Resistance training and co-supplementation with creatine and protein in older subjects with frailty: a small-scale exploratory study

Jodie Collins

Submitted in fulfilment of the requirements for the degree of
Master of Science (Sport and Exercise Science)

Centre for Sport Science and Human Performance

Waikato Institute of Technology

Supervisor: Prof Bruno Gualano,

Co-Supervisor: Dr Glynis Longhurst

July 2015

Keywords

Ageing, Creatine, Lean mass, Strength, Whey protein

Abstract

This is a small-scale exploratory trial from the Pro-Elderly study ("Protein Intake and Resistance Training in Aging") aimed at gathering knowledge on the feasibility, efficacy and safety of co-supplementation with creatine and protein supplementation, in conjunction with resistance training, in older individuals with frailty.

Methods: A 14-week, double-blind, randomized, parallel-group, placebo controlled exploratory trial was conducted between in Hamilton (New Zealand). The subjects were randomly assigned to compose either one of the following groups: 1) whey protein and creatine co-supplementation (WHEY+CR) or 2) whey protein supplementation (WHEY). All the subjects undertook a supervised exercise training program for 14 weeks and were assessed at baseline and after 14 weeks.

The main dependent variables were muscle function (handgrip, timed-stands and timed-up-and go tests) and body composition (free fat mass, fat mass, and bone mass). Self-reported adverse events were recorded throughout the trial and blood parameters were assessed.

Results: We found a main time-effect in handgrip (WHEY+CR = 26.65 ± 31.29; WHEY = 13.84 ± 14.93 Kg; $p = 0.0005$), timed-up-and-go (WHEY+CR = -11.20 ± 9.37; WHEY = -17.76 ± 21.74; $p = 0.006$ s), and timed-stands-test (WHEY+CR = 47.50 ± 35.54; WHEY = 46.87 ± 24.23 reps; $p = 0.0001$), suggesting that WHEY+CR and WHEY were similarly effective in improving muscle function.

Exploratory analyses further suggested that most of the subjects experienced improvements in all muscle function tests. In addition, all of the subjects showed improvements in at least two of the three tests, regardless of their treatments. Neither within- nor between-group differences were detected in any of the body composition variables (all $p > 0.05$). No important adverse effect was observed and blood parameters remained unaltered.

Conclusion: Co-supplementation with creatine and whey protein was tolerable and free of adverse events in older subjects with frailty undertaking resistance training. Furthermore creatine supplementation did not augment the adaptive effects of resistance training along with whey protein on body composition or muscle function in this population.

This exploratory study will be insightful in designing a larger and more comprehensive randomized controlled trial to confirm or refute these findings. Clinicaltrials.gov: NCT01890382.

Table of Contents

Keywords i

Ageing, Creatine, Lean mass, Strength, Whey protein Abstract i

Table of Contents iv

List of Figures v

List of Tables vi

Statement of Original Authorship vii

Acknowledgements viii

CHAPTER 1: INTRODUCTION 1

1.1 Background 1

1.2 Context 1

1.3 Purposes 2

CHAPTER 2: LITERATURE REVIEW 3

2.1 Historical Background 3

2.2 Creatine 3

2.3 Whey Protein 4

2.4 Summary and Implications 5

CHAPTER 3: RESEARCH DESIGN 6

3.1 Methodology and Research Design 6

3.1.1 Methodology 6

3.1.2 Research Design 6

3.2 Participants 7

3.3 Instruments 7

3.4 Procedure and Timeline 8

3.5 Analysis 9

3.6 Ethics and Limitations 10

CHAPTER 4: RESULTS 11

CHAPTER 5: CONCLUSIONS 21

REFERENCES 24

APPENDICES 27

Appendix A Timed up & go protocol 27

Appendix B Timed Stand protocol 28

Appendix C Resistance Training Program 29

Appendix D Participant Forms 31

List of Figures

Figure 1 **Flowchart of subjects included in the study (n=18) and subjects who withdrew (n=2) during the study.....11**

Figure 2. **Results from the handgrip, timed up-and-go and the timed stand and sit test showing similar improvements between both groups. 14**

Figure 3. **No significant differences between groups (p>0.05).....15**

List of Tables

Table 1. ***Main characteristics of subjects*** **.....12**

Table 2. ***Food intake variables before and after supplementation with creatine and protein (CR+WHEY or whey supplementation alone (WHEY) in older subjects undertaking resistance training.*** **.....13**

Table 3. ***Blood variables before and after supplementation with creatine and protein (CR+WHEY) or whey supplementation alone (WHEY) in older subjects undertaking resistance training.*** **.....16**

Table 4. ***Data for muscle function and body composition data at Pre and Post*** **.....19**

Statement of Original Authorship

The work contained in this thesis has not been previously submitted to meet requirements for an award at this or any other higher education institution. To the best of my knowledge and belief, the thesis contains no material previously published or written by another person except where due reference is made.

Signature: 

Date: 10 July 2015

Acknowledgements

Fonterra (New Zealand) and Alzchem (Germany) who donated the supplements for this study.

Todd Foundation and WINTEC for Grants to fund medical testing for this study.

Bruno Gualano and Hamilton Roschel are supported by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq).

Chapter 1: Introduction

1.1 BACKGROUND

With the ageing process, the body goes through physiological changes; of most interest are the declines in lean muscle mass. Longitudinal studies suggest that there is a gradual loss of muscle mass from the age of 40. It is estimated to reduce at a rate of 0.64 - 1.0% per year from the age of 75, additionally muscle strength is shown to reduce at a rate of 2.5 – 4.0% (Mitchell, et al., 2012). In elderly this loss of muscle mass and strength also results in a loss of muscle function which vastly impairs the ability to perform daily tasks and increases the risk of falls and illness. If resistance training with supplementation of whey protein and creatine could increase lean muscle mass and strength faster and if it was in safe tolorable amounts we could see a marked improvement in the health of our elderly.

1.2 CONTEXT

The ageing population in New Zealand is growing at a high rate and the living age expectancy is increasing. It is estimated that by 2036 23% of our population will be over the age of 65 (Statistics New Zealand, 2000), this in turn increases the demands on health, disability and social services.

Through resistance training lean muscle mass and strength in the elderly improve. Increasing lean muscle mass and strength can decrease the rate of muscle loss and improve muscle function. This will reduce the risk of falls and age related diseases along with improve recovery following illness or surgery (Hunter, McCarthy, & Bamman, 2004).

With the use of supplementation such as whey protein and creatine is believed to improve lean muscle and strength at a faster rate than resistance training alone.

1.3 PURPOSES

This is a small-scale exploratory trial from the Pro-Elderly study ("Protein Intake and Resistance Training in Aging") aimed at gathering knowledge on the feasibility, efficacy and safety of co-supplementation with creatine and protein supplementation, in conjunction with resistance training, in older individuals with frailty.

Chapter 2: Literature Review

2.1 HISTORICAL BACKGROUND

Resistance training has been established as important in the prevention and treatment of disabilities and co-morbidities secondary to aging, increasing muscle function and lean mass (Cadore & Izquierdo, 2013), (Breen & Phillips, 2011). Furthermore, there is evidence suggesting that some dietary interventions, including creatine and whey protein supplementation, can act synergistically to resistance training by enhancing its beneficial effects in elderly individuals (Gualano, et al., 2014), (Gualano, Roschel, Lancha-Jr, Brightbill, & Rawson, 2012), (Wall, Cermak, & van Loon , 2014), (Tieland, et al., 2012), (Neves, et al., 2011).

2.2 CREATINE

Creatine plays an important role in rapid energy provision during muscle contraction involving the transfer of N-phosphoryl group from phosphorylcreatine to ADP in mitochondria to regenerate ATP through a reversible reaction catalyzed by phosphorylcreatine kinase (CK). In addition to its function as a temporal energy buffer, phosphorylcreatine also acts a spatial energy buffer to shuttle high-energy phosphates between mitochondria and cellular ATP utilization sites (Wallimann, Wyss, Brdiczka, Nicolay, & Eppenberger, 1992).

It has been systematically shown that creatine supplementation can increase muscle creatine and phosphylcreatine content, thereby leading to improvements in training volume, which, in turn, could translate into greater adaptations to resistance exercise. In fact, there is growing evidence demonstrating creatine supplementation and resistance training seem to act synergistically in improving lean mass, fatigue resistance, muscle strength, performance of activities of daily living largely than resistance training alone (Tarnopolsky, et al., 2007) (Candow, et al., 2008) (Eijnde, et al., 2003).

