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THE EFFECTS OF ULTRA-FILTERED MILK CONSUMPTION ON STRENGTH AND PERFORMANCE FOLLOWING RESISTANCE TRAINING IN FEMALE COLLEGIATE ATHLETES

David A. Ferrer Georgia State University

Jeffery S. Otis Georgia State University

Brett J. Wong Georgia State University

Andrew J. Doyle Georgia State University

Sarah T. Henes Georgia State University

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ACCEPTANCE

This dissertation, THE EFFECTS OF ULTRA-FILTERED MILK CONSUMPTION ON STRENGTH AND PERFORMANCE FOLLOWING RESISTANCE TRAINING IN FEMALE COLLEGIATE ATHLETES, by DAVID A. FERRER, was prepared under the direction of the candidate's Dissertation Advisory Committee. It is accepted by the committee members in partial fulfillment of the requirements for the degree, Doctor of Philosophy, in the College of Education and Human Development, Georgia State University.

The Dissertation Advisory Committee and the student's Department Chairperson, as representatives of the faculty, certify that this dissertation has met all standards of excellence and scholarship as determined by the faculty.

Jeffrey. S. Otis, Ph.D. Committee Co-Chair Brett J. Wong, Ph.D. Committee Co-Chair

J. Andrew Doyle, Ph.D. Committee Member Sarah T. Henes, Ph. D., RD, LDN Committee Member

Date

Mark Geil, Ph.D. Chairperson, Department of Kinesiology and Health

Paul A. Alberto, Ph.D. Dean College of Education & Human Development

AUTHOR'S STATEMENT

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David A. Ferrer Department of Kinesiology and Health College of Education & Human Development Georgia State University Atlanta, GA 30303

The director of this dissertation is:

Jeffrey S. Otis and Brett J. Wong Department of Kinesiology and Health College of Education & Human Development Georgia State University Atlanta, GA 30303

CURRICULUM VITAE

David A. Ferrer, MS, EP-C, CF-L1

ADDRESS:	Department of Kinesiology and Health
	College of Education & Human Development
	Georgia State University
	Atlanta, GA 30303

EDUCATION:

	Ph.D.	2017	Georgia State University
	Master's Degree	2012	Kinesiology Georgia State University Exercise Science
	Bachelor's Degree	2009	Georgia State University Exercise Science
PROFESSI	ONAL EXPERIENCE:		
	2016-2017		Crossfit Level 1 Coach Crossfit AGX/Airo Chamblee, GA
	2014-2016		Volunteer Coach Strength and Conditioning Department of Athletics Georgia State University Atlanta, GA
	2012-Present		Part-Time Instructor Dept. of Kinesiology and Health Georgia State University Atlanta, GA
	2011-Present		Graduate Research Assistant Exercise Psychology Laboratory Exercise Physiology Laboratory Dept. of Kinesiology and Health Georgia State University

Atlanta, GA

PRESENTATIONS AND PUBLICATIONS

Ferrer, D. A., & Ellis, R. (2017). A Review of Physical Activity Interventions Delivered via Facebook. Journal of Physical Activity and Health, 1-32.

Ferrer, D. (2016, January). *So You Want to Get Fit?* Invited, guest speaker for the Tighter Grip E-Board. Georgia State University, Atlanta, GA.

Ferrer, D. & Ellis, R. (2015, April). *Using Mobile Apps to Promote Exercise Motivation and Behavior Among Adult*. Paper presented at the Kinesiology and Health Research Symposium, Georgia State University, Atlanta, GA.

Ferrer, D. & Ellis, R. (2015, February). *Using Mobile Apps to Promote Exercise Motivation and Behavior Among Adults*. Poster Presented at annual meeting of Southeast American College of Sports Medicine. Jacksonville, Florida 2015.

Ferrer, D. and Ellis, R. (2015). Using Mobile Apps to Promote Motivation and Behavior Among Adults. Dept. of Kinesiology & Health, Georgia State University, Atlanta, GA 30302. *SEACM Annual Meeting 2015*.

Biber, D., Ellis, R., Ransaw, C.J., Saringer, C., **Ferrer. D.**, Hamilton, A., Abbott, L. (2015). Motivational Correlates of Physical Activity Among University Employees Dept. of Kinesiology & Health, Georgia State University, Atlanta, GA 30302. *SEACM Annual Meeting 2015*.

Ferrer, D. A., Brandenberger, K. J., Baumann, C. W., & Otis, J. S. Physical motivation influences race performance over a 24 hour ultra-marathon. *International Journal of Sport Studies* 5(10),1162-1169. 2015.

Baumann, C. W., Brandenberger, K. J., **Ferrer, D. A.**, & Otis, J. S. (2014) Physiological Parameters Associated with 24 Hour Run Performance. *Age* (y), 41(11.41), 43-28

PROFESSIONAL SOCIETIES AND ORGANIZATIONS

2012-Present	American College of Sports Medicine
2016-Present	National Strength and Conditioning Association

THE EFFECTS OF ULTRA-FILTERED MILK CONSUMPTION ON STRENGTH AND PERFORMANCE FOLLOWING RESISTANCE TRAINING IN FEMALE COLLEGIATE ATHLETES

By

DAVID A. FERRER

Under the Direction of Jeffrey S. Otis, Ph. D. and Brett J. Wong, Ph. D.

ABSTRACT

Resistance training is beneficial in the improvement of skeletal muscle functionality. Improvements in performance, increased resistance to injury, and great force production are associated with resistance training. Hypertrophy of skeletal muscle mass is important for improving fitness, decreasing body fat percentage, improvements in whole-body metabolism, and enhancements in quality of life. The ability to recovery properly following subsequent training sessions is critical for maximizing training adaptations. Nutrient supplementation has been previously studied. The supplementation of carbohydrates has been shown to replenish muscle glycogen stores. The consumption of carbohydrates following resistance training benefits muscle protein balance by attenuating muscle protein breakdown. Another commonly consumed supplement is amino acids/protein. Supplementation of protein has demonstrated improvements in body composition (i.e. increased fat free mass), increases in hypertrophy, and muscular strength. Two type of proteins used by individuals that resistance train are whey protein and casein protein. Whey protein is a fast digesting protein that leads to quick stimulation of protein synthesis. Casein protein is a slower digesting protein that also attenuates the breakdown of muscle protein. Milk is a natural product that contains carbohydrates, whey protein, and casein protein. Whole milk, low fat milk (i.e., 1-2%), and fat free milk have shown positive results in the ability to improve muscle protein synthesis, lean body mass, strength gains. Therefore, the purpose of the following dissertation is to compare the effects of higher protein, less sugar content chocolate milk to traditional low fat chocolate milk on adaptations to (1) strength and performance measures and (2) body composition following resistance training.

THE EFFECTS OF ULTRA-FILTERED MILK CONSUMPTION ON STRENGTH AND PERFORMANCE FOLLOWING RESISTANCE TRAINING IN FEMALE COLLEGIATE ATHLETES

By

DAVID A. FERRER

A Dissertation

Presented in Partial Fulfillment of Requirements for the

Degree of

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In

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In

Department of Kinesiology and Health

In

The College of Education & Human Development

Georgia State University

Atlanta, GA

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DEDICATION

Nothing that I have been able to accomplish would have been possible without the love and support of my wife Emily. Without her, this would have been a journey that could not have possible. Thanks for helping me keep my head up and pushing me.

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ABBREVIATIONS

3RM	One repetition max
4E-BP1	4E binding protein
p70S6K1	70KDa ribosomal S6 protein kinase
ÂA	Amino Acids
Akt	Protein Kinase B
BCAA	Branched Chain Amino Acids
CAS	Casein Protein
CC	Calf Circumference
СНО	Carbohydrate
СК	Creatine Kinase
СМ	Chocolate Milk
CSA	Cross-Sectional Area
DOMS	Delayed Onset Muscle Soreness
EAA	Essential Amino Acids
EMG	Electromyography
FFM	Fat-Free Milk
FoxO1	Forkhead Box Protein O1
GSK3β	Glycogen Synthase Kinase 3 ^β
IGF-1	Insulin-like Growth Factor-1
LDH	Lactate Dehydrogenase
LBM	Lean Body Mass
mTOR	Mammalian target of rapamycin
MAA	Mixed Amino Acids
MPB	Muscle Protein Breakdown
MPS	Muscle Protein Synthesis
MuRF1	Muscle RING-Finger Protein-1
MVC	Maximal Voluntary Contractions
NAA	Non-Essential Amino Acids
PGC-1γ	Peroxisome Proliferator-Activated Receptor γ Coactivator 1
PI3K '	Phosphatidylinositol 3-kinase
PIP3	Phosphatidylinositol-3, 4, 5, triphosphates
PLA	Placebo
PRO	Protein
RFD	Rate of Force Development
TC	Thigh Circumference
UFM	Ultra-filtered Chocolate Milk
VJ	Vertical Jump
WM	Whole Milk
WHP	Whey Protein
	-

Chapter One

Post Exercise Recovery Drinks

Introduction

It is well-known that resistance exercise can enhance the functionality of skeletal muscle in a myriad of ways such as increased force production, improved resistance to injury, and increased physical performance (Westcott, 2012) . Hypertrophy of skeletal muscle mass is important for improving fitness, decreasing body fat percentage, improvements in whole-body metabolism, and enhancements in quality of life (Gonzalez, Hoffman, Stout, Fukuda, & Willoughby, 2016). Resistance training also promotes neural adaptations such as improved motor unit recruitment, motor unit synchronization, and improved rate of force production (Maffiuletti et al., 2016). Neural adaptions in response to resistance training lead to improvements in muscular hypertrophy, strength, power, and local muscular endurance (Deschenes & Kraemer, 2002).

Increases in muscle mass due to protein accretion are referred to as hypertrophy, which is characterized by an increase in the number of myofilaments, myosin and actin, as well as an increase in cross-sectional area (CSA). Hypertrophy is strongly correlated to strength gains in skeletal muscle. With an increase in hypertrophy, lean body mass (LBM) also increases. Lean body mass is imperative in maintaining healthy body weight; and in turn, combats the potential for the development of metabolic diseases (Lumeng & Saltiel, 2011). Strength gains in skeletal muscle following resistance exercise can be measured by assessments of muscle functionality (i.e., improved muscular strength, and endurance). While resistance exercise is beneficial, proper recovery from subsequent bouts is imperative to maintain training gains.

