes. Yeah, yeah very cool. Um, did you find the story that guided the case study to be helpful or not helpful to understand the information and why it matters?

#9: Um, it was probably more helpful because you didn't see it on just like the cellular level, you saw the effects that it had to the person. So like he gained weight and he had a slow metabolic rate and stuff like that and like how it progressed over the years. So like seeing it as a person it was also kind of culture shock to also not do that to yourself see how that could happen (she smiles).

A: Yeah! That's so cool to hear you say that. So um, you like that you know it looked things both from a realistic level like reality and then also molecularly?

#9: Yeah, yeah a realistic level is a good way to put it.

A: Very cool! Good. Um, so did you find uh these flowcharts (show flowcharts) to be helpful or not so helpful?

#9: Extremely helpful (points and smiles).

A: Really?

#9: Yes.

A: Good!

#9: Probably cause whenever Rhodes is like you need to make a sequence of events and you gotta break it, ever, I never did that for any other like way to study and so that made sense probably because that's like how we're learning in our class right now, and so that was a good way to see it like as it progresses.

A: So, prior to Ashley's class, you never made flowcharts?

#9: Not that in depth, I would make flow, like

A: Simple?

#9: Yeah like simple ones, but like not each like every single step (smirk). Because you don't really realize all the steps that are going on there.

A: No, and you don't realize like even the little step, if that breaks, that can effect something else. Um cool, so did you find these interactive figures to be helpful or not so helpful? (show interactive figures)

#9: Um it was hard at first just to um just cause I was like uhh what the...(waves her arms in the air with confusion).

A: What is this?

#9: Uh but the instructions part showing it and then like when it had you label the arrows so that you could try to figure it out I liked just how interactive it was so then it was like you're learning it but then if you would read the paragraph after you'd be able to like check your work to kind of see that

A: Yeah, so you think it was more helpful, the interactive part more than the finished product? What helped you understand it more?

#9: Um, yeah the interactive I guess the interactive helped me yeah it helped solidify what I knew what was going on.

A: Yeah

#9: Um, because like I wouldn't have been able to just like look at that picture and know what's moving in and out of where.

A: Yeah, cool. Very cool. Um, uh did you find these like bolded texts in the paragraphs, did you find that to be distracting or helpful?

#9: Um, probably more helpful just because just like the way I've read most...

A: TexTbooks?

#9: TexTbooks, yeah. It was, it was nice to um I would uh like scan over that word because I like it if it's also in italics too just so I know I can like connect that bolded word with what's around it and then just like relate it to whatever else. And then it would also help me not get certain definitions confused with the other ones.

A: Mhm, cool. Um, so what did you think about the length of the case study, did you think it was too long, too short or just right?

#9: I probably felt like it was kind of long just cause I was in there for a while, but I thought that every part was like a fairly good amount, like the page wasn't just straight like reading and um I didn't think it was, it definitely wasn't too little, but I thought it was like just the right amount. Cause it wasn't like too much to where I was overloaded, and it was, it wasn't too little to where I didn't know what I just read. So I thought it was a good amount.

A: Yeah, good. Good. Um, would you have like to discuss this in a group, or with the class, or did you like completing it on your own?

#9: Um, I liked completing it on my own. I think if, it's nice talking in a group cause then you can hear everybody's um things, but sometimes that can either help or hurt you if helps, but I think doing it by myself cause then I know that the correct answer is in the reading and I can check it with myself.

A: Yeah, so you would rather do it alone at first and then maybe...

#9: Yeah.

A: Yeah. K, that's what I would like to do as well. Um, do you think you would rather learn this information using the case study, or watching a YouTube video, listening to a lecture, or a multimedia, which is defined as an animation with narration that explains the information?

#9: I probably wouldn't want, I wouldn't want listening to a lecture cause I need, I think I'm more of a visual and an auditory learner; um and then also writing. So I think the um, the multimedia one would be kinda cool. Cause I'll go, I'll go home and look up YouTube videos to help with Ashley's lectures or something.

A: Really?

#9: Yeah cause it'll be helpful to like, I'll listen to her lecture, take those extra notes, and then I'll look at the YouTube videos just to be able to see what she's talking about instead of just like that 2D picture.

A: Yeah.

#9: So I think the multimedia would be cool.

A: Cool! That's really good feedback, um I wish I had the time to design one. Um, cool, so do you have any questions for me?

#9: No...what are you guys doing with the case study?

Interviewee #10:

A: Okay, um, so have you ever had a case study in a class before? Did you ever use a case study before?

#10: Yes in ecology.

A: Okay, what was it like compared to this one?

#10: Um, very similar, it um I mean obviously dealing with ecology and just it didn't have like as much of a like personal um aspect to it. It was very just like here are the facts and like stuff that we got. It wasn't like a personal story like Timothy or whatever.