However, most studies have primarily focused on healthy elderly individuals. Recently, we showed that creatine supplementation was able to enhance the effects of resistance training on appendicular lean mass and muscle function in vulnerable older individuals (i.e., those who commonly complain of being ‘‘slowed up’’ or have

disease symptoms) (Gualano, et al., 2014). Nonetheless, the number of studies involving older frailer individuals being supplemented with creatine is very limited.

2.3 WHEY PROTEIN

Another dietary intervention potentially able to improve lean mass and muscle function in the aging population is protein supplementation. Indeed, recent evidence has shown that higher dietary protein ingestion is beneficial to support good health, promote recovery from illness, and maintain functionality in older adults (Bauer, et al., 2013). According to recent review papers, the need for more dietary protein is in part because of a declining anabolic response to protein intake in older people; more protein is also needed to offset inflammatory and catabolic conditions associated with chronic and acute diseases that occur commonly with aging (Breen & Phillips, 2011) (Wall, Cermak, & van Loon , 2014) (Bauer, et al., 2013) (Wolfe, 2013).

Furthermore, older individuals usually eat less protein compared to younger adults (Deutz, et al., 2014). Importantly, an imbalance between protein supply and protein can result in loss of skeletal muscle mass because of a chronic disruption in the balance between muscle protein synthesis and degradation (Breen & Phillips, 2011) (Wall, Cermak, & van Loon , 2014). As a result, older individuals may lose muscle mass and strength and eventually experience physical disability. On the other hand, dietary protein supplementation promotes protein synthesis and can enhance recovery of physical function in older individuals (Breen & Phillips, 2011) (Wall, Cermak, & van Loon , 2014) (Bauer, et al., 2013) (Wolfe, 2013).

Interestingly, a recent meta-analysis provided evidence that protein supplementation can augment the adaptive response of skeletal muscle to resistance training in the aging population (Cermak, Res, de Groot, Saris, & van Loon, 2012). However, caution should be exercised because longer-term studies investigating the possible synergistic effects of protein supplementation and resistance training in older individuals remain scarce.

2.4 SUMMARY AND IMPLICATIONS

Studies exploring the potential beneficial effects of co-supplementation with protein and creatine in resistance trained older individuals. Bemben et al., showed that a resistance-training program for 14 weeks in middle-aged and older men (48-72 yrs.) was able to increase muscular strength and muscle mass with no additional benefits from creatine (5 g/day) and/or whey protein supplementation (35 g/day) (Bender, et al., 2005). Similarly, Villanueva et al., demonstrated that creatine (0.3 g/kg/day for 5 days followed by 0.07 g/kg/day) and whey protein (35g/day) supplementation did not provide additional benefits in older adults (60-80 yrs.) performing 12 weeks of periodised resistance training to augment muscular and functional performance (Villanueva, He, & Schroeder, 2014).

Furthermore, Eliot et al. failed to show any effect of supplementation with creatine (5g/day) and whey protein (35 g/day), alone or combined, in middle-aged men (48-72) undergoing 14 weeks of resistance training (Eliot, Knehans, Bemben, Witten, & Bemben, 2008). In contrast, Candow et al., showed that creatine (0.1 g/kg/d) combined with protein supplementation (0.3 g/Kg/d) during a 10-week resistance training program increased lean tissue mass and bench press strength (but not leg press strength), as well as reduced muscle protein degradation and bone resorption in older individuals (59-77 yrs.) (Candow, et al., 2008).

These findings are hard to reconcile because of the heterogeneous characteristics of samples, training protocols, supplementation regimens, and research outcomes. Furthermore, these studies evaluated apparently heathy subjects; to date, no study has assessed the effects of co-supplementation with creatine and protein, along with resistance training, in older individuals with frailties.

Chapter 3: Research Design

3.1 METHODOLOGY AND RESEARCH DESIGN

3.1.1 Methodology

A 14-week, double-blind, randomized, parallel-group, placebo controlled exploratory trial was conducted between March 2014 and December 2014 in Hamilton (New Zealand). This study is part of a larger clinical trial aimed at investigating the role of proteins, amino acids and derivatives along with resistance training in pre-frail and frail older subjects ("The Pro-Elderly Study", registered at clinicaltrials.gov as NCT01472393). This small-scale exploratory study will be followed by a larger-sample confirmatory study to be conducted in Sao Paulo (Brazil). This two-stage analysis consents flexibility in analyses, while allowing further planning on how to confirm the findings herein (Nuzzo, 2014).

The subjects were randomly assigned (1:1) to either one of the following groups: 1) whey protein and creatine co-supplementation (WHEY+CR) or 2) whey protein supplementation (WHEY). All the subjects undertook a supervised exercise training program for 14 weeks. The subjects were assessed at baseline and after 14 weeks. Self-reported adverse events were recorded throughout the trial and blood parameters (i.e., urea, creatinine, sodium, potassium, bilirubin, alkaline phosphatase, gamma glutamyltransferase, alanine aminotransferase, aspartate aminotransferase, glucose, cholesterol, HDL, LDL, triacylglycerides, and creatine kinase) were assessed before and after the intervention.

Subjects were given a food diary to record their dietary intake daily for 7 days before trial and then daily throughout the trial. This was to determine dietary status prior to trial and to monitor changes in food habits and changes in gastrointestinal effects during trial.

3.1.2 Research Design

This is a quantitative study with the main dependent variables being muscle function (handgrip, timed-stands and timed-up-and go tests) and body composition (free fat mass, fat mass, and bone mass).

3.2 PARTICIPANTS

The subjects were recruited into the study by advertising at newspapers, newsletters, rest homes, retirement villages, local health centers, and agencies. The sample was comprised by postmenopausal women and men aged ≥ 65 years physically inactive. The exclusion criteria were cancer, uncontrolled cardiovascular diseases and/or musculoskeletal disturbances that could preclude exercise participation, and any difficulties to consume the supplements.

All the participants were classified into one of the following categories: "3 - well, with treated comorbid disease" (i.e., those whose disease symptoms are well controlled compared with those in category 4); "4 - apparently vulnerable" (i.e., those who commonly complain of being ''slowed up'' or have disease symptoms); or "5 – mildly frail" (those with limited dependence on others for instrumental activities of daily living), according to the Canadian Study of Health and Aging (CSHA) clinical frailty scale, which varies from 1 ("very fit") to 7 ("severely ill") (Rockwood, Song, MacKnight, Bergman, Hogan, & McDowell, 2005). The study was approved by the local ethical committee and all of the subjects signed the informed consent.

3.3 INSTRUMENTS

Muscle function assessments: Handgrip test assesses the isometric strength using the Jamar Handgrip Dynamometer.

Timed-up-go test assesses the time that a subject requires to rise from a standard arm chair, walk to a line on the floor three meters away, turn, return, and sit down again. This is a functional muscle test. (Podisiadlo & Richardson, 1991) (Appendix A).

Timed-stand test evaluates the number of stand-ups that a subject can perform from a standard armless chair for 30 seconds. (Newcomer, Krug, & Mahowald, 1993). (Appendix B).

Muscle strength

1 Repetition Max (RM) was used. It describes the mathematical relationship between the numbers of repetitions performed to fatigue. It is less intensive than the actual 1RM, with less muscle soreness or chance of injury. The 10RM is considered more practical, and a safer alternative to maximal exertion testing.

Bone mineral density and body composition: Bone mineral density and body composition was measured by dual energy x-ray absorptiometry (DEXA).

3.4 PROCEDURE AND TIMELINE

Prior to muscle function assessments, two familiarisation tests were conducted on two separate days with at least 48 hours between trials. Muscle function was measured by handgrip, timed-stands and timed-up-and-go tests.

All subjects underwent a 14-week supervised resistance training program (Appendix C). Exercise sessions occurred twice-a-week and were monitored by fitness professionals. The exercise program was performed at the WINTEC (Waikato Institute of Technology) gymnasium (Hamilton, New Zealand).

Resistance training program was comprised of step-ups, shoulder press, squats, seated row, leg press, biceps curl, triceps extension, calf rise, sit-ups, and leg extension. The subjects were required to perform a five minute warm-up on a stationary bike followed by three sets of 8 repetition maximum of all 10 exercises. Two-min rest intervals were allowed between sets. Progression of exercise load was implemented when the subject could perform more than 8 repetitions on a given exercise set. The workout was then finished with a cool-down which consisted of a series of stretches. All of the following physical tests were performed prior to the 14 week program and after the program:

Handgrip tests: the dynamometer was adjusted for the subjects hand grip, the dynamometer was then set to zero. The subject stood in an upright position holding a dynamometer with the dominant hand, with arm at 90 degrees in front of them. The subject was asked to squeeze and lower the arm to rest beside their leg. Three maximal attempts (with 1-min interval between them) are performed, all scores were recorded.