Recovery is crucial in accumulating benefits of resistance exercise. One common method to improve recovery is the consumption of a post-exercise nutritional beverage (Beelen, Burke, Gibala, & Van Loon, 2010a; Ferguson-Stegall et al., 2011; Howatson & Van Someren, 2008; Lynch, 2013a; Pritchett, Pritchett, & Bishop, 2011; Rankin et al., 2004; Sousa, Teixeira, & Soares, 2014). Dietary protein is a commonly used macronutrient as a post-exercise nutritional beverage due to its ability to attenuate muscle damage while also increasing muscle protein synthesis (MPS) (Howatson & Van Someren, 2008; Koopman, Pennings, Zorenc, & Van Loon, 2007; D. Moore, Atherton, Rennie, Tarnopolsky, & Phillips, 2011) . The aim of this literature review is to detail functional and physiological adaptations of skeletal muscle to resistance exercise, explain the signaling mechanisms that regulate hypertrophy, and the effects of utilizing protein rich post-exercise nutritional beverages.

Skeletal Muscle Adaptations to Resistance Exercise

Neural Adaptions

Neural adaptations are primarily responsible for initial strength gains in humans for roughly the first six months of resistance training (Folland & Williams, 2007; Gabriel, Kamen, & Frost, 2006). Neural adaptations to resistance training include improved motor unit recruitment, firing frequency, motor unit synchronization, and agonist-antagonist interaction (Cormie, McGuigan, & Newton, 2011; Folland & Williams, 2007; Gabriel et al., 2006). Depending on the force needed, motor units are recruited based on the size principle. Size principle dictates that smaller motor neurons within type I fibers will be recruited first followed by type IIa and IIx fibers as force production increases (Henneman, Clamann, Gillies, & Skinner, 1974; Henneman, Somjen, & Carpenter, 1965). The ability to recruit more motor units as well as higher threshold motor units is crucial for greater force production. While humans are not able to completely (~95%) activate muscles voluntarily, resistance training can improve total activation (Gabriel et al., 2006). Resistance training lowers the recruitment threshold for high-threshold motor units, thus making it easier to produce greater forces voluntarily. Cormie et al. (2011) explain that increases in firing rates can heighten the magnitude of force per contraction as well as impact rate of force development (RFD), both influencing the development of muscular power. Aagaard, Simonsen, Andersen, Magnusson, and Dyhre-Poulsen (2002) were able to increase RFD in participants following 14wks of resistance training. Increases in electromyography (EMG) increased up to 143% in contractions and 106% in the early phase of contractions indicating enhanced neural drive. Similarly, simultaneous activation of multiple motor neurons known as synchronization has been implied to increase force production.

Motor unit synchronization was studied early by Milner-Brown and Stein (1975). Briefly, their group studied motor unit synchronization in hand muscles following strength training in bus drivers using surface EMG (SEMG). The use of SEMG was disputed (Yue, Fuglevand, Nordstrom, & Enoka, 1995), but J. Semmler and Nordstrom (1998) directly observed motor unit synchronization in strength-trained individuals, musicians, and controls. Controls, skilled musicians, and strength-trained individuals had varying, increasing synchronization respectively. This indicates that motor unit synchronization increases with increases in skilled movement or patterns (J. G. Semmler, Sale, Meyer, & Nordstrom, 2004). The more efficient the movement or pattern, the increased ability to have motor unit synchronization could increase RFD, thus increasing strength.

Greater motor control and proficiency in skill may also lead to greater force production in resistance-trained individuals (Dettmers et al., 1996; Folland & Williams, 2007; Gabriel et al.,

2006). Nozaki, Nakazawa, and Akai (2005) demonstrated a wide range of muscle activity variability in knee extension not only between subjects, but also within subjects. The improvement in motor skill is commonly seen in the ability to increase agonist activation (Folland & Williams, 2007; Gabriel et al., 2006). In regards to the capacity to produce maximal force, co-activation of antagonists is problematic. With the activation of antagonists, force is produced upon the joint in the opposite direction of the agonist preventing complete activation of the agonist. While important for the stability of the joint in ballistic movements, this limits force production. Carolan and Cafarelli (1992) found a decrease in antagonistic activation in the biceps femoris with increased activation of agonist, vastus lateralis, following 8wks of knee extensor training.

Skeletal Muscle Hypertrophy

Hypertrophy is characterized by the increase in the number of contractile proteins, thus increasing the cross-sectional area of skeletal muscle fibers, and is a primary determinant of strength gains in response to resistance exercise. Exercise type elicits differences in activation pathways and gene expression, which ultimately dictates the primary adaptation observed in skeletal muscle. Satellite cells act as the primary stem cell for skeletal muscles in humans. Due to the post-mitotic and multinucleated nature of skeletal muscle, satellite cell activation occurs as well to induce hypertrophy. Specific to skeletal muscle satellite cells, myogenic precursor cells are activated via mechanical stress and differentiate into myocytes that then fuse with myofibers (shown in figure 1); with this fusion, the recently differentiated myocytes will then add to protein synthesis by the addition of their nuclei leading to hypertrophy (Hawke & Garry, 2001; Kadi & Thornell, 2000). The mechanical signals of resistance exercise prompt hypertrophy signaling

pathways, which regulate transcriptional and translational activity resulting in muscle protein synthesis.

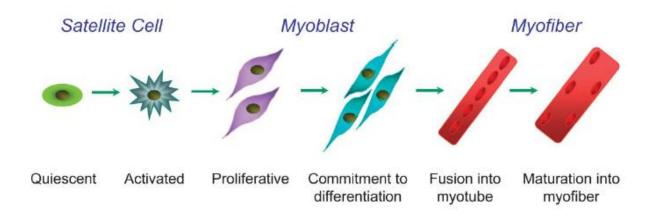


Figure 1: Schematic of satellite cell myogenesis created by Zammit, Partridge, and Yablonka-Reuveni (2006).

IGF-PI3K-AKT-mTOR Signaling Pathway

Growth hormone (GH) has a vital role in growth by regulating insulin like growth factor-1 secretion from skeletal muscle. The secretion of GH from the pituitary gland promotes lipolysis, amino acid transportation, glucose transport, and protein synthesis (Clemmons, 2004). Insulin like growth factor-1 (IGF-1) is a signaling molecule implicated in skeletal muscle hypertrophy using a variety of methods from localized infusion of IGF-1 in control muscles (Adams & McCue, 1998), infusion of IGF-1 to atrophied skeletal muscle (Chakravarthy, Davis, & Booth, 2000), to overexpression of IGF-1 (Musarò et al., 2001). Insulin like growth factor-1 is secreted from the liver after mechanical stress on skeletal muscle and binds to insulin receptor substrate within the myocyte, which is an activator of phosphatidylinositol 3-kinase (PI3K; Backer et al. (1992). PI3K then activates phosphatidylinositol-3, 4, 5, triphosphate (PIP3) causing the activation of protein kinase B (Akt). The primary isoform of Akt responsible for hypertrophy is Akt1 which acts as an upstream regulator of mammalian target of rapamycin (mTOR), which is responsible for regulating both 4E binding protein (4E-BP1) and 70KDa ribosomal S6 protein kinase (p70S6K1) (Glass, 2003, 2005). mTOR is associated with the increases in protein synthesis and cell size (Ali & Sabatini, 2005; Bodine et al., 2001; Ohanna et al., 2005). The phosphorylation of p70S6K1 has been implicated to induce skeletal muscle hypertrophy (Coffey & Hawley, 2007; Egan & Zierath, 2013; Glass, 2003, 2005; Lai et al., 2004; Nader, 2005; Nader & Esser, 2001; Rommel et al., 2001). This process is illustrated in figure 2. 4E-BP1 is responsible for the inhibition of protein synthesis, but it is phosphorylated and suppressed by mTOR.

Akt also plays a role in the inhibition of catabolic processes, protein degradation, by inhibiting the activity of glycogen synthase kinase 3β (Glass, 2003, 2005; Nader, 2005) and forkhead box protein O1 (Latres et al., 2005; Stitt et al., 2004). The inactivation of FoxO1 is vital due to its transcriptional ability to regulate muscle RING-finger protein-1 (MuRF1) and Atrogin, two genes implicated in muscle atrophy (Stitt et al., 2004). The regulatory role of FoxO1 on MuRF1 has been previously demonstrated in atrophy models (Sandri et al., 2004; Stitt et al., 2004). The role atrogin plays in protein breakdown is through the degradation of MyoD (Tintignac et al., 2005).

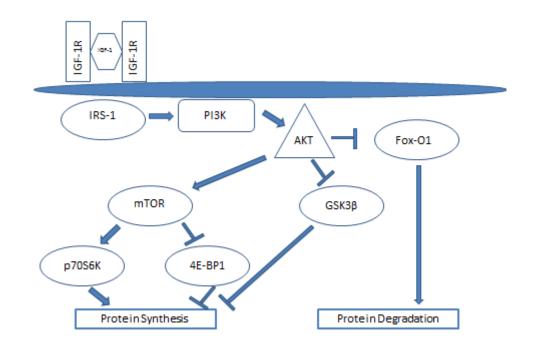


Figure 2. Schematic representation of IGF-PI3K-AKT-mTOR pathway to promote MPS and inhibit protein degradation adapted from Hitachi and Tsuchida 2014. Abbreviations: IGF-1R, insulin-like growth factor-1 receptor; IGF-1, insulin like growth factor-1; IRS-1, insulin receptor substrate; PI3K, phosphatidylinositol 3-kinase; Akt, protein kinase B; mTOR, mammalian target of rapamycin; p70S6K, 70KDa ribosomal S6 protein kinase; 4E-BP1, 4E binding protein; GSK3β, glycogen synthase kinase 3β; Fox O1, forkhead box protein O1.

Carbohydrate Supplementation

Carbohydrates are classified as monosaccharides, disaccharides, and polysaccharides. Monosaccharides, such as glucose, fructose, and galactose are considered simple sugars which contain 6-carbon atoms. Glucose is stored in the body in the form of glycogen within the liver or skeletal muscle. Glycogen is broken down via glycogenosis to yield glucose. Glucose is then able to enter the glycolytic pathway in order to yield energy in the form of adenosine triphosphate (ATP). Resistance exercise has been demonstrated to deplete muscle glycogen stores, carbohydrate supplementation is thought to benefit recovery and prevent decreases in performance (Haaf et al., 2010).

The idea behind this practice is to reload endogenous fuel substrates that have been used during activity. Muscle glycogen is the predominant carbohydrate (CHO) source during moderate to high intensity exercise (Egan & Zierath, 2013). The rate at which skeletal muscle utilizes CHO is dependent upon intensity and duration of activity. With higher intensity resistance training (i.e., increased loads or volume) carbohydrates from muscle or liver glycogen are utilized. Along with the phosphocreatine system, the need for carbohydrate replenishment is important for subsequent bouts of activity considering the use of carbohydrate as a fuel source during resistance exercise (Egan & Zierath, 2013; Haff, LEHMKUHL, MCCOY, & STONE, 2003; Macdougall et al., 1999; Schoenfeld, 2010).