A: Yeah, very cool. Um, did you find the case study to be a helpful or detrimental learning tool?

#10: Helpful, definitely.

A: Why is that?

#10: Um, so like even when I like go through my notes from lecture I kind of do the same thing of like I make flow charts, I kind of go through like the process of like what kind of questions could be asked on this material. And so, I naturally kind of go through that same process, and so when I have it you know already written down and already kind of the steps going out of it that kind of just helps guide that along I guess.

A: Yeah so it was like a guide of what you've already done.

#10: Yeah

A: Good! Um, did you find the information in the case study to be interesting or dull?

#10: Interesting. I liked the fact that it had that personal element of like there was an actual person and kind of gave back story without being like okay like this is kinda weird but...

A: Yeah, mhm, cool! Um, so you actual liked the science part or did you more so just like the story part?

#10: Eh definitely like the science part but um, but like learning the facts and like the figures is a lot easier I guess to take in and more like, it almost felt more important especially going into healthcare its like this is going to be something I deal with everyday so it's kind of nice to have that.

A: Yeah, that's awesome to hear you say that. Um, did you find the story, you kind of already said this, to be helpful or not helpful to understand?

#10: Yeah it definitely aided in it.

A: Yeah, did it? Um, so did you find those flowcharts helpful? The flowcharts where it was like, you know, mouth, stomach...

#10: Mhm.

A: You liked that?

#10: I did like those except for on, I think it was the second flowchart, I can't remember exactly what it was, I think it was like the absorption one of those...but um

A: Yeah, I can grab one

#10: I think it's just like the way my thought process was I think a little bit different so I had to add one more box to it, but other than that yeah it was definitely help guide, especially

Abi hands the case study to Rachel

#10: It was this one, I had to like add a box like right here or something

A: But it made sense for the most part?

#10: Yes, uh huh.

A: So what did you think about these figures here? Did you find those to be helpful?

#10: Yes, um just I'm a very visual learner, so definitely any time you have um a graphic like that and I especially liked learning the information like so reading it first and then seeing the graphic.

A: You like that order?

#10: Mhm, that's just the way I guess I learn the best um because then you can kind of take the information you know and kind of see like what you can like put on the graphic yourself.

A: Yeah, yeah. Very cool. Um, well good you're just flying right through this. Um, was it the interaction that helped you the most or the final product? Did you find um actually interacting with the tool more helpful or the, the final product and like reviewing it?

#10: Um, I think interacting.

A: Interacting helped you most?

#10: Mhm.

A: Um, did you find the bolded text, so like the bolded words, did you find those to be helpful or...

#10: Yes, definitely because I feel like filling out the flowcharts is kinda like the words that you actually used in that, but um I could see how like if this was handed out to a class it would be very easy for someone because of those bolded words to kinda just like do it absent mindedly

A: Fill them in

#10: Yeah. If you're engaged, it was helpful

A: Yeah but it can also be enabling right

#10: Mhm

A: Um, what did you think about the length of the case study? Was it too long, too short, or just right?

#10: Umm, I mean it was kind of lengthier but I don't see how it could be you know any shorter to get that information in. If you think of it in like terms of you know that's a whole lecture, so 50 minutes of a lecture compared to I mean to like 20 minutes or so to read through that, I think if you put it in those terms, it's definitely the right length.

A: You think so?

#10: Mhm.

A: Um, would you have liked to discuss this in a group, or with the class, or did you like completing this alone?

#10: I like completing it alone just because I work better alone um and just kind of have my own thought process and I like wrote notes out to the side and stuff. But I think it would be beneficial to kinda like at the end maybe gather as like a class and like go over it or just seeing important details just cause it brings in different perspectives or you know different details you might have missed.

A: Absolutely, I agree. Um, do you think that you would have rather learned this information through watching a YouTube video, listening to a lecture, or watching some sort of multimedia, which would be like an animation with narration.

#10: Umm, I definitely like this the best, I feel like the YouTube videos or like the graphic narration things, I feel like those are helpful tools to like aid me in like after I know the material and like kind of watch and make sure like I thought of the information in the same way umm, but like I kinda said earlier like if when I have lecture and I go through the notes, I kinda do the same thing afterwards so this kinda almost, not took out a step, but was a little more direct in how I learned the material.

A: Which you liked, you preferred that? It helped you?

#10: Yes

A: So you would have rather had the case study over a lecture?

#10: Mhm

A: And you would have rather had like multimedia or a YouTube video over the case study, if you had to like group those?

#10: I think case study first.

A: Really? Okay!

#10: Mhm, that's just the way I learn.

A: That's, no that's great! I mean everybody learns differently, so this is um all good, all good stuff. Well, do you have any questions for me?

#10: I don't think so!

A: Well that was short and sweet and I appreciate your time!