Timed-up-go test: A Chair with arms was positioned behind a line marked out with tape. Another line was marked exactly three meters away. Subjects were asked to sit upright in the chair on the command they were to stand and walk the 3 meters, turn and sit down. The timer was started and was not stopped until the subject was fully seated. Subjects performed three attempts and all scores were recorded.

Sit and stand test: A chair with no arms was used, subjects were asked to sit upright in the chair, the must refrain from touching the back of the chair. Subjects

were then instructed on the command to stand and sit as many times as they could in 30 seconds. A stopwatch was used and a countdown of 3 seconds was given.

10RM Assessment: Fitted with heart rate monitors subject the initial exercise weight was selected based on subjects body weight and perceived ability. Subjects were asked to perform the exercises in a slow and controlled manner the load was conservatively increased until the subject performed ten but not eleven repetitions.

The following exercises were performed: Leg extension, seated shoulder press and leg press. All exercises were performed in accordance to the exercise protocol for the specified weighted machines. The subjects 1RM will be predicted using the Brzycki mathematical equation. Weight ÷ (1.0278 - (0.0278 × Number of repetitions)).

Blood tests were conducted at Medlab and Pathlab Laboratories situated in Hamilton, Waikato, New Zealand, prior to starting and after finishing the 14 week resistance and supplementation period. Subjects were required to fast at least 10 hours prior to blood tests.

DEXA scans were performed using a GE Linar DPX MD+densitometer at Med-imaging situated in Hamilton, Waikato, New Zealand, analysis software used was encore 2013 Version 15 SP2. Two scans were carried out at the start of the program and after finishing the 14 week resistance training and supplementation period.

Bone mineral density was determined at the following sites: lumbar spine, femoral neck, total femur, and whole body. The precision errors for bone mineral density measurements were determined based on the standard protocols from The International Society for Clinical Densitometry (ISCD) (Shepherd, et al., 2006). The least significant change is considered to be 0.033 g/cm^2, 0.039 g/cm^2, and 0.010 g/cm^2 for lumbar spine, femoral neck, femur, and whole body, respectively (Lopes, et al., 2011).

3.5 ANALYSIS

Data was analysed using mixed-model analysis for repeated measures with Kenward-Roger correction for unequal samples. Tukey *post-hoc* was used when necessary. In addition, absolute delta changes were compared between groups using unpaired two-tailed T-tests. Effect sizes (ES) were calculated for the main dependent variables, according to Cohen (Cohen, 1988). Fisher's test was applied to assess

possible between-groups differences in the incidence of chronic diseases and use of supplements and medications.

Data are presented as mean ± standard deviation, delta scores, and individual responses. The significance level was previously set at $p < 0.05$.

3.6 ETHICS AND LIMITATIONS

Full ethics approval was granted by the Wintec Ethics Committee December 2013. Subjects were invited to an information morning where a presentation of the study was outlined, along with their requirements as participants to fulfil the study. Questions were encouraged before subjects filled in consent forms and questionaries (Appendix D). Before commencing on the study subjects were required to seek medical approval.

Chapter 4: Results

Subjects

A total of 47 subjects were screened for participation and 18 met the inclusion criteria. These patients were randomly assigned into WHEY+CR (n = 9) and WHEY (n = 9). Two subjects from WHEY withdrew due to personal reasons (n = 1) and medical conditions (n = 1). Thus, 16 subjects completed the trial and were analyzed (n = 9 and n = 7 in WHEY+CR and WHEY, respectively). A flowchart of participants is shown in Figure 1. Also followed by the main characteristics of subjects are expressed in Table 1.

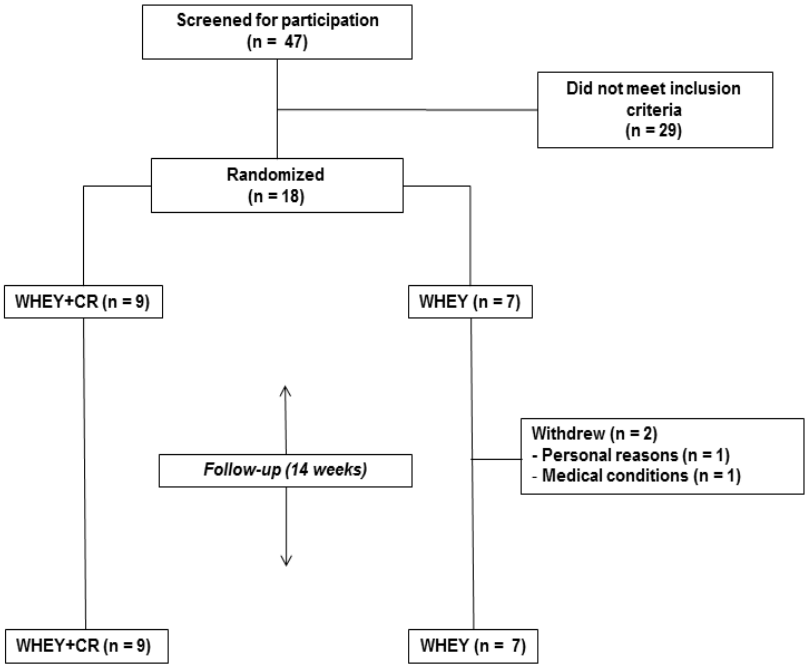

Figure 1 Flowchart of subjects included in the study (n=18) and subjects who withdrew (n=2) during the study.

Table 1. *Main characteristics of subjects*

	WHEY+CR	WHEY+P
Age (years)	70.33	69.43
Height (m)	1.67	1.67
Age (years)	70.33	69.43
Height (m)	1.67	1.67
Weight (kg)	86.31	86.56
BMI (kg/m2)	30.92	30.53
Total BMD (Z-score)	1.2	1.7
Diseases/morbidities, n (%)		
Hypertension	6	5
Type-2 diabetes	1	
Obesity	4	3
Depression	2	2
Osteoarthritis	4	3
Osteoporosis		1
CSHA clinical frailty scale, n (%)		
III	2	4
IV	5	2
V	2	1

Participants reported their adherence to the dietary interventions, exercise program and food intake: All participants reported, in food diaries, that they were complying with the supplement regimes. The adherence to the exercise program was comparable between the trained groups. Total energy and macronutrient intake was not significantly different between groups ($p > 0.05$). (WHEY+CR: 1125.50 ± 417; WHEY: 1107.44 ± 281) (Table 2).

Table 2. *Food intake variables before and after supplementation with creatine and protein (CR+WHEY or whey supplementation alone (WHEY) in older subjects undertaking resistance training.*

Variable	WHEY+CR		WHEY	
	Pre	Post	Pre	Post
Energy Intake (kcal)	1127.32	1125.50	1112.18	1107.44
Carbohydrate (g)	112.10	97.6	124.21	118.78
Carbohydrate (%)	39.78%	34.69%	44.67%	42.90%
Lipid (g)	45.53	43.80	31.98	32.22
Lipid (%)	36.35%	35.02%	25.88%	26.18%
Protein (g)	67.80	79.20	60.27	62.78
Protein (%)	24.05%	28.15%	21.68%	22.67%

Muscle function

Figure 2 shows muscle function data. We found a main time-effect in handgrip (WHEY+CR = 26.65 ± 31.29; WHEY = 13.84 ± 14.93 Kg; p = 0.0005), timed-up-and-go (WHEY+CR = -11.20 ± 9.37; WHEY = -17.76 ± 21.74; p = 0.006 s), and timed-stands-test (WHEY+CR = 47.50 ± 35.54; WHEY = 46.87 ± 24.23 reps; p = 0.0001), suggesting that WHEY+CR and WHEY were similarly effective in improving these variables.