Following resistance training, both muscle protein synthesis and muscle protein breakdown are elevated and remains in a negative balance in a fasted state (Kumar, Atherton, Smith, & Rennie, 2009). Due to the disruption in muscle protein balance, carbohydrate supplementation has been studied to improve the muscle protein synthesis-muscle protein balance (MPS-MPB) balance as a means to achieve an anabolic response. The ingestion of carbohydrates leads to increases in blood glucose, thus increasing rates of insulin secretion from the pancreas. An increase in insulin, in turn, should then enhance protein synthesis via the IGF-1/PI-3K/Akt, mTOR pathway. The literature is mixed in regards to the effects of CHO supplementation on MPS following resistance exercise.

Protein supplementation is widely accepted as a nutritional strategy to improve MPS, but to compare the effects of CHO on MPS, researchers have compared the use of CHO+PRO versus

PRO alone demonstrating the lack of a synergistic effect of insulin on MPS when comparing CHO+PRO and PRO alone (Koopman, Beelen, et al., 2007; Staples et al., 2011). Some studies have proposed the mechanism of which CHO benefits muscle protein balance are derived from its ability to attenuate MPB rather than improve MPS suggesting CHO promotes a positive muscle protein balance following resistance training (Beelen et al., 2010a; Børsheim et al., 2004; B. Roy, Tarnopolsky, MacDougall, Fowles, & Yarasheski, 1997; B. D. Roy, Fowles, Hill, & Tarnopolsky, 2000). For example, Roy and colleagues (1997) provided participants with 1g/kg of a CHO supplement or a placebo immediately and 1h following a bout of unilateral knee extensor exercise. Fractional muscle protein synthesis rate was 36% greater in the CHO group compared to the placebo, but this was not statistically significant. Roy et al (1997) suggested CHO supplementation was able to attenuate muscle protein breakdown, resulting in a less negative whole body protein balance. Increases in plasma insulin concentration have been attributed to the attenuation of muscle protein breakdown (Biolo, Tipton, Klein, & Wolfe, 1997; Greenhaff et al., 2008). When CHO is added to a protein mix compared to protein alone, CHO does not enhance MPS (Creer et al., 2005; B. Roy et al., 1997; Staples et al., 2011). In contrast, CHO supplementation does increase muscle glycogen stores.

Low levels of muscle glycogen influence net muscle protein balance. Transcriptional rates of metabolic and myogenic genes such as myogenin are suppressed (Churchley et al., 2007) and can promote MPB (Lemon & Mullin, 1980) in muscle that is glycogen-depleted. For individuals who engage in resistance training, replenishment of muscle glycogen is vital. With CHO supplementation, acceleration of muscle glycogen resynthesization occurs and can return to resting levels 24 hours following training. Athletes typically have multiple training sessions daily. Training sessions are not limited to resistance exercise such as strength training, but sport

related activities (i.e. practice) also add to the training regimen. Such training sessions require the use of muscle glycogen and can be separated by a few hours (e.g., weight training in the morning followed by practice in the afternoon/evening). CHO supplementation assists with short-term muscle glycogen replenishment thus benefiting subsequent training sessions. Consumption of CHO post-exercise seems to be the optimal recovery period to enhance recovery and performance (C. Kerksick et al., 2008; Pritchett et al., 2011). The consumption of CHO, regardless of liquid or solid form, increases the rate of muscle glycogen resynthesis. Comparatively, consumption of CHO within 30 minutes following resistance exercise is more beneficial in replenishing glycogen stores than delaying consumption past 1 hour (Ivy, Ding, Hwang, Cialdella-Kam, & Morrison, 2008).

Protein/Amino Acid Supplementation

Athletes who are involved in intense training programs require more daily protein, 1.6g/kg body weight, compared to that of the United States Recommended Daily Allowance of 0.8g/kg of body weight , (Wu, 2016). This recommendation is similar between endurance and resistance training individuals, 1.3g/kg and 1.6g/kg respectively. Dependent on the amino acid content of a protein source, proteins may be considered complete or incomplete proteins. Proteins that contain all amino acids are considered complete proteins and those that do not are incomplete proteins (C. M. Kerksick et al., 2006). The concentration of branched chain amino acids (BCAAs) within a protein source has an effect on protein synthesis, with high concentrations promoting greater rates of protein synthesis (Børsheim, Tipton, Wolf, & Wolfe, 2002). Protein and/or amino acid supplementation has been studied in a variety of exercise modes. Modes ranging from sub-maximal running to eccentric resistance exercise have proven to benefit from protein supplementation (Burd, Tang, Moore, & Phillips, 2009; Pasiakos,

Lieberman, & McLellan, 2014). Feeding in general as well as resistance training increases muscle protein synthesis, but following a fasting period, muscle protein breakdown occurs in similar rates causing a net neutral balance in regards to muscle protein synthesis. The combination of resistance training along with feeding (i.e., protein supplementation) following training increases muscle protein synthesis above and beyond normal rates while attenuating the rates of muscle protein breakdown, causing greater fed gains (i.e. muscle protein synthesis) than fasted losses (i.e. muscle protein breakdown) resulting in a positive net balance of muscle protein balance. This relationship is illustrated in figure 3. While resistance exercise does enhance MPS, muscle protein net balance is further improved to a positive state more so with PRO/AA supplementation compared to a fasted state (Burd et al., 2009; Reidy & Rasmussen, 2016). Protein supplementation is also associated with increases in fat free mass, hypertrophy, and muscular strength (Cermak, de Groot, Saris, & van Loon, 2012).

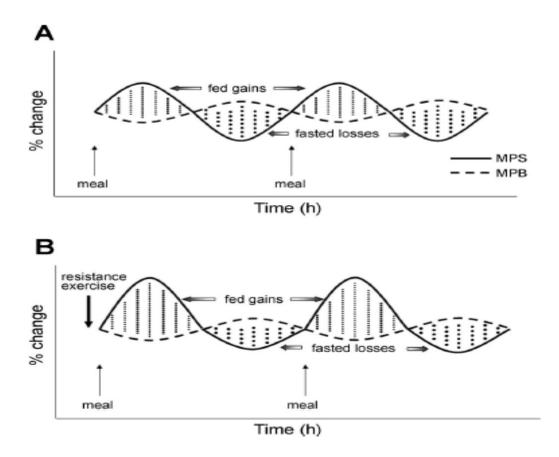


Figure 3. *A*: changes in muscle protein synthesis and muscle protein breakdown following feeding. *B* changes in muscle protein synthesis and muscle protein breakdown following the combination of resistance training and feeding (Burd et al., 2009).

Muscle Protein Anabolism

While there is clear evidence of increased MPS with protein/amino acid supplementation, improving the turnover rate to reach a net positive balance has also been strongly supported (Beelen et al., 2010a; Børsheim et al., 2004; Elliot, Cree, Sanford, Wolfe, & Tipton, 2006b; Koopman, Beelen, et al., 2007; Rasmussen, Tipton, Miller, Wolf, & Wolfe, 2000; Reidy et al., 2014; Reitelseder et al., 2014; Tang et al., 2007; Tipton et al., 2007; Tipton et al., 2004; Tipton, Ferrando, Phillips, Doyle, & Wolfe, 1999; Wilkinson et al., 2007; Witard et al., 2014). Tipton and colleagues (1999) compared the supplementation of either 40g of mixed amino acids, 40g of essential amino acids, or a placebo in untrained young, adults. Rate of MPB was not significant among groups, but the mixed and essential amino acid groups significantly increased net protein balance in comparison to the placebo. No group differences were observed between mixed and essential amino acids. Since net protein balance was not significantly different between the mixed amino acid group and the essential amino acid group, their study demonstrated nonessential amino acids were unnecessary for improving net protein balance. Børsheim et al. (2002) investigated the effect of essential amino acid (EAA) supplementation on the stimulation of net muscle protein balance. Their group concluded that since non-essential amino acid (NEAA) levels were maintained following a bout of resistance training, NEAA supplementation was unnecessary to increasing net muscle protein.

Similar results were found utilizing milk as Elliot et al (2006) compared fat-free milk, whole milk, and isocaloric fat-free milk in untrained, young adults. Each group consumed their milk 1h following 10 sets of 8 repetitions of knee extensions. Ingestion of all three milk groups improved glucose levels as well as blood amino acid concentration and net protein balance. Increases in net protein balance are not limited to untrained populations. Witard et al (2014) studied 48 resistance trained, young men. Participants performed a bout of leg presses and leg extension then ingested varying amounts (0, 10, 20, or 40g) of whey protein immediately following exercise. Those whom consumed 20g of whey protein increased muscle protein synthesis 49%. Similar results were observed in both recreational athletes as well as resistance trained individuals (Borsheim et al 2002; Borsheim et al 2004; Rasmussen et al 2000; Reidy et al 2014; Reitelseder et al 2014; Witard et al 2014).

Timing of Supplementation

An important consideration for promoting the benefits of protein supplementation is the timing during which the supplementation is consumed. Appropriate timing of protein ingestion is thought to aid in the enhancement of adaptations in body composition, strength, hypertrophy, MPS, and recovery (Phillips 2013; Stark et al., 2014; Wilborn et al., 2013). Generally, the consumption of a protein/AA supplement pre- and/or post-resistance exercise has shown to improve training adaptations (Phillips 2013; Stark et al., 2014; Wilborn et al., 2013).

Amino acid supplementation before resistance exercise has been tested with equivocal results. Initially, Tipton et al. (2001) sought to determine if ingesting amino acids immediately before resistance exercise would be more effective in increasing MPS compared to consumption post-resistance exercise. Due to increased muscle protein breakdown during resistance exercise, Tipton and colleagues hypothesized that inducing hyperaminoacidemia before training would attenuate the rate of muscle protein breakdown during training. Participants performed 10 sets of 8 repetitions of leg press at 80% of their 1RM. Participants received an oral AA+CHO solution either immediately before or immediately after the exercise bout. Consumption of amino acids prior to the bout of exercise was able to increase AA delivery to the leg significantly faster than post, resulting in higher levels of MPS. The results of this study were contrary to the outcome of the previous work of Rasmussen et al (2000). The exercise protocol and composition of the oral supplementation Tipton et al utilized was identical to that of Rasmussen et al. For the Rasmussen et al (2000) study, MPS was much greater with post-exercise consumption than that of Tipton et al.