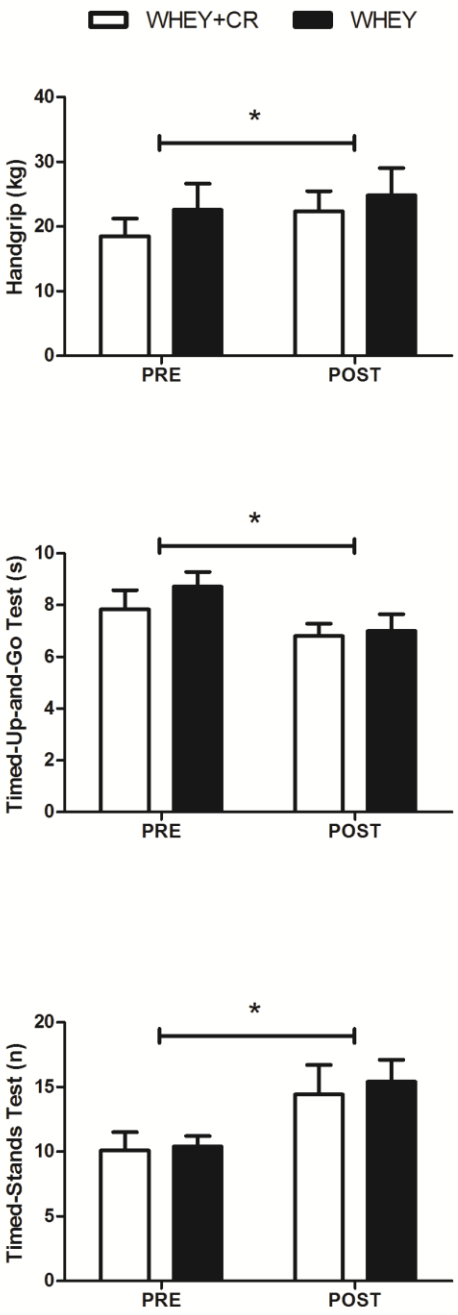

Figure 2. Results from the handgrip, timed up-and-go and the timed stand and sit test showing similar improvements between both groups.

Body composition

Figure 3 shows body composition data. Neither within- nor between-group differences were detected in any of the body composition variables (all $p > 0.05$). Also, relative changes were comparable across groups in all parameters (Free fat mass: WHEY+CR = 1.72 ± 5.07 and WHEY = 1.97 ± 2.36%); Fat mass: WHEY+CR = -2.66 ± 5.07 and WHEY = 0.59 ± 6.97%; Bone mineral content: WHEY+CR = -2.125 ± 5.82 and WHEY = -0.27 ± 8.23%; Total body bone mineral density: WHEY+CR = 0.08 ± 3.00 and WHEY = 1.97 ± 2.81%; Spine bone mineral density: WHEY+CR = -0.23 ± 5.74 and WHEY = -2.85 ± 6.44%; Femur bone mineral density: WHEY+CR = -0.64 ± 0.85 and WHEY = 0.56 ± 3.43%).

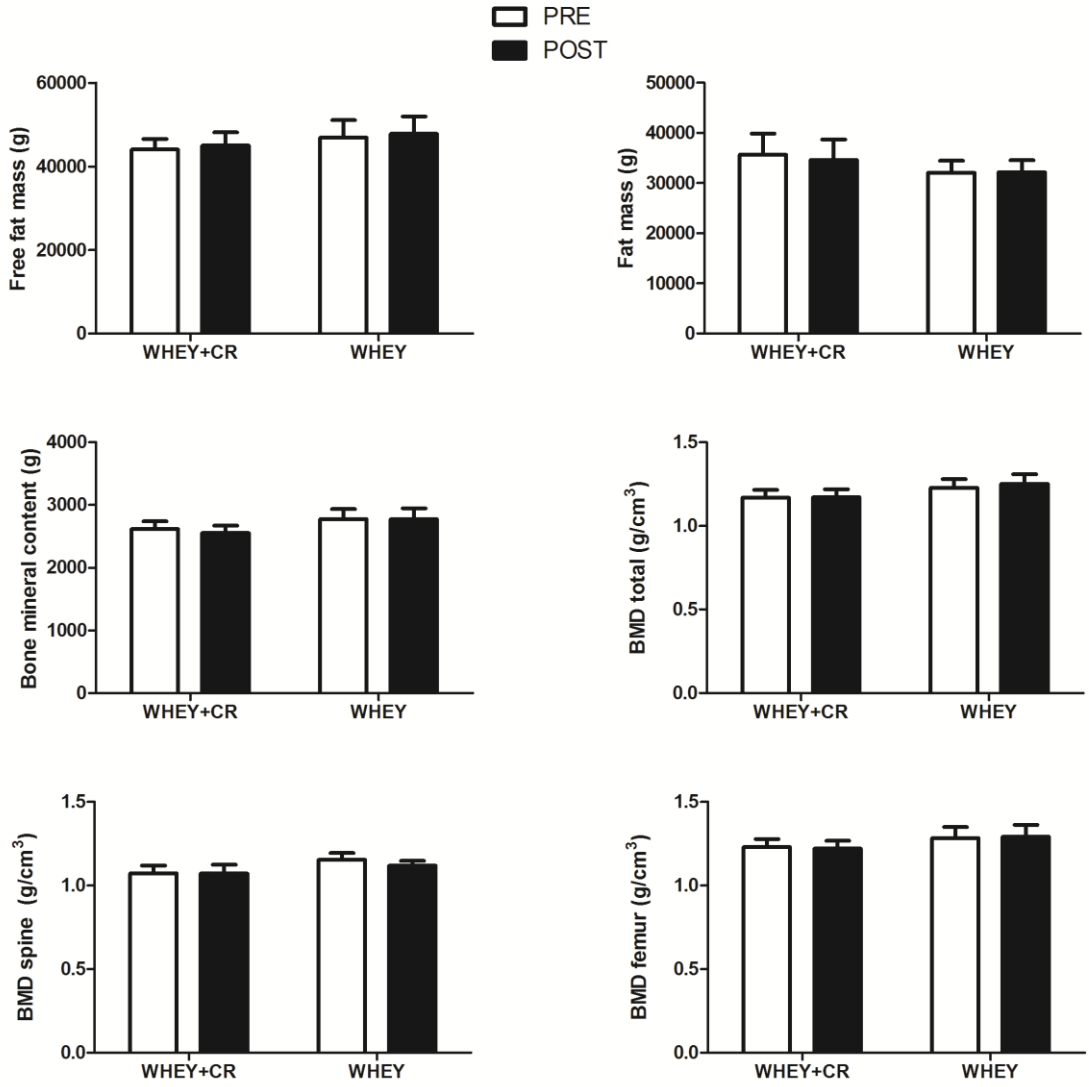

Figure 3. No significant differences between groups ($p>0.05$).

Adverse effects

There were no self-reported side effects throughout the study, except for one single episode of mild upset stomach reported by a subject from WHEY, which did not cause the subject exclusion from the study. Clinical examinations did not reveal adverse events potentially associated with creatine/protein supplementation or resistance training. Moreover, blood parameters (i.e., urea, sodium, potassium, bilirubin, alkaline phosphatase, gamma GT, alanine aminotransferase, aspartate aminotransferase, glucose, cholesterol, HDL, LDL, triglycerides, and creatine kinase) remained within normal range and results and did not change across time (all $p > 0.05$). The only exception was creatinine levels being slightly elevated in the WHEY group pre and post (Table 3).

Table 3. *Blood variables before and after supplementation with creatine and protein (CR+WHEY) or whey supplementation alone (WHEY) in older subjects undertaking resistance training.*

	WHEY+CR	WHEY	Reference Values
Urea			
Pre	5.10 ± 1.55	5.60 ± 1.90	3.2 - 7.7
Post	6.64 ± 1.75	6.63 ± 2.09	
Creatinine			
Pre	79.67 ± 17.52	98.57 ± 27.36	45 - 90
Post	81.56 ± 20.53	98.43 ± 30.37	
Sodium			
Pre	141.78 ± 2.22	141.00 ± 0.82	135 - 145
Post	141.11 ± 2.57	140.29 ± 0.76	
Potassium			
Pre	4.27 ± 0.53	4.47 ± 0.46	3.5 - 5.2
Post	4.61 ± 0.92	4.40 ± 0.22	
Bilirubin			
Pre	10.22 ± 3.96	11.07 ± 5.52	2 - 20
Post	9.57 ± 3.74	9.00 ± 2.58	
ALP			
Pre	83.11 ± 17.09	83.71 ± 51.59	

Post	93.67 ± 21.42	79.00 ± 31.80	30 - 150
GGT			
Pre	58.11 ± 76.95	58.56 ± 62.69	10 - 35
Post	64.86 ± 86.01	37.71 ± 22.43	
ALT			
Pre	24.22 ± 10.27	25.89 ± 12.73	0 - 30
Post	27.71 ± 18.91	28.71 ± 13.46	
AST			
Pre	30.11 ± 22.27	27.00 ± 13.56	10 - 50
Post	26.00 ± 12.60	26.71 ± 10.19	
Glucose			
Pre	5.02 ± 0.70	5.11 ± 1.12	3.5 - 6.0
Post	5.23 ± 0.66	5.59 ± 1.55	
Cholesterol			
Pre	5,06 ± 0.72	4.67 ± 1.23	< 5.0
Post	4.89 ± 0.61	4.61 ± 0.96	
HDL			
Pre	1.58 ± 0.37	1.45 ± 0.52	< 3.4
Post	1.60 ± 0.41	1.40 ± 0.53	
LDL			
Pre	2.93 ± 0.64	3.04 ± 0.96	> 1.0
Post	2.77 ± 0.66	3.00 ± 1.02	
TAG			
Pre	1.21 ± 0.40	1.46 ± 0.50	< 2.0
Post	1.12 ± 0.47	1.76 ± 0.81	
CK			
Pre	93.67 ± 39.00	109.43 ± 32.81	30 - 180
Post	111.22 ± 50.28	113.00 ± 47.31	

Further exploratory analysis

Table 4 shows individual data for muscle function and body composition data at Pre and Post as well as absolute change values (i.e., delta), ES, and specific p values for between-group comparisons. Muscle function parameters (i.e., handgrip, timed-stands test, and timed-up-and-go test) were beneficially affected in both groups alike, with medium to large ES. The large majority of the subjects experienced improvements in all muscle function tests (considering any improvement above zero). In addition, all of the subjects showed improvements in at least two of the three tests, regardless of their treatments.

In contrast, our analyses revealed that body composition variables (i.e., fat mass, free fat mass and bone mass) were neither clinically nor statically changed on average in both groups, despite the high individual variability observed.

Collectively, individual analysis, ES and delta changes along with inferential statistics suggested that the interventions (i.e., creatine plus whey protein vs. whey protein alone) were not significantly different from each other for any variable. The high individual variability found in this study seems to have occurred in both groups alike.

Table 4. *Muscle function and body composition data at Pre and Post for WHEY+C and WHEY groups. Effect size (ES) improvemnts were seen in muscle function tests regardless of treatment. Body composition changes were not statistically significant.*

Subject	**Handgrip (Kg)**		**TUG (s)**		**TST (reps)**		**FFM (Kg)**		**FM (Kg)**		**BMC (g)**		**BMD total (g)**		**BMD spine (g)**		**BMD femur (g)**	
WHEY+CR	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post
I	25.3	31.3	5.9	5.6	12	23	55.1	56.3	22.4	22.7	2865	2474	1.2	1.2	1.1	1.1	1.3	1.3
II	28.0	28.7	5.3	5.2	14	16	46.5	45.0	38.8	34.6	2762	2798	1.2	1.2	1.2	1.2	1.2	1.2
III	13.3	13.3	10.5	7.4	9	12	35.4	36.7	17.8	17.7	1887	1795	0.9	0.9	0.8	0.8	1.0	0.9
IV	9.7	10.3	8.2	7.6	10	11	45.5	44.7	20.8	20.4	2422	2517	1.1	1.1	0.9	0.1	1.2	1.2
V	16.3	19.0	6.1	5.7	13	21	40.0	42.1	34.1	32.2	2608	2532	1.1	1.1	1.0	0.9	1.1	1.1
VI	27.0	30.7	5.5	5.3	12	21	38.2	37.5	42.3	44.2	2633	2760	1.3	1.3	1.2	1.2	1.3	1.3
VII	4.7	9.3	10.2	9.0	9	12	37.3	37.0	45.7	45.8	2568	2606	1.3	1.3	1.3	1.3	1.3	1.3
VIII	21.3	31.7	10.5	8.5	0	1	56.6	64.0	48.9	44.0	3296	3078	1.3	1.4	1.1	1.1	1.5	1.4
IX	21.0	27.0	8.3	6.8	12	13	41.8	41.6	50.6	50.0	2502	2438	1.1	1.1	1.1	1.1	1.2	1.2
Mean (SD)	18.5(8.1)	22.4(9.3)	7.8(2.2)	8.7(1.5)	10(4)	14(7)	44.1(7.6)	4.5(9.4)	35.7(12.7)	32.1(6.3)	2616(376)	2779(413)	1.2(0.1)	1.2(0.4)	1.1(0.1)	1.1(0.2)	1.2(0.1)	1.2(0.1)

Delta Change	3.8(3.3)		-1.0(1.0)		4(4)		0.9(2.7)		-1.1(2.2)		-60(163)		0.0(0.0)		0.0(0.1)		0.0(0.0)	
ES	0.48		-0.47		1.04		0.12		-0.09		-0.02		0.01		-0.01		-0.06	
WHEY																		
I	30.7	32.0	7.5	7.2	11	19	52.1	54.0	32.5	32.2	3334	3234	1.3	1.3	1.3	1.1	1.4	1.3
II	7.3	8.3	8.9	6.6	12	18	35.5	36.9	28.3	25.4	2512	2437	1.2	1.1	1.2	1.0	1.1	1.1
III	32.3	34.0	6.8	6.8	12	14	55.0	54.0	29.2	30.0	3003	3050	1.2	1.2	1.1	1.2	1.3	1.3
IV	22.0	24.0	9.3	10.6	8	9	57.9	57.8	34.7	34.8	2923	3381	1.4	1.4	1.2	1.2	1.5	1.5
V	34.0	38.7	11.3	5.4	13	22	56.7	59.3	37.9	36.1	2851	2709	1.3	1.4	1.1	1.1	1.5	1.5
VI	22.3	23.3	9.6	6.7	8	13	39.1	39.8	40.4	42.5	2803	2508	1.2	1.3	1.3	1.2	1.2	1.3
VII	9.3	13.7	7.5	5.7	9	13	32.2	32.9	21.8	24.3	2026	2074	1.0	1.0	1.0	1.0	1.0	1.0
Mean (SD)	22.6(10.8)	24.9(11.0)	8.7(1.5)	7.0(1.7)	10(2)	15(4)	46.9(10.9)	47.7(10.9)	32.1(6.3)	32.1(6.3)	2779(413)	2770(472)	1.2(0.1)	1.2(0.1)	1.1(01)	1.1(0.1)	1.3(0.2)	1.3(0.2)
Delta change	2.3(1.6)		-1.7(2.3)		5(3)		0.9(1.2)		0.9(1.9)		-8(237)		0.0(0.0)		0.0(0.1)		0.0(0.0)	
ES	0.21		-1.12		2.42		0.08		0.01		-0.02		0.10		-0.35		0.05	
WHEY+CR *vs.* WHEY(P)	0.27		0.44		0.71		0.96		0.29		0.61		0.23		0.33		0.34	

Chapter 5: Conclusions

This study showed that co-supplementation with creatine and whey protein was well-tolerable and free of important adverse effects in older subjects with frailty. In addition, creatine supplementation did not enhance the effects of resistance training combined with whey protein on body composition or muscle function parameters in this population.

Frailty in the aging represents one of the most significant problems the public health that gives rise to muscle dysfunction and body composition disturbances (e.g., loss of bone and muscle mass), thereby resulting in vulnerability (Rockwood, Song, MacKnight, Bergman, Hogan, & McDowell, 2005) (Rockwood, Fraility and its definition: a worthy challenge, 2005). To date, the first-choice interventions to manage this syndrome consist of resistance training and a few dietary interventions (Breen & Phillips, 2011) (Wolfe, 2013). In fact, a sufficient body of literature has evidenced the beneficial role of resistance exercise in counteracting muscle dysfunction and loss of free fat mass in older individuals. Furthermore, growing evidence has indicated that supplementary high-quality proteins can also be supportive of muscle anabolism in the elderly, particularly when combined with resistance training (Cermak, Res, de Groot, Saris, & van Loon, 2012).

More recently, a few studies have shown that creatine supplementation along with resistance training can improve muscle function, bone, and muscle mass in older subjects with frailty (Gualano, et al., 2014) (Chilibeck, Candow, Landeryou, Kaviani, & Paus-Jenssen, 2014), although confirmatory studies remain necessary.