Strength and Body Composition Adaptations

Adequate consumption of protein is necessary to maximize adaptations from resistance training (Schoenfeld, Aragon, and Krieger 2013). Theoretically, the ability of protein consumption following training to increase muscle protein anabolism would promote greater gains in lean mass and skeletal muscle hypertrophy leading to gains in skeletal muscle performance. The supplementation of protein concurrent with resistance training has shown to increase strength gains and skeletal muscle hypertrophy (Anderson et al., 2005; Bird, Tarpenning, and Marino 2006; Coburn et al., 2006; Cribb et al., 2007; Willoughby, Stout, and Wilborn 2006). Bird, Tarpenning, and Marino (2006) studied untrained men following a 2d/wk, 12wk total body resistance training protocol. Participants consumed one of four drinks: carbohydrate, essential amino acids, carbohydrate + essential amino acids or a placebo following each training session. Following training, those consuming the CHO + EAA supplement significantly increased their 1- repetition max (1RM) on the leg-press compared to their placebo counterpart. Results were similar in regards to hypertrophy with the CHO + EAA group increased functional cross-sectional area in both type-1 and type-II fibers compared to placebo. While hypertrophy was significantly greater for all treatment groups, there were no differences between groups for decreases in fat mass. All treatment groups were able to increase fat-free mass, with the CHO + EAA group having the greater gains compared to the placebo. Similarly, Anderson et al (2005) compared protein to carbohydrate supplementation immediately before and after resistance training. Their program consisted of 3d/wk, 14wk lower body resistance training. Participants executed two types of vertical jump: a squat jump and a countermovement jump. Along with jump performance, isokinetic knee extensor peak torque was also measured. The protein supplement group was able to increase both type-I and type-II muscle fiber CSA by

 $18\% \pm 5\%$ and $26\% \pm 5\%$ respectively whereas the carbohydrate group did not experience any significant changes. Similarly, squat jump performance was significantly increased in the protein group, but not the carbohydrate group.

Along with strength gains, supplementation of protein has shown to improve body composition by increasing lean mass (Burke et al., 2001; Cermak et al., 2012; Candow et al., 2006; Volek et al., 2013). Candow et al (2006) studied healthy adults training 3d/wk for 6wks following a full-body training program. Participants consumed a whey supplement, soy supplement, or maltodextrine placebo before and after training. Participants tested a 1-RM bench press and 1-RM hack squat as well as dual energy X-ray absorptiometry (DEXA) prior and following 6wks of training. Regardless of protein supplement, strength improved for both bench press (whey: +8.2 kg or 14%; soy: +7.6 kg or 13.4%; placebo: +4 kg or 7.1%) and hack squat (whey: +26.7 kg or 38.6%; soy: +23.7 kg or 34%; placebo:+14.1 kg or 19.7%) significantly greater compared to the placebo group. Lean tissue mass also increased significantly greater with protein supplementation compared to placebo (whey: +205 kg or 4.7%; soy: +1.7 kg or 3.1%; placebo: +0.3 kg or 0.5%). Volek et al (2013) were able to demonstrate increases in lean body mass with the supplementation of whey protein. Participants engaged in a non-linear periodized program for 9 months (~96 training sessions). During this time, participants consumed similar supplements as Candow et al; supplements included whey protein, soy, and a maltodextrine placebo. Lean body mass was measured at 3, 6, and 9 months. While all groups increased body mass, whey protein had significantly greater increases in lean body mass compared to both the soy and placebo groups at all-time points.

Protein Supplementation

Amino Acids

Twenty amino acids are involved to synthesize proteins. Of those amino acids, nine are classified as essential amino acids. These particular amino acids are essential due to the inability to synthesize these amino acids within the body and therefore must be obtained via dietary sources. Essential amino acids include histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine. The remaining eleven are non-essential amino acids because of their ability to be synthesized naturally within the body. Non-essential amino acids include aspartic acid, glutamic acid, proline, glycine, serine, alanine, cysteine, arginine, asparagine, glutamine, and tyrosine. Another form of amino acids studied is BCAAs. BCAAs consist of isoleucine, leucine, and valine. Amino acids have been investigated to improve MPS, decrease MPB, improve net muscle protein balance, decreases markers of muscle damage, increase performance, and recovery from intense exercise.

Amino acids have been associated with increases in MPS (Biolo et al., 1997; Blomstrand, Eliasson, Karlsson, & Köhnke, 2006; Børsheim et al., 2002; Crozier, Kimball, Emmert, Anthony, & Jefferson, 2005; Rasmussen et al., 2000; Sharp & Pearson, 2010; Tipton et al., 1999; Tipton et al., 2001; Volpi, Kobayashi, Sheffield-Moore, Mittendorfer, & Wolfe, 2003). Biolo and colleagues (1995) studied the rate of protein synthesis and degradation from the vastus lateralis within untrained adults. Their studied demonstrated increased levels of leucine, lysine, and alanine with an improved muscle protein balance utilizing both arteriovenous blood samples and biopsies from the vastus lateralis. While muscle protein balance was improved, it was not increased to a positive state. Similarly, Tipton et al (2001) achieved small, but non-significant

increases in muscle protein balance. This is largely in contrast with later studies that found increases in net muscle protein balance due to increased mTOR activation. p70S6K plays a primary role in skeletal muscle hypertrophy. Activation of mTOR and phosphorylation of p70S6K has been associated with EAA or BCAA supplementation leading to net muscle protein balance (Blomstrand et al., 2006; Crozier et al., 2005; Karlsson et al., 2004; Rennie, Bohé, Smith, Wackerhage, & Greenhaff, 2006). Karlosson et al (2004) investigated how resistance exercise alone compared to a combination of resistance exercise and oral intake of BCAA on phosphorylation of p70S6K. BCAA supplementation increased plasma concentrations compared to PLA during both exercise and up to 2h post-exercise. The BCAA group also increased p70S6K phosphorylation 2.5-fold during recovery. In a similar fashion, leucine alone has been able to increase MPS and phosphorylation of p70S6K (Crozier et al 2005; Rennie et al 2006). Increases in p70S6K phosphorylation would presume beneficial in hypertrophic adaptations.

Improvement in DOMS due to supplementation of AA has been observed (Howatson et al., 2012; Jackman, Witard, Jeukendrup, & Tipton, 2010; Matsumoto et al., 2009; Nosaka, Sacco, & Mawatari, 2006; Shimomura et al., 2006). Shinomura et al (2006) investigated the ability of BCAAs to enhance recovery and alleviate DOMS. Their participants were untrained, young adult males and females that followed a squat protocol. Compared to the placebo group, BCAA experienced peak soreness on the second day after exercise, while the placebo group had peak soreness three days after. Up to five days later in females, the BCAA group had significantly lower DOMS compared to that of the placebo. Males tended to have peak DOMS only two days post-exercise and was significantly lower in the BCAA group. Shinomura hypothesized the amount of BCAA consumption as a potential reason for differences in DOMS.

Males consumed less BCAA per kg of body weight compared to females (77±3mg/kg and 92±2 mg/kg respectively).

Markers of muscle damage have also been reduced with supplementation (Howatson et al., 2012; Matsumoto et al., 2009; Sharp & Pearson, 2010). Sharp and Pearson (2010) used three weeks of high intensity resistance training (HIRT) in untrained young males to induce exercise-induced muscle damage. BCAA supplementation has been associated with reduction in creatine kinase (CK). Similarly, Howatson et al (2012) studied twelve young, trained adult males after a sport specific bout of damaging exercise. Muscle damage markers, CK, maximal voluntary contraction (MVC), DOMS, vertical jump (VJ), thigh circumference (TC), and calf circumference (CC) were measured. VJ, TC, and CC were not different between groups, but significant reductions in CK and recovery of MVC was greater in the BCAA group compared to a placebo suggesting BCAA supplementation reduced exercised-induced muscle damage while aiding in recovery.

Whey Protein & Casein Protein

Whey protein is most commonly found in dairy milk and is considered a complete protein due to its composition of every EAA with a high concentration of BCAA. Whey protein is a rich source of leucine, which is a augmenter of p70S6K phosphorylation (Cribb & Hayes, 2006; Koopman et al., 2005). Furthermore, Koopman et al (2005) demonstrated the addition of leucine to a whey protein WHP supplement increased plasma insulin, muscle protein synthesis, and whole body protein balance when compared to a WHP+CHO alone. WHP has become a popular supplement for those engaging in resistance training and is typically consumed as either WHP concentrate (per 100g, 80% is pure whey protein) or WHP isolate (per 100g, 90% is pure whey

protein) (Cribb et al., 2006). WHP is considered a fast digested protein due to its ability to exit the stomach rapidly while increasing AA levels and whole body anabolism (Boirie et al., 1997; Devries & Phillips, 2015). Alternately, another commonly consumed post-exercise supplement is casein protein (CAS). Casein is a dairy protein that is digested slowly and takes longer to leave the stomach and small intestine (Devries and Phillips, 2014). Supplementation with WHP concurrent with resistance training has shown improvements in MPS, hypertrophy, increases in LBM, decreases in fat mass, and strength improvements (Hulmi, Lockwood, & Stout, 2010; Pasiakos et al., 2014; Pasiakos, McLellan, & Lieberman, 2015). Similarly, CAS supplementation has been associated with the promotion of muscle building (Hartman et al., 2007) and improvement in overall muscle protein balance (Boirie et al., 1997; Dangin et al., 2001).

As with AA supplementation, improvements in MPS has been well documented with WHP supplementation (Boirie et al., 1997; Burd et al., 2012; Hulmi et al., 2010; Koopman, Beelen, et al., 2007; D. Moore et al., 2011; Tipton & Phillips, 2013). Boirie and colleagues (1997) studied the effects on postprandial protein synthesis with WHP or CAS supplementation in sixteen healthy, young adults. Subjects received either whey protein or casein. Results demonstrated different rates and uses between WHP and CAS. WHP supplementation had a rapid and large increase in dietary AA indicating greater MPS, but no changes in MPB. CAS had a different metabolic response. Rates of dietary AA appearance were slower with minor increases in MPS, but noticeably inhibited MPB. The benefits from this study show the different absorption rates of different PRO sources. Similarly, Burd et al (2012) compared pure WHP isolate to micellar CAS supplementation, but compared MPS at rest and up to 4h after unilateral leg resistance exercise. The levels of leucine and blood AA were greatest after 1h of consumption with the rested leg experiencing 65% higher rates of MPS with WHP compared to CAS. Similarly, rates of MPS were greater in WHP versus CAS. While not statistically different than WHP, it is worth noting CAS had significantly higher levels of MPS thru 3h post exercise compared to immediately following exercise suggesting the ability of CAS to promote positive muscle protein balance for a longer duration which could lead to increases in hypertrophy. Moore et al (2011) studied seven healthy, young males performing unilateral resistance exercise. Subjects immediately consumed 25g of WHP and similarly compared MPS rates at rest and after resistance exercise as Burd and colleagues with one difference in methodology being MPS rates were taken at 1, 3, and 5h post exercise. With supplementation, the rested leg experienced increased levels of MPS only 1h post ingestion while p70SPK and eukaryotic elongation factor 2 (eEF2) phosphorylation were heightened 1, 3, and 5h after ingestion. It is known that resistance exercise increases MPS with MPB ultimately rising above the rates of MPS. It has been demonstrated that increasing and sustaining the levels of MPS with WHP supplementation is beneficial to increasing hypertrophy after resistance exercise (Pasiakos et al 2015).