Co-supplementation with creatine and protein might be expected to promote synergistic effects based on their differential mechanisms of actions. Protein supplementation has been shown to positively modulate muscle protein balance, stimulating protein synthesis in elderly subjects (Yang, Burd, Churchwood-Venne, Tarnopolsky, & Phillips, 2012). Interestingly, it has been suggested that proper doses and types of protein supplementation (e.g., 30-40 g per meal or 1.2 g/Kg/day of isolate whey or whole milk-proteins) can partially offset the so-called “anabolic resistance”, a condition characterized by a blunted-response to anabolic stimulus (i.e., exercise and protein intake) in the elderly (Breen & Phillips, 2011) (Wall,

Cermak, & van Loon , 2014) (Bauer, et al., 2013) (Wolfe, 2013). Creatine supplementation, in turn, appears not to directly affect muscle protein balance, at least in young subjects (Louis, et al., Creatine supplementation has no effect on human muscle protein turnover at rest in the postabsorptive or fed states., 2003) (Louis , Poortmans, Francaux, Berre, Boisseau, & Brassine, No effect of creatine supplementation on human myofibrillar and sarcoplasmic protein synthesis after resistance exercise., 2003); the positive effect of this amine upon muscle mass seems to be a result of its bioenergetics role in enhancing ATP resynthesis, which could lead to increased training volume and, hence, muscle mass gains (Gualano, et al., 2014) (Rawson & Venezia, 2011) (Gualano, Artioli, Poortmans, & Lancha Jnr, 2009).

Thus, we hypothesized that creatine could add to whey protein in improving muscle function and body composition parameters in older subjects undergoing a resistance training program. Our findings, however, failed to support any beneficial effect of creatine.

These findings seem to be in agreement with those of previous studies showing no synergistic effects of creatine and protein in elderly subjects engaged in resistance training (Eliot, Knehans, Bemben, Witten, & Bemben, 2008), (Villanueva, He, & Schroeder, 2014). It is possible to speculate that the effects of creatine supplementation on muscle function and body composition parameters were too small to be detected by small-scale trials. In addition, there are subjects unresponsive to creatine supplementation, which might have further precluded us to find positive effects associated with this dietary supplement. In fact, previous studies investigating the possible additive effects of creatine and protein supplements have small samples too, which may partially explain the lack of positive outcomes.

Certainly, additional studies with larger sample sizes are necessary to verify as to whether creatine supplementation adds to protein supplementation in promoting positive adaptations in older subjects undergoing resistance training. Furthermore studies should determine muscle creatine/phosphoryilcreatine in order to provide a better link between changes in functional and morphological variables and possible increases in muscle high-energy phosphagens.

Another interesting data from this study was the beneficial effect of resistance training and whey protein supplementation (regardless of creatine) in increasing muscle function, but no free fat mass, in older subjects with frailty. There are meta-

analytic data to suggest that protein supplementation additively to prolonged resistance training can augment both free fat mass and muscle function (Cermak, Res, de Groot, Saris, & van Loon, 2012). The lack of increases in free fat mass in this study may be a result of a too short follow-up period (i.e., 14 weeks in the current study *vs.* 24 weeks in the meta-analysis). Importantly, it is worth noting that our experimental design did not allow us to distinguish the effects of resistance training from those of protein supplementation, nor the effects of training and supplementation from those of no intervention at all. Further studies with more comprehensive designs allowing full comparisons between creatine and whey protein supplementation, combined or alone, and placebo will be elucidative.

It is important to highlight that co-supplementation with creatine and protein is safe and well-tolerable among older subjects with frailty. All of them adhered to supplementation protocols. No adverse effects, except for a single episode of mild stomach upset, were reported. Blood parameters also revealed no abnormalities in muscle, kidneys or liver metabolism.

Finally, the compliance with the resistance training protocol was highly satisfactory, with no incidence of injuries or any other training-related adverse events. Taking together, these observations allow concluding that our intervention is feasible and safe and, as such, merits further investigation.

Important lessons were learned from this exploratory study. Firstly, based on the ES found in this trial, larger sample sizes seem to be necessary to show any additive effect of creatine and protein. Secondly, longer-term follow-ups appear to be particularly important to show any beneficial effects of creatine and/or protein supplementation in resistance-trained elderly subjects. Thirdly, resistances training along with whey protein seem to be a tolerable, safe and effective strategy in improving muscle function in the elderly with frailty. The role of each one of the interventions applied in this trial requires further investigation. All of these gaps will be taken into consideration in a larger comprehensive study to confirm or refute these preliminary data.

In conclusion, co-supplementation with creatine and whey protein was tolerable and free of adverse events in older subjects with frailty undertaking resistance training. Moreover, creatine supplementation did not augment the adaptive effects of resistance training along with whey protein on body composition or muscle function in this population.

References

Bauer, J., Biolo, G., Cederholm, T., Cesari, M., Labarque, V., Taes, Y., & Verbessem, P. (2013). Evidence-based recommendtions for dietary protein intake in older people: A position paper from the PROT-AGE study group. *Journal of American Diretics Association, 14*(8), 542-59.

Bender, A., Auer, D. P., Merl, T., Reilmann, R., Saemann, P., Yassouridis, A., . . . Klopstock, T. (2005). Creatine supplementation lowers brain glutamate levels in Huntington's disease. *Journal of Nuerology, 252*(1), 36-41.

Breen, L., & Phillips, S. M. (2011). Skeletal Muscle protein metabolism in the elderly: Interventions to counteract the anabolic resistance of ageing. *Journal of Nutrition and Metabolism*, 8-68.

Cadore, E. L., & Izquierdo, M. (2013). How to simultanouesly optimize muscle strength, power, functional capacity and cardiovascular gains in elderly: an update. *AGE, 35*(6), 2329-44.

Candow, D. G., Little, J. P., Chilibeck, P. D., Abeysekara, S., Zello, G. A., Kazachkov, M., . . . Yu, P. H. (2008). Low-dose creatine combined with protein during resistance training in older men. *Medicine Science and Sports Exercise, 40*(9), 1645-52.

Cermak, N. M., Res, P. T., de Groot, L. C., Saris, W. H., & van Loon, L. J. (2012). Protein supplementation augments the adaptive response of skeletal muscle to resistance-type exercise training: a meta-analysis. *American Journal of Clinical Nutrition, 96*(6), 1454-64.

Chilibeck, P. D., Candow, D. G., Landeryou, T., Kaviani, M., & Paus-Jenssen, L. (2014). Effects of crfeatine and resistance trainingon bone health in postmenopausal women. *Medical Science Sports and Exercise*.

Cohen, J. (1988). Statistical power analysis for the behavioral sciences:. *Lawrence Erlbaum Associates*.

Deutz, N. E., Bauer, J. M., Barazzoni, R., Biolo, G., Boitie, Y., Bosy-Westphal, A., . . . Calder, P. C. (2014). Protein intake and exercise for optimal muscle function with aging: recommendations from the ESPEN Expert Group. *Clinical Nutrition, 33*(6), 929-36.

Eijnde, B. O., van Leemputte, M., Goris, M., Labarque, V., Taes, Y., Verbessem, P., . . . Hespel, P. (2003). Effects of creatine supplementation and exercise training on fitness in men 55-75 years old. *Journal of Applied Physiology, 95*(2), 818-28.

Eliot, K. A., Knehans, A. W., Bemben, D. A., Witten, M. S., & Bemben, M. G. (2008). The effects of creatine and whey protein supplementation on body composition in men aged 48 to 72 years during resistance training. *Journal of Nutrition Health and Ageing, 12*(3), 208-12.

Gualano, B., Artioli, G. G., Poortmans, J. R., & Lancha Jnr, A. H. (2009). Exploring the therapeutic role of creatine supplementation. *Amino Acids*.

Gualano, B., Macedo, M., Alves, C., Roschel, H., Benatti, F., Takayama, L., . . . Pereira, R. M. (2014). Creatine supplementation and resistance training in vulnerable older women. *Experimental Gerontology, 53*, 7-15.

Gualano, B., Roschel, H., Lancha-Jr, A., Brightbill, C., & Rawson, E. (2012). Interventions to counteract the anabolic resistance of ageing. *Nutrition and Metabolism, 2*(43), 519-29.

Hunter, G. R., McCarthy, J. P., & Bamman, M. M. (2004). Effects of resistance training on older adults. *Sports Medicine, 345*, 329-348.

Lopes, J. B., Danilevicius, C. F., Takayama, L., Caparbo, V. F., Menezes, P. R., Scazufca, M., . . . Pereira, R. M. (2011). Prevalence and risk factors of radiographic vertebral fracture in Brazilian community-dwelling elderly. *Osteoporosis International, 22*(2), 711-9.

Louis , M., Poortmans, J. R., Francaux, M., Berre, J., Boisseau, N., & Brassine, E. (2003). No effect of creatine supplementation on human myofibrillar and sarcoplasmic protein synthesis after resistance exercise. *American Journal Endocrinol Metabolism., 285*(5), 1089-94.

Louis, M., Poortmans, J. R., Francaux, M., Hultman, E., Berre, J., Boisseau, N., . . . Rennie, M. J. (2003). Creatine supplementation has no effect on human muscle protein turnover at rest in the postabsorptive or fed states. *American Journal of Physiology Endocrinol Metabolism, 284*(4), 1349-62.

Mitchell, K. W., Williams , J., Atherton, P., Larvin, M., Lund, J., & Narici, M. (2012). Sacopenia, Dynapenia and the impact of advancing age on human skeletal muscle size and strength; a quantitive review. *Fronteirs in Physiology, 260*(3). doi:10.3389/fphys.2012.00260.

Neves, M. J., Gualano, B., Roschel , H., Fuller, R., Benatti, F. B., Pinto, A. L., . . . Bonfa, E. (2011). Beneficial effect of creatine supplementation on knee osteoarthritis. *Medicine and Science in Sport and Exercise, 43*(8), 1538-43.

Newcomer, K. L., Krug, H. E., & Mahowald, M. L. (1993). Validity and reliability of the timed-stands test for patients with rheumatoid arthritis and other chronic diseases. *Journal of Rheumatol, 20*(1), 21-7.

Nuzzo, R. (2014). Scientific method: statistical errors. *506*(7487), 150-2.

Podisiadlo, D., & Richardson, S. (1991). The timed "Up & Go": a test of basic functional mobility for frail elderly persons. *Journal of American Geriatric Society, 20*(1), 142-8.

Rawson, E. S., & Venezia, A. C. (2011). Use of creatine in elderly and evidence for effects on cognitive function in young and old. *Amino Acids, 40*(5), 1346-62.

Rockwood, K. (2005). Fraility and its definition: a worthy challenge. *Journal of American Geriatric Association, 53*(6), 1069-70.

Rockwood, K., Song, X., MacKnight, C., Bergman, H., Hogan, D. B., & McDowell, I. (2005). A global clinical measure of fitness and frailty in elderly people. *Canadian Medical Association Journal, 173*(5), 489-95.

Shepherd, J. A., Lu, Y., Wilson, K., Fuerst, T., Genant, H., Hangartner, T. N., . . . Leib, E. S. (2006). Cross-calibration and minimum precision standards of dual-energy X-ray absorptiometry: the 2005 ISCD Official Positions. *Journal of Clinical Densitometry, 9*(1), 31-6.

Statistics New Zealand. (2000, January/Febuarary .). *National population estimates.* Retrieved from Statistics New Zealand: http://www.stats.govt.nz/browse_for_stats/people_and_communities/older_people/pop-ageing-in-nz.aspx

Tarnopolsky, M., Zimmer, A., Paikin, J., Safdar, A., Aboud, A., Pearce, E., . . . Doherty, T. (2007). Creatine monohydrate and conjugated linoleic acid improve strength and body compisition following resistance exercise in older adults. *PLOS One, 2*(10).

Tieland, M., Dirks, M., van der Zwaluw, N., Verdijk, L., van de Rest, O., de Groot, L., & van Der Loon, L. J. (2012). Protein supplementation increses muscle mass gain during prolonged resistance-type exercise training in frail elderly people: a randomized, double blind, placebo-controlled trial. *Journal of American Medical Directors Association, 13*(8), 713-9.

Villanueva, M. G., He, J., & Schroeder, E. T. (2014). Periodized resistance training with and without supplementation improves body compesition and performance in older men. *European Journal Applied Physiology, 114*(5), 891-905.

Wall, B., Cermak, N., & van Loon , L. (2014). Dietary protein considerations to support active ageing. *Sports Medicine, 2*(44), 185-94.

Wallimann, T., Wyss, M., Brdiczka, D., Nicolay, K., & Eppenberger, H. M. (1992). Intracellular compartmentation, structure and function of kinase isoenzymes in tissues with high and fluctuating energy demands: the phosphocreatine circuit for cellular energy homeostatis. *Biochemical Journal, 281*(1), 21-40.

Wolfe, R. R. (2013). Perspective: Optimalprotien intake in elderly. *Journal of American Dir Association, 14*(1), 65-66.

Yang, Y., Burd, N. A., Churchwood-Venne, T. A., Tarnopolsky, M. A., & Phillips, S. M. (2012). Myofibrillar protein synthesis following ingestion of soy protein isolate at rest and after resistance exercise in elderly men. *Nutrition Metlab, 9*(1), 57.

Appendices

Appendix A
Timed up & go protocol

Timed Up and Go (TUG) Test[1,2]

1. Equipment: arm chair, tape measure, tape, stop watch.

2. Begin the test with the subject sitting correctly in a chair with arms, the subject's back should resting on the back of the chair. The chair should be stableand positioned such that it will not move when the subject moves from sitting to standing.

3. Place a piece of tape or other marker on the floor 3 meters away from the chair so that it is easily seen by the subject.

4. Instructions : "On the word *GO* you will stand up, walk to the line on the floor, turn around and walk back to the chair and sit down. Walk at your regular pace.

5. Start timing on the word "*GO*" and stop timing when the subject is seated again correctly in the chair with their back resting on the back of the chair.

6. The subject wears their regular footwear, may use any gait aid that they normally use during ambulation, but may not be assisted by another person. There is no time limit. They may stop and rest (but not sit down) if they need to.

7. Normal healthy elderly usually complete the task in ten seconds or less. Very frail or weak elderly with poor mobility may take 2 minutes or more.

8. The subject should be given a practice trial that is not timed before testing.

9. Results correlate with gait speed, balance, functional level,the ability to go out, and can follow change over time.

10. Interpretation
    - ≤ 10 seconds = normal
    - ≤ 20 seconds = good mobility, can go out alone, mobile without a gait aid.
    - < 30 seconds = problems, cannot go outside alone, requires a gait aid.

A score of more than or equal to fourteen seconds has been shown to indicate high risk of falls.

1. Podsiadlo D, Richardson S. *The Time "Up & Go": A Test of Basic Functional Mobility for Frail Elderly Persons.* Journal of the American Geriatrics Society 1991; 39(2): 142-148
2. Shumway - Cook A, Brauer S, Woollacott M. *Predicting the Probability for Falls in Community-Dwelling Older Adults Using the Timed Up & Go Test.* Physical Therapy 2000 Vol 80(9): 896-903.

Saskatoon Falls Prevention Consortium, Falls Screening and Referral Algorithm, TUG, Saskatoon Falls Prevention consortium, June, 2005

Appendix B

Timed Stand protocol

- **Purpose:** This test assesses leg strength and endurance.
- **Equipment required:** a straight back or folding chair without arm rests (seat 17 inches/44 cm high), stopwatch.
- **Procedure:** Place the chair against a wall, or otherwise stabilize it for safety. The subject sits in the middle of the seat, with their feet shoulder width apart, flat on the floor. The arms are to be crossed at the wrists and held close to the chest. From the sitting position, the subject stands completely up, then completely back down, and this is repeated for 30 seconds. Count the total number of complete chair stands (up and down equals one stand). If the subject has completed a full stand from the sitting position when the time is elapsed, the final stand is counted in the total.
- **Scoring:** the score is the number of completed chair stands in 30 seconds.