With the ability of both proteins to further stimulate whole body MPS after resistance exercise, it is logical to assume this may lead to increases in skeletal muscle hypertrophy and lean mass gains (Hulmi et al., 2009). Farup et al. (2014) compared the effects of eccentric or concentric resistance training in combination with whey protein hydrolysate + CHO or CHO supplementation alone on muscle and tendon hypertrophy. Participants were young (age 23.9±0.8 years) recreationally active men. Participants performed maximal knee extensor training one leg performing eccentric contractions and the other concentric contractions. Regardless of contractile mode, participants consuming whey protein hydrolysate + CHO increased both quadriceps and patellar tendon hypertrophy. Hulmi et al (2009) were able to demonstrate improved hypertrophy in the quadriceps following 21 weeks of bi-weekly resistance

training while consuming WHP immediately before and after resistance training when compared to a placebo. Their study observed decreased rates in *myostatin* (inhibitor of muscle growth) and increases in *myogenin* mRNA (induces myogenesis) which indicate protein supplementation's role in increasing cell growth. Along with increases in hypertrophy, WHP may improve body composition by increasing lean body mass.

Volek et al. (2013) demonstrated the supplementation of WHP increased LBM. Nonresistance-trained males and females were recruited to participate in a whole-body, periodized resistance-training program over nine months. Participants were randomized into a CHO, WHP, or soy protein group. Total protein intake, including supplementation was 1.1 g/kg body mass for the CHO group and 1.4g/kg body mass for the WHP and soy groups. Both protein groups received roughly 22g/day of their respective supplement. Supplements were consumed with breakfast on non-training days and immediately after exercise on training days. Those receiving WHP supplementation experienced significant changes in body composition (i.e., increases in lean body mass) at 3, 6, and 9 months compared to both the CHO and soy groups. The elevated levels in both resting and exercise induced plasma leucine content with WHP supplementation could explain the increases in LBM.

Separately, WHP is able to increase MPS to greater levels than CAS (Tang, Moore, Kujbida, Tarnopolsky, & Phillips, 2009), but the combination of the two have yielded promising results in regards to LBM. C. M. Kerksick et al. (2006) compared the ingestion of a whey (40g/d) + casein (8g/d) supplement to a whey (40g/d) + BCAA (3g/d) + L-glutamine (5g/d) supplement as well as a CHO (48g/d) placebo on performance measures and body composition. Participants were resistance trained adult males that participated in a 4d/wk, 10wk split-body program. Participants ingested their supplement within 2h of completing each workout and in

the morning on non-training days. Those receiving the mix of WHP and CAS experienced the greatest increase in lean mass and FFM compared to both placebo and WHP + BCAA + $_{L}$ -glutamine groups. One commonly available food source which contains both WHP and CAS is milk.

Milk & Chocolate Milk

Due to WHP's fast digestion leading to quick stimulation of protein synthesis as well as the slow digesting capabilities of CAS to suppress muscle protein breakdown, a supplement containing both would presumably be an ideal product. Similarly, consumption of CHO in close proximity to resistance exercise has proven beneficial in recovery between bouts or training. Milk is a natural food that contains WHP, CAS, and carbohydrates. While a natural food, dairy cattle receive Food and Drug Administration regulated bovine growth hormone (rbGH). This particular growth hormone is not biologically active in humans (Juskevich and Guyer 1990). Supplementation of bovine colostrum has shown to increase serum IGF-1 concentration in humans (Mero et al., 1997), but the amount found in cow's milk is minimal (~1mg/l; Haug, Hostmark, and Harsad 2007). Cade et al. (1991) performed one of the earliest studies to suggest milk as a post-exercise supplement. Their group examined if muscle damage occurred following high-intensity training as well as to find if a milk protein supplement could affect muscle damage. Creatine kinase (CK) and lactate dehydrogenase (LDH) were used as markers of muscle damage in Division I National Collegiate Athletic Association (NCAA) swimmers over a six-month training period. Swimmers were placed into four different groups that received a glucose-electrolyte solution, milk protein, glucose-electrolyte + sucrose, glucose-electrolyte + milk. During six-weeks of training of progressive increases in intensity, both milk groups experienced decreased levels of CK in both males and females. Following the six-weeks, only

eight participants continued to the final phase of the study, which included performing identical workouts one day a week for four weeks. Each week the beverage consumption changed. Milk protein supplementation post-exercise returned CK and LDH levels to control values more rapidly than the sucrose solution. This study led to future studies of milk as a post-exercise supplement to improve recovery (Cockburn, Bell, & Stevenson, 2013; Cockburn, Hayes, French, Stevenson, & St Clair Gibson, 2008; Gilson et al., 2010; Wojcik, Walber-Rankin, Smith, & Gwazdauskas, 2001). Further research has demonstrated the effects of milk consumption on muscle protein synthesis, lean body mass increases, and strength gains.

Elliot and colleagues (2006) studied the effects of varying types of milk on net muscle protein synthesis (MPS). Participants received 237g of fat free milk (FM), 237g of whole milk (WM), and 393g of fat-free milk isocaloric with WM (IM). Each group consumed their respective beverages following a one-hour bout of leg resistance exercise. Increased levels of amino acid uptake, particularly threonine and phenylalanine, were greater following milk consumption. The underlying contributions to these increases were not examined although increased metabolism of proteins was demonstrated. Wilkinson et al (2007) were able to demonstrate increased rates of MPS with consumption of fat-free milk following resistance exercise in young (21.6±0.3 years), trained males. Beverages of nonfat milk or isonitrogenous, isoenergetic, and macronutrient-matched soy protein were consumed following an exhaustive leg workout. Fractional synthetic rate of muscle proteins was elevated at greater levels 3h into recovery with the consumption of nonfat milk compared to soy protein consumption.

Rankin et al. (2004) was the first to compare the effects of consumption of milk and CHO drinks immediately following resistance-exercise on body composition during a ten-week training program for untrained young men (18-25 years). Beverage consumption occurred within

five minutes following training. Fat free mass was not significantly different between groups, but trended towards more increases with the consumption of milk compared to CHO. One possible explanation for this lack of differences could be the total energy consumed and similarities in protein consumption between the groups. Protein consumption was 1.2g/kg of body weight for the CHO group and 1.3g/kg of body weight for the milk group. Conversely, Hartman et al (2007) were able to promote greater levels of hypertrophy in novice weightlifters with the consumption of fat-free milk compared to an isoenergetic soy or carbohydrate supplement. Subjects were young males who trained 5d/week for 12 weeks utilizing a split-body resistance-training program. Beverages were consumed immediately before and 1h after training. The milk beverage contained ~17.5g protein, ~25.7g CHO, and ~0.4g fat. Subjects consuming milk experienced a 5.5% decrease in fat mass and a 6.2% increase in fat and bonefree mass, both of which were significantly different from the CHO and soy group. Milk consumption concurrent with resistance training also increased both type I and type II fiber CSA compared to CHO and soy groups. As opposed to Rankin et al (2004), total energy intake and protein intake were similar across all groups while changes in LBM were different between the groups.

Josse, Tang, Tarnopolsky, and Phillips (2010) performed a study on young women comparing the effects of supplementing fat-free milk to an isoenergetic carbohydrate beverage on lean mass gains and strength gains following a 5d/week, 12wk program. Participants within the CHO group gained weight, while both groups increased lean mass. However, those in the milk group gained a greater amount of lean mass and experienced a decrease in fat mass compared to the CHO group (-1.6 ± 0.4 kg vs -0.3 ± 0.4 kg,). While the milk group did not experience strength gains in all exercises, they did have subtle increases in upper body exercises

(i.e. bench press and chest fly) compared to the CHO group. With the potential of milk to improve training adaptations, a more appealing flavor to the palate that has demonstrated similar benefits is chocolate milk.

Chocolate milk contains cocoa which has potential benefits to exercise based on the ability to have an anti-inflammatory effect as well as being a rich source for antioxidants. Cocoa contains flavonoids that have an anti-inflammatory response to the body (Engler & Engler, 2006; Ramiro-Puig & Castell, 2009; Selmi, Mao, Keen, Schmitz, & Gershwin, 2006). Cocoa contains falvanols that modulate the synthesis of eicosanoids, mediators of inflammatory responses, while also stimulating tumor growth factor (TGF)-b production, an anti-inflammatory cytokine (Selmi et al., 2006). The antioxidants contained in cocoa would provide to be beneficial in the reduction of free radicals (Ramiro-Puig and Castel 2009). Goldfarb, Bloomer, and Mckenzie (2005) studied the effects of an antioxidant treatment after eccentric exercise on nonresistance trained females. Their supplementation of vitamin C, E, and selenium attenuated the increase of protein carbonyls and malondialdehyde, markers of oxidative stress. Similarly, Arent et al. (2009) studied the consumption of a post-workout nutraceutical drink on division I college football players. The drink contained carbohydrate, protein, fat, and anti-oxidants and was consumed following training sessions. Over seven weeks of training, those supplementing with the nutraceutical drink had greater increases in peak power and decreases in body fat percentage and fat mass. The treatment group also experienced greater increases in recovery and decreases in inflammation. Due to the make-up of chocolate milk (i.e., proteins, carbohydrates, fats, cocoa), chocolate milk has the potential to positively affect resistance training adaptations.

Chocolate milk (CM) has demonstrated benefits to aerobic training, body composition, and reduced rate of perceived exertion (RPE). Ferguson-Stegall et al (2009) compared CM,

isocaloric CHO, and a PLA supplement following 4.5 weeks of submaximal aerobic training. Participants within the CM groups experienced improvements in lean and fat mass differential for the trunk as well as whole body measures. Maximal oxygen consumption (VO₂ max) was assessed before and after training. Those within the CM group experienced a greater increase in VO₂ max compared to the CHO and PLA, pointing to the benefit of CM to improve aerobic power. Similarly, CM has demonstrated the ability to increase time to exhaustion (Lunn et al., 2012; Thomas, Morris, and Stevenson 2009). Thomas et al (2009) found CM was able to increase time to exhaustion following a glycogen-depleting bout of exercise. Participants completed a glycogen-depleting trial, followed by a 4hr recovery period, finishing with the completion of a cycle to exhaustion at 70% of VO₂ max. Comparatively, those consuming CM cycled 51% and 43% longer than their CHO and PLA counterparts. Improvements in aerobic performance have been documented, but there is a lack of literature on the benefits of CM following resistance training.

Wallace and Abel assessed the effects of CM on RPE, muscular peak strength, and fatigue following a bout of resistance training on the lower body. For their randomized crossover study, participants consumed skim chocolate milk and a non-caloric placebo. When consuming CM, participants reported significantly lower rates of perceived exertion as well as greater increases in knee flexion peak torque. These improvements would be beneficial for those who may perform multiple or subsequent resistance training bouts. Similar improvements in recovery were demonstrated in college soccer players. Gilson et al (2010) assessed RPE, serum creatine kinase, myoglobin, muscle soreness, fatigue ratings, and isometric quadriceps force following a period increased training duration. Training consisted of soccer-specific training along with strength and sprint training. Athletes consumed either CM or a CHO supplement

following training. Of their measures, only serum creatine kinase levels were significantly lower for the CM group.

Hypothesis

Whole milk, low fat milk (i.e., 1-2%), and fat free milk have shown benefits by increasing MPS, LBM, and strength gains. Calorically, white milk and chocolate milk differ in macronutrient content. For instance, whole white milk contains roughly 150kcal, 8g protein, 8g fat, and 12g carbohydrate per 236mL. Chocolate milk contains 150kcal, 9g protein, 3g fat, and 24g of carbohydrates per 236mL. Chocolate milk contains added sucrose and cocoa, but the amount varies between manufacturers. The difference in carbohydrate content would presume to be more beneficial in regards to muscle glycogen replenishment as well as the attenuation of muscle protein breakdown (J. L. Ivy & Ferguson-Stegall, 2014).

With milk displaying beneficial improvements in body composition and performance, there is potential for an ultra-filtered counterpart to promote similar, if not greater improvements. Recently, commercially available ultra-filtered milk in the form of whole, fat-free, 2% white, and 2% chocolate milk have been produced. This particular product contains half the sugar content, by removing lactose, and nearly a 50% increase in the protein content of their traditional counterparts. Research using chocolate milk supplementation as a post-recovery beverage has shown to improve fractional synthesis rate following endurance exercise, increased aerobic power, improved body composition, increased toque, and increased recovery (Ferguson-Stegall et al., 2009; Gilson et al., 2010; Lunn et al., 2012; Spaccarotella & Andzel 2011; Wallace & Able 2010. To date, no studies have examined these particular types of milk to one another. The benefit of ultra-filtered milk is the ability to supply 25g of protein with without the addition of

excess calories. For instance, 500mL of this product will supply an individual with 27g of protein and carbohydrates resulting in nearly 300kcals. In order to receive 27g of protein, one would have to drink 1000mL of traditional chocolate milk, but this would result in nearly 76g of carbohydrates. Therefore, the purpose of the following dissertation is to compare the effects of higher protein, less sugar content chocolate milk to traditional low fat chocolate milk on (1) strength gains (2) increased performance and (3) body composition following resistance training adaptations.

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Chapter Two

The Effects of Ultra-Filtered Milk Consumption on Strength and Performance Following Resistance Training in Female Collegiate Athletes

ABSTRACT

Purpose: To date, no studies have examined the consumption of ultra-filtered milk to chocolate milk to one following resistance training. The purpose of this study was to identify any unrealized gains of ultra-filtered milk to traditional low-fat chocolate milk on athletic performance and body composition in collegiate, female athletes. Methods: Division I NCAA athletes were counterbalanced to drink either ultra-filtered milk (UFM; n = 8) or chocolate milk (CM; n = 7) immediately following resistance training. Subjects trained 3 days per week for 10 weeks. Body composition changes were measured by dual-energy x-ray absorption. Upper body strength was measured by 3-repition max bench press and lower body strength measured by 5repition max back squat. Performance was measured by 5-10-5 agility shuttle and vertical jump. All measures were taken before and after training. **Results:** Body composition changes were similar for both groups with UFM trending to greater percent changes in body mass (-0.09% to +0.03%), body fat percentage (-3.06% to -2.31%), and lean mass (+1.5% to +0.51%). Both groups experienced time effects for strength and performance measures with CM trending towards greater improvements in bench press (+12.8% to +8.47%), back squat (+12.5% to +11.2%), 5-10-5 (-3.1% to -2.58%), and vertical jump (+13% to +6.99%). Conclusion: Consumption of chocolate milk immediately following resistance training improved strength and performance measures in collegiate, female athletes. While not significantly different from one another, ultra-filtered chocolate milk trended more positively for body composition changes while traditional chocolate milk trended more positively for strength and performance measures.

INTRODUCTION

Resistance training is an important mode of training for athletes as it is shown to increase athletic performance by improving muscular strength, endurance, power, balance, coordination, and hypertrophy (Kraemer & Ratamess 2004). The adaptations experienced by skeletal muscle have been credited to increased rates of protein synthesis as well as neurological adaptations (Gabriel, Kamen, & Frost, 2006; D. R. Moore et al., 2009)(D. R. Moore et al., 2009). Resistance training increases the rate of protein synthesis above and beyond that of protein degradation causing positive skeletal muscle protein balance. Sustained positive protein balance during recovery leads to positive adaptations following resistance training such as hypertrophy, strength gain, and improved body composition (i.e. increased fat free mass and decreased fat mass) (Deschenes & Kraemer, 2002). In addition, proper recovery from bouts of resistance training is critical to maximize the benefits of each training session. Resistance training, especially those of higher intensities, can deplete muscle glycogen causing decreased force production and strength (Haff et al. 2003). One method to optimize benefits of resistance training is the consumption of nutrients around the training session (Beelen, Burke, Gibala, & Van Loon, 2010b; Ferguson-Stegall et al., 2011; Howatson & Van Someren, 2008; Lynch, 2013b; Pritchett et al., 2011; Rankin et al., 2004; Sousa et al., 2014).

Carbohydrate supplementation following training sessions is commonly used to replenish muscle glycogen (Egan & Zierath, 2013; Haff et al., 2003; Macdougall et al., 1999; Schoenfeld, 2010). Haff et al (2000) studied the effects of carbohydrate supplementation on muscle glycogen and resistance training performance on highly resistance trained males. Subjects performed bouts of isokinetic leg exercise before and after isotonic resistance exercise. Compared to a placebo, subjects consuming carbohydrates prior to and during their session experienced less muscle

glycogen degradation. High intensity (e.g. load or volume) resistance training and/or multiple training sessions in one day benefit from carbohydrate supplementation as it restores glycogen levels to resting levels (Haaf et al., 2003). The consumption of carbohydrates within 30 minutes post-exercise, whether in liquid or solid form, is more beneficial in replenishing glycogen stores (Ivy et al., 2008). Similarly, high quality protein is beneficial in the augmentation of resistance training adaptations.

Whey protein and casein protein are considered highly quality proteins due to their amino acid profile and high levels of branched-chained amino acids. The benefits of consuming whey protein around resistance training has been documented in several reviews including: improved rates of muscle protein synthesis, increased hypertrophy, improved body composition (i.e. increases in lean body mass and decreases in fat mass), and increased strength gains (Hulmi et al., 2010; Pasiakos et al., 2014; Pasiakos et al., 2015). The addition of protein to a carbohydrate supplement decreases the amount of carbohydrate needed to maximally increase the rate of muscle glycogen synthesis (J. L. Ivy & Ferguson-Stegall, 2014). Farup et al. (2014) compared differing contraction resistance training (i.e. eccentric vs concentric) in combination with whey protein + carbohydrate or carbohydrate supplementation alone on muscle and tendon hypertrophy. Recreationally active, young men performed maximal knee extensor training with one leg performing eccentric contractions and the other performing concentric contractions. Contractile mode had no effect, but those consuming the whey protein + carbohydrate supplement increased both quadriceps and patellar tendon hypertrophy. A product that combines proteins and carbohydrates would seem ideal to elicit the benefits of the two macronutrients.

Milk contains a combination of high quality proteins (i.e. whey and casein) and carbohydrates, and has been demonstrated to improve strength, hypertrophy, and body

composition in both untrained and resistance trained individuals (Hartman et al., 2007; Josse et al., 2010; Rankin et al., 2004). Chocolate milk, which contains added sugar, has similar benefits as milk. Studies utilizing chocolate milk as a post-exercise recovery drink have shown improvements in aerobic training, body composition, and rate of perceived exertion. Both Lunn et al (2012) and Thomas et al (2009) studies found improved aerobic performance in trained subjects. As for resistance training, Wallace and Able (2010) studied the effects of chocolate milk supplementation on perceived exertion and muscular strength following resistance training in recreationally active males. Compared to a non-caloric placebo, those consuming chocolate milk had lower levels of perceived exertion as well as lower muscle fatigue. Wallace and Able concluded chocolate milk reduce muscular fatigue and be beneficial for those with multiple, daily training sessions.

With the current literature reporting benefits, chocolate milk has been adopted as a posttraining recovery drink for various teams and is currently used at Georgia State University with several varsity teams. Recently, ultra-filtered milks with nearly 50% more protein and 50% less sugar compared to traditional counterparts have become commercially available. UFM (*Fairlife*) undergoes an ultra-filtration process that results in nearly half the carbohydrate content (13g vs. 24g) and nearly 50% more protein (13g vs. 9g) compared to CM (*Glenview Farms*). UFM contains 4.5g of fat compared to 3g in CM. During the filtration process, lactose is also removed from the milk, resulting in lower carbohydrate content. The decreased amount of carbohydrates from the ultra-filtered milk should not have adverse effects compared to the CM (J. L. Ivy & Ferguson-Stegall, 2014). To date, no studies have examined ultra-filtered milk to chocolate milk to one another. Therefore, the purpose of our study was to identify any unrealized gains of UFM and compare the effects of ultra-filtered milk to traditional low-fat chocolate milk on athletic

performance (strength, power, agility) and body composition (body weight, fat-free mass, and fat mass) in resistance-trained collegiate athletes. We hypothesized that athletes consuming ultrafiltered milk would out perform traditional chocolate milk by increasing strength, increasing performance, and improving body composition.