Appendix C Resistance Training Program

Resistance Program Example				
Exercise	**Description**	**Load**	**Reps**	**Sets**
Warm-up	Stationary cycle – Seat will be adjusted for subject height. Subject is required to remain seated with both hands on hand grips.	5 Minutes No load – Subjects are required pedal at a moderate constant speed. The subject must remain seated and both hands on the handlebars.		
Step Ups	Stand close to a handrail or stable support. Stand in front of a step and starting with your right foot step up (heel first) your whole foot must be on the step. Lift your left leg up to meet the right foot pause and lower left leg.	No load Performed using graduated step using lowest setting. Height will be adjusted at 4, 8 and 12 weeks.	8	3
Seated shoulder press	Sit in a chair keeping back straight, feet shoulder width apart. Raise the hand and arm above head and lower to shoulder height in a controlled manner. Alternatively this can be performed on shoulder press machine if subject lift weight is more than 5kgs.	50% 1RM increased at 4, 8 and 12 weeks.	8	3
¼ - ½ Squat	Stand feet shoulder width in front of an armless chair. Extend arms and lean forward slightly at the hips. Lower yourself to the chair controlled and ensuring knees do not go past toes. Pause for 2 seconds and rise in the same controlled manner.	No load Height of chair will be determined by subject ability.	8	3
Seated row	Using machine back straight grip handles and using arm and back muscles pull towards you slowly. Go back to start position in a controlled manner. Alternatively seated row with light weights.	Load determined by the individual.	8	3
Leg Press	Using the machine adjusted for subject. Ensure leg is straight throughout the movement. The knee should track in the direction of the foot.	50% 1RM increased at 4, 8 and 12 weeks.	8	3

Biceps curl	This exercise can be performed either sitting or standing. Using hand weights one in each hand. Feet shoulder width apart, palms facing thighs. Slowly lift weights toward shoulders. Upper arms and elbows remain close to the body. Pause before lowering the weights in a controlled manner.	50% 1RM increased at 4, 8 and 12 weeks.	8	3
Leg Extension	Using either a chair or weight machine, sit all the way back in the seat. Feet should barely reach the ground. With a flexed foot slowly extend one leg until knee is straight (do not lock the knee). Pause and lower in a controlled manner.	50% 1RM increased at 4, 8 and 12 weeks.	8	3
Triceps extension	Sit in a chair keeping back straight, feet shoulder width apart. Raise the hand and arm towards the ceiling and bend at the elbow. This is the starting position. Bending at the elbow raise arm up towards ceiling and then lower in a controlled manner.	Load determined by individual	8	3
Calf raise	Using a chair or bench for support stand shoulder width apart. To the count of four raise up onto toes, hold for four seconds then lower	No load	8	3
Sit-ups	Lying on a comfortable surface, legs bent arms across chest or placed behind neck. Raise shoulders off the ground slightly while contracting abdominals. Alternatively start from upright position and lower to the floor slowly.	No Load	10	3
Cool down	Quadriceps stretch Hamstring stretch Chest and arm stretch Upper backs stretch Shoulder stretch			

Appendix D
Participant Forms

Wintec Research information

Project Information Sheet

Project Title: The effect of creatine combined with protein supplementation on lean muscle mass and muscle function in elderly subjects undergoing resistance training.

Researcher: Jodie Collins **Supervisors:** Dr Bruno Gualano and Dr Glynis Longhurst

Project Purpose: The purpose of this study is to assess the effectiveness of creatine + protein supplementation combined with resistance training on lean muscle mass, muscle strength and muscle function in frail elderly subjects.

Eligibility: 60 years or older, not exercising, No uncontrolled medical conditions and no chronic conditions such as cancer or neurological diseases

Involvement: You will be required to attend two sessions per week for 16 Weeks starting the first week of August 2014 and ending November 2014. Training sessions will be at least 48 hours apart. Scheduled times will be given based on your availability. Sessions will be held at WINTEC in the Sport & Exercise Science building.

You will be require to fill in the consent forms and get GP medical clearance. You will also be required to have 2 blood tests and two DEXA (Bone Density) Scans before and after your program begins.

A familiarisation of the exercise protocol will take place 1 week before testing begins. This session will allow you to become familiar with exercise equipment and technique protocols. You will be given information on monitoring equipment and there will be opportunity to ask questions.

On the day of testing the session will last approximately 30mins. You will be required to perform some of the program exercises. With these exercises I would like to determine your 10 repetition max (RM), this will enable the 16 week resistance program to be set at your correct level. To determine your 10 RM you will be required to lift a given weight 10 times, weights will be adjusted if load is too heavy or too light. You will have 5 minutes to rest between sets and exercises. If at any stage you experience discomfort please stop and advise the researcher.

Following testing you will be allocated into either group 1 or group 2 you will be given the allocated supplement in which you will be required to start taking daily from August. Supplement whey protein powder should be mixed with milk or water along with two chewable creatine tablets, taken twice a day with breakfast and dinner. Giving a daily total of 20g Protein and 4g creatine.

Please allow at least 60mins for each training session, all sessions will be recorded and monitored by the researcher. Heart rate monitors must be worn for safety and will be monitored, if you have any discomfort during the sessions please stop and advice the supervisor. Showering facilities are available.

If at any stage you are unable to make a training session, a catch up session will need to be completed prior to the next session occurring. Please let me know, using the contact details at the bottom of this sheet, so we can sort out a time and place for this to occur.

Please wear gym shoes and comfortable clothing to exercise in.

You are also required to fill in a food diary each day and will be handed in weekly. The food diary sheets will be given to you each week.

Testing Length: A Pre & Post Test will be done each approx. 30min

Training Length: You will be required to attend 2 resistance trainings per week for 16 weeks.

Data Collection: Data collection will take place at Wintec Rotokauri (Avalon) campus in the School of Sport and Exercise Science.

Information: All information and data on participants will be kept private and secure unless waived by the participant (written consent).

Participation: It is not compulsory to take part in this investigation. Please take the opportunity to talk through your potential involvement with family, friend and/or spouse. You may bring a support person with you to the testing session.

Withdrawal: Participants may withdraw from the study at any stage of any reason without consequence. If a participant desires to withdraw they may converse with the researcher directly or use any means of contact (email, text or call, shown below).

Participant Acknowledgment/Privacy: Participants will not be recognized by name in text or analysis. Collected data will be coded to ensure privacy from persons outside of the research coordinators.

Research Results: Feedback and results will be made available to any participant that requests it.

Further Inquiries: For any further inquiries or questions about the investigation please contact using the below details.

Name: Jodie Collins

Contact Details: Phone: (07) 8297800 (021) 709504

Email: jodiec@clear.net.nz

Participant Consent Form
(one copy to be retained by the Research Participant and one copy to be retained by Researcher)

I …… (participant's name) consent to being a participant in the above named research project, and I attest to the following:

1. I have been fully informed of the purpose and aims of this project.

2. I understand the nature of my participation and have full Doctor's medical clearance.

3. I understand the benefits that may be derived from this project.

4. I understand that I may review my contributions at any time without penalty.

5. I understand that I will be treated respectfully, fairly and honestly by the researcher/s, and I agree to treat the other participants in the same way.

6. I understand that I will be offered the opportunity to debrief during, or at the conclusion of this project.

7. I have been informed of any potential harmful consequences to me by taking part in this project.

8. I understand that I may withdraw from the project at any time (without any penalties)

9. I understand that personal and testing information gathered from me will be treated with confidentiality, except where I consent to waive that confidentiality.

10. Due to the training group environment required for this project I agree to maintain the anonymity and privacy of other participants, and the confidentiality of the information they contribute.

11. I understand information from this research may be used for academic publication purposes, knowing I will not be able to be identified

Participant ……
Date……

Principal Researcher Jodie Maree Collins Date…08/07/2014

Health Questionnaire

Name:	
Address:	
Contact Phone Number(s):	
Email Address:	
Date Of Birth:	
Emergency Contact Person:	**Address:** **Phone:**
Doctor:	**Medical Centre Information:**

Please answer the following questions with full disclosure

Have you ever smoked – cigarettes, pipes or cigars? **If YES do you currently smoke?** **On average how many would you smoke a day?**	**YES / NO** **YES / NO**
Are you on medication? **If Yes please list medications and conditions below:**	**YES / NO**
Do you have Diabetes?	**YES / NO**
Do you have High Blood Pressure?	**YES / NO**
Do you have High Cholesterol?	**YES / NO**
Do you have Arthritis or Osteoarthritis?	**YES / NO**

Do you suffer from any other joint, muscle or bone problems? **If YES please explain:**	**YES / NO**
Have you ever suffered from heart problems? **(This includes angina, heart attack, genetic defects etc...)** **If YES please explain:**	**YES / NO**
Do you participate in regular physical activity? **(Regular = at least 3 times a week for 30min or more)** **If YES please explain:**	**YES / NO**
Have you ever experienced any adverse effects while doing exercise? **(Shortness of breath, tight chest, dizziness etc...)** **If YES please explain:**	**YES / NO**
Have you had surgery in the last year? **If YES please explain:**	**YES / NO**
Do you or will you get medical clearance to exercise if you are included in this research study?	**YES / NO**
Is there anything you want to add?	**YES / NO**

Signature: **Date:**