METHODS

Participants. Members of both a Division I NCAA women's beach volleyball team and women's softball team were recruited to participate in the study. Subjects were current roster members and injury free. Participation in the study was voluntary and not required by sport coaches.

Experimental Design. Each group was counterbalanced to achieve an equal number of beach volleyball and softball members per group. Groups were assigned treatments as follows: (1) ultra-filtered chocolate milk (UFM) or (2) 1% Chocolate Milk (CM). Subjects completed an informed consent approved by the Georgia State University Institutional Review Board as well as a medical history questionnaire. Athletes free of injury (i.e., not limited in practice or workouts) were included in the study. Similar to Wilkenson et al (2007), milk products were compared to one another. All groups underwent one week of familiarization for the performance measures located in the Georgia State University Athletic Weight Room. After the familiarization week, one week was spent collecting subjects pre-training strength, performance, and body composition measures. After the pre-testing week, subjects began the ten-week resistance training program based off testing numbers. Immediately following each strength training session, the research staff or a member of the strength and conditioning staff provided

post-workout either UFM or CM (Elliot, Cree, Sanford, Wolfe, & Tipton, 2006a; Wilkinson et al., 2007).

Dietary logs were collected during week 1, week 5, and week 10 as previously described (Burke et al., 2001; Josse et al., 2010; Willoughby, Stout, & Wilborn, 2007). During these weeks, dietary logs were kept for three days of each week: one day for strength training, one day for speed/conditioning training, and one day for an off day. Subjects used MyFitnessPal as an electronic log to keep track of their food. Total caloric intake as well as macronutrient intake was collected using the USDA nutrient database and compared between the two groups. Following the ten weeks of training, one week was used to collect post-training strength, performance, and body composition measures.

Experimental Methods.

Performance Measures. Performance measures were taken the week before week one of training (PRE) as well as one week after week ten of training (POST). Upper body strength was assessed via three repetition maximal effort barbell bench press (3RM BP). A successful rep for bench press was considered when the athlete made contact with their chest and returned to a locked out position. Lower body strength was assessed via five repetition maximal effort barbell back squat (5 RM BS). For the back squat, a successful rep was considered once the athlete's knees reached or surpassed the hips and returned to an extended position. Subjects used 4-5 warm-ups and were advised by coaching staff on weight increases per attempt. Vertical jump (VJ) was assessed using the VERTEC (Sports Imports, Columbus, Ohio) vertical jump assessment tool. To score VJ, each subject performed two attempts jumping off two feet without stepping. The average of the two scored. A 5-10-5 shuttle run was used to assess speed and agility. The 5-10-5 shuttle run

consists of sprinting forward five yards, quick change of direction sprint ten yards backwards (not a backwards sprint), and finish by sprinting forward five yards. The 5-10-5 shuttle was manually recorded via stopwatch. Subjects performed two attempts with the average of the two scored. A member of the coaching staff was assigned to collect one measure and was responsible for that exercise for both pre-and post- testing (e.g. coach one collects every bench press attempt). For the strength and performance measures, two minutes of rest was allotted between attempts. Testing measures and training sessions occurred Monday, Wednesday, and Friday at 8:00am.

Body Composition. Height, body weight, and body composition (i.e. fat-free mass and fat mass) were measured one week prior to training (PRE) and again one week following training (POST). Body composition was measured using dual-energy X-ray absorptiometry (DEXA, Lunar Prodigy, General Electric, Madison, WI).

Resistance Training Protocol. Resistance training was performed three days per week with two days of speed and agility training using a periodized model designed in collaboration with National Strength and Conditioning Association certified coaches in the GSU athletic department (Table 1). Specifically, weeks 1-3 focused on hypertrophy development with rep schemes ranging from 8-12 and intensity (i.e. percent of 1RM) ranging from 50-65% 1RM. Weeks 4-7 focused on strength development with reps ranging from 1-5 and intensity ranging from 65-90% 1RM. Week 8 was used as a de-load week, followed by weeks 9 and 10 for peaking. During Weeks 9 and 10, one to two reps were used at high intensities of 85-100% 1RM. Strength training sessions were total body (e.g. bench press, squat, deadlift, and jumping) focused on strength, power, and hypertrophy development. The first two days (Monday and Wednesday) of training were focused primarily on total body hypertrophy, power, or strength development

depending on the week (see Table 1). Day 3 (Friday) differed as it focused on muscular endurance and conditioning. The goal was to complete prescribed rounds of 15-25 reps of wall ball shots, med ball drop squat, and burpees. Additionally, the following exercises were performed in sand for a total of 100ft: trap bar carries at 135lbs, kettlebell rope pulls at 70lbs, plate drags at 45lbs, and plate lunge at 25lbs to broad jump. Table 2 displays exercise information for the training program.

Table 1. Weekly breakdown of 10-week training program.

Weeks 1-5					
1RM%	50-55%	55-60%	60-65%	65-75%	85-100%
Sets x Reps	4x12	4x10	4x8	6x5	2,2,1,1
Weeks 6-10					
1RM%	80%	90%	60-75%	85%	85-100%
Sets x Reps	4x4	4x2	6x3	2x1	2,2,1,1,1

RM = Repetition Max

 Table 2. Exercise selection for10-week training program

Day 1	Day 2	Day 3
Back Squat	Deadlift	Wall Ball Shots
Jump/Swing	Jump/Swing	Trap Bar Carry
Overhead Press	Bench Press	Medicine Ball Drop Squat
Overhead Pull	Horizontal Pull	Kettlebell Rope Pulls
Romanian Deadlift	Glute Ham	Burpees
Dumbbell Curl to	Dumbbell Pull Overs	Plate Drags
Press		Plate Lunge to Broad Jump

Chocolate Milk Supplementation. Participants were blinded to which milk they received. In order for both groups to receive similar caloric content, the UFM (*Fairlife*) group received 500mL (~16.9oz) while the CM (*Glenview Farms*) group received 16oz (~473mL). For the UFM group, this resulted in 296 calories (9.5g fat, 27.5g protein, 27.5g carbohydrate). For the CM group, they received 300 calories (6g fat, 18g protein, 58g carbohydrate). Subjects had 30 minutes following each training session to consume their assigned drink under the supervision of the coaching staff.

Statistical Analysis. Power analyses were performed using G*Power (Heinrich-Heine-Universität Düsseldorf). A 2 x 2 repeated measures (group x time) mixed ANOVA was used to analyze changes for each measure from pre- to post-program within and between groups. Statistical analysis was performed with the Statistical Package for the Social Sciences (SPSS; V21.0.) Statistical significance was set at $p \le 0.05$.

Results

Participants. Twenty-four athletes were recruited for the study. Of those, 6 withdrew from the study (3 from each group; 3 dropped for injuries sustained outside of the study; 2 dropped for personal reasons; 1 dropped from adverse effects of milk) and another 3 were unable to complete all strength and/or performance measures (2 from UFM group and 1 from CM group) leaving a total of 15 (UFM, n = 8; CM, n = 7). Both groups were of similar age (UFM: $20.67 \pm .88$; CM: $19.58 \pm .42$) and not statistically different. Compliance with consumption of post-training drinks was consistent between both groups as well as completing each training session.

Body Composition.

No significant time effects or group by time interactions were experienced for any variable (Table 3), but trended towards improvements. Body mass, total body fat percentage, fat mass, and lean mass were not different at baseline. Body mass remained consistent for both groups over the course of training. Total body fat percentage and fat mass saw small decreases. Lastly, lean body mass remained constant through training. Percent changes from pre-to-post testing are displayed for body mass all measures in figures 4a-4d.

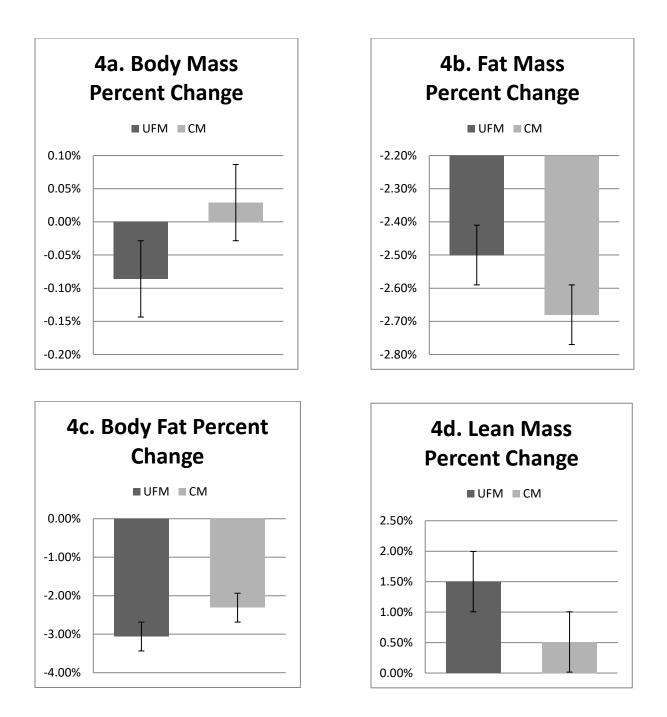


Figure 4a-4d: Changes are expressed in percent changes from pre to post data collection. Dark grey bars represent ultra-filtered chocolate milk = UFM. Lighter grey bars represent chocolate milk = CM. Error bars represent SEM. 4a represents percent changes from pre to post testing for changes in body mass. 4b represents percent changes from pre to post testing for changes in fat mass. 4c represents percent changes from pre to post testing for changes in total body fat percentage. 4d represents percent changes from pre to post testing for changes in lean mass.

Table 3. Group Body Composition

	UFM (n = 8)		CM (N = 7)		
	PRE	POST	PRE	POST	ANOVA Group x Time Interaction
Body Mass (kg)	68.9±6.8	68.7±6.7	67.3±4.9	67.4±5.6	p = .814
Total Body Fat Percentage	26.5±3.2	25.3±4.0	26.7±4.0	25.8±3.8	p = .716
Fat Mass (kg)	17.5±3.4	16.7±4.0	17.4±3.7	16.8±4.0	p = .881
Lean Mass (kg)	48.0±4.1	48.8±3.1	47.2±2.3	47.6±2.8	p = .513

Weight in kilograms. Total body fat in percentage. Fat mass in kilograms. Lean mass in kilograms. Expressed in Mean ± Standard Deviation. UFM = Ultra-filtered chocolate milk. CM = chocolate milk.

Performance & Strength Measures.

No significant group by time interactions for any strength or performance measure was observed. The UFM group was significantly stronger than the CM group at baseline for the 3RM bench press (p = .016). There was a time effect as both groups increased all strength and performance measures from baseline. These results are displayed in Table 5. Percent changes from pre-to-post testing for both strength and performance measures are displayed in figures 5a-5d.

	UFM (n = 8)		CM (N = 7)		
	PRE	POST	PRE	POST	ANOVA Group x Time Interaction
5-10-5 Agility (sec)	5.2±.24	5.1±.18	5.3±.23	5.2±.17	p = .467
Vertical Jump (cm)	51.6±9.1	55.88±9.4	49.8±5.6	59.2±5.6	p = .904
3 RM Bench Press (kg)	43.4±4.3	47.9±5.3	36.6±5.1	43.4±3.6	p = .020*
5 RM Back Squats (kg)	68.0±11.4	77.1±13.9	63.5±5.1	73.9±5.4	p = .448

Table 4. Group Performance and Strength

5-10-5 in seconds. Vertical Jump in centimeters. Bench Press in pounds. Squat in pounds. Expressed in Mean \pm Standard Deviation. UFM = Ultra-filtered chocolate milk. CM = chocolate milk. RM = repetition max. * indicated significance at the alpha <0.05 level.



Figure 5a-5d: Changes are expressed in percent changes from pre to post data collection. Dark grey bars represent ultra-filtered chocolate milk = UFM. Lighter grey bars represent chocolate milk = CM. Error bars represent SEM. 5a represents percent changes from pre to post testing for changes in 3-repitition max bench press. 5b represents percent changes from pre to post testing for changes in 5-repitition max. back squat 5c represents percent changes from pre to post testing for changes in agility shuttle performance. 5d represents percent changes from pre to post testing for changes in vertical jump performance.

Dietary Intake.

Subjects were inconsistent with submission of food logs. Due to a lack of submitted food logs, sufficient dietary data was not attained. From the UFM group, 5 subjects submitted food logs and for the CM group, 4 subjects submitted logs. Of those subjects, all submitted logs for week one, 3 from UFM at week 5, and all from CM at week 5. No subjects submitted logs at week ten. The last log submitted were used, but varied from weeks 7, 8, and 9. Only one subject from UFM submitted a log past week 5. Table 6 displays the daily caloric intake and macronutrient intake for both groups.

	Week 1 $(n = 5)$	Week 5 $(n = 3)$	
	1647.8 kcal/day	1772.3 kcal/day	
UFM	197.67 g/day carbohydrate	211.22 g/day carbohydrate	
OFM	66.27 g/day fat	68.67 g/day fat	
	71.47 g/day protein	66 g/day protein	
	Week 1 (n = 4)	Week 5 $(n = 4)$	
	1473.0 kcal/day	1570.4 kcal/day	
СМ	181.5 g/day carbohydrate	169.92 g/day carbohydrate	
CIVI	56.58 g/day fat	70.67 g/day fat	
	57.17 g/day protein	67.41 g/day protein	

Table 5. Energy consumption by group is listed in kcal/day (1kcal = 4.184 kjoule). Ultra-filtered milk = UFM. Chocolate = CM. g = grams. Food logs were collected via the MyFitnessPal app. Electronic logs were emailed to the researchers and analyzed via the USDA Food Composition Database. Logs were collected after weeks 1, 5, and 10 of training. Insufficient data were collected during week 10; only 4 (UFM = 1; CM = 3) subjects submitted logs, therefore the data were not reported.

Discussion

We report no differences in body composition, strength, and performance measures

between ultra-filtered chocolate milk and chocolate supplementation immediately following 10

weeks of resistance training. Regardless of post-exercise beverage, a 3 d/week, 10-week resistance training program was able to increase strength and performance measures above pre-training values.

Our data support current literature regarding chocolate milk supplementation following resistance training. These results are similar to studies that utilize milk proteins (e.g. whey and casein protein). Wilborn et al. (2013) compared the effects of whey protein supplementation versus casein protein on female, NCAA Division III collegiate basketball players following 8 weeks of resistance training. Similar to our study, their study found no difference between proteins, but saw both groups increase performance in regards to lower body strength, upper body strength, vertical jump, broad jump, and 5-10-5 agility shuttle time. Similarly, Josse et al (2010) observed increases in upper body strength in young, recreationally active healthy females following 12 weeks of resistance training.

However, we were unable to observe changes in body composition. The consumption of milk surrounding resistance training influences the rate of skeletal muscle protein synthesis, increasing hypertrophy (Elliot et al., 2006a; Wilkinson et al., 2007). The increases in muscle hypertrophy improve body composition by increasing lean mass and decreasing fat mass (Hartman et al., 2007; Josse et al., 2010; Wilborn et al., 2013). Although body composition improvements were not statistically significant, UFM trended towards changes that are more positive in body mass, lean mass, and decreases in fat mass. One possible explanation why we failed to see changes in body composition was the rigor of training program. For example, our subjects engaged in a 10-week program compared to the Hartman and Josse studies, which had their subjects resistance train 5 days a week for 12 weeks. It should be noted our participants at baseline had lower fat mass and greater lean mass than those within the Josse study. It is

plausible that our athletes did not experience significant changes in body composition due to their training background (NCAA Division I athletes compared to untrained) and had less room for improvement compared to an untrained counterpart.

Because our subjects were collegiate athletes engaged in pre-season resistance training programs, we were unable to control for dietary intake. While we were not able to control for the diet, we aimed to assess and compare food logs. Due to our study not controlling for diet, as well as depending on dietary recall, we faced noticeable limitations regarding dietary intake. In order to compare dietary intake, athletes were asked to log their food consumption 3 days per week similarly to Josse et al (2010). We attempted to collect food logs from athletes at week 1, week 5, and week10 of the study. This collection method was similar to previous studies (Burke et al., 2001; Josse et al., 2010; Willoughby et al., 2007). Unfortunately, we were unable to collect consistent data from both groups due to a lack of response to emails requesting logs. Therefore, we cannot rule out the possibility that dietary intake played a role in the lack of body composition changes. The training period ended around Thanksgiving break for our studentathletes. Due to the testing week ending the Friday before Thanksgiving, participants were unresponsive to emails during that week off from school. To address this limitation, future studies could benefit from collecting food logs in person as opposed to digitally. For instance, with a Monday, Wednesday, Friday training schedule, hand out physical logs on Monday, athletes log for Monday (RT day) and Tuesday (non-RT day), and collect logs on Wednesday. On Friday, hand out another log for athletes to record their weekend day then collect logs on Monday.

Another limitation from the lack of consistent dietary data is the comparison of protein intake following training. Week 1 was the only week where comparable logs were submitted. At

week 1, the UFM group consumed 1.03g/kg bodyweight of protein compared to the .85g/kg bodyweight of the CM group. At week 5, the UFM group consumed 0.96g/kg and the CM group consumed 1.0g/kg body weight; this does not include the added protein content of either chocolate milk. The UFM contained 27.5g of protein while the CM contained 18g of protein. Without dietary data, it is difficult to tell if the 9.5g of protein consumed immediately after would play a role in adaptations. Previous research would suggest that minor of a difference would not play a role due to both groups receiving what is considered near optimal (~20g), or above, protein following training (Witard et al., 2014). Furthermore, the timing of the protein intake may not have played as great of a role in adaptations due to their inadequate total protein intake indicated from weeks 1 and week 5. The consumption of adequate protein (~1.6g/kg body weight; Wu, 2016) throughout the day in combination with resistance training is imperative for maximizing skeletal muscle protein accretion (Schoenfeld, Aragon, and Krieger 2013). Without the dietary logs, we were unable to assess if the athletes were meeting recommended daily protein intake. Future studies should attempt to collect dietary logs in person or control for diet.

Another limitation is the practicality of working with collegiate athletes due to the inability to include a placebo within the study. The athletes within this study were accustomed to receiving a post-training serving of milk, during previous semesters and the coaches were not willing to have their athletes receive a placebo. It should be noted these particular athletes were trained during the fall semester and were not receiving milk supplementation during three months prior to commencing training, as they were not training on campus during the summer. Furthermore, athletes who consumed protein powders within 30 days prior to the study were not included in the study. The lack of a placebo group prevents a direct comparison of effects on supplementation versus non-supplementation. It is plausible to consider the changes in strength

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and performance was due to both the training protocol and supplementation because both groups consumed a nutrient dense and calorically similar product following training. The aim of this study was to identify unseen benefits of ultra-filtered milk in collegiate beach volleyball and softball players. Due to the use of this specific population, total number of participants were limited. Comparatively, the availability of highly trained athletes compared to the general population adds another limitation to our study.

Lastly, our study was underpowered with 15 subjects. To achieve an effect size of f=0.5 and 0.95 power with an alpha of 0.05 regarding strength, performance, and body composition measures a total of 34 subjects needed to be recruited. There are 42 combined members for the beach volleyball and softball teams. The population pool for elite athletes is limiting. It must also be noted that participation in the study was voluntary and not a requirement by the coaches. Of the 42 members, 24 voluntarily participated for the study. With a limited recruitment pool and unforeseen circumstances such as injuries sustained outside of training further limit available athletes to recruit. One potential benefit from our study was the ability to train overhead athletes from two different sports simultaneously. While a benefit for our study, not every sport trains identically, thus reducing the capability to conduct this study with numerous teams. Typically, different teams utilize different training programs as well as different members of the strength and conditioning staff. While having the two teams train together improved our recruitment capabilities, future studies would benefit from potentially recruiting several athletes over numerous off-season training periods. Maintaining similar progressions for major lifts, such as bench press, squat, or deadlift over several off-seasons would help ensure the training program is similar over multiple off-seasons. This is ideal for a laboratory setting, but is difficult to implement as strength coaches typically vary aspects of their training program over the years. If

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these changes can be made, this could provide researchers the ability to conduct a longitudinal study over several seasons or increase recruitment by recruiting more athletes over time. Another possibility would be to collaborate with multiple institutions with the same sports and conduct the study, increasing overall participants.

In conclusion, no group differences were observed between the ultra-filtered chocolate milk and the traditional chocolate milk post workout supplementation in regards to strength, performance, or body composition. Both groups were able to increase strength and performance measures from pre to post testing. These results lend themselves as an option for lactose intolerant athletes to use ultra-filtered milk as a post resistance training supplement. Also, milk provides NCAA athletes the opportunity to consume a product to help increase strength and performance that is a natural food product without the potential of taking a product that contains banned substances.

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