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**The effect of bovine colostrum supplementation in older adults during resistance training**

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**Running Head:** Colostrum and resistance training

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24

25 **Abstract**

26 Bovine colostrum is the first milk secreted by cows after parturition and has high levels  
27 of protein, immunoglobulins, and various growth factors. We determined the effects of 8 weeks  
28 of bovine colostrum supplementation versus whey protein during resistance training in older  
29 adults. Males (N = 15, 59.1±5.4y) and females (N=25, 59.0±6.7y) randomly received (double  
30 blind) 60g/d of colostrum or whey protein complex (containing 38g protein) while participating  
31 in a resistance training program (12 exercises, 3 sets of 8-12 reps, 3d/wk). Strength (bench press  
32 and leg press 1-RM), body composition (by dual energy X-ray absorptiometry), muscle thickness  
33 of the biceps and quadriceps (by ultrasound), cognitive function (by questionnaire), plasma  
34 insulin-like growth factor-1 (IGF-1) and C-reactive protein (CRP, as a marker of inflammation),  
35 and urinary N-telopeptides (Ntx, a marker of bone resorption) were determined before and after  
36 the intervention. Participants on colostrum increased leg press strength (24±29 kg; p<0.01) to a  
37 greater extent than participants on whey protein (8±16 kg) and had a greater reduction in Ntx  
38 compared to participants on whey protein (-15±40% vs. 10±42%; p<0.05). Bench press strength,  
39 muscle thickness, lean tissue mass, bone mineral content, and cognitive scores increased over  
40 time (p<0.05) with no difference between groups. There were no changes in IGF-1 or CRP.  
41 Colostrum supplementation during resistance training was beneficial for increasing leg press  
42 strength and reducing bone resorption in older adults. Both colostrum and whey protein groups  
43 improved upper body strength, muscle thickness, lean tissue mass, and cognitive function.

44 Clinicaltrials.gov identifier: NCT01792297

45

46 **Keywords:** muscle, bone, inflammation, cognitive function, IGF-1

47

## 48 **Introduction**

49           Skeletal muscle is lost after approximately the age of 50y, potentially leading to  
50 sarcopenia and loss of muscle strength and function (International Working Group on  
51 Sarcopenia, 2011; Roubenoff, 2003). Muscle loss may be related to a decrease in anabolic  
52 hormones and/or increased catabolism driven by inflammation (Roubenoff, 2003; Visser et al.,  
53 2002). Older muscle is more sensitive to damage via less effective anti-oxidant systems leading  
54 to an altered response of satellite cells in regeneration of damaged muscle (Degens, 2010;  
55 Thalacker-Mercer, Dell'Italia, Cui, Cross, & Bamman, 2010). This response is linked to  
56 differential expression of skeletal muscle specific genes, with up-regulation of transcripts related  
57 to stress, inflammation, and protein degradation, and down-regulation of some transcripts related  
58 to protein synthesis in old versus young muscle (Degens, 2010; Thalacker-Mercer et al., 2010).  
59 Chronic low-grade systematic inflammation is the main factor contributing to the attenuated  
60 hypertrophic response of older muscle to strength training (Degens, 2010) and plays an important  
61 role in the development of disability (Visser et al., 2002). Increased inflammation associated  
62 with aging diminishes the efficacy of insulin-like growth factor-1 (IGF-1), an anabolic hormone  
63 responsible for muscle hypertrophy and regeneration (Degens, 2010) and therefore inflammation  
64 is associated with lower muscle mass and strength in older adults (Visser et al., 2002). Insulin-  
65 like growth factor-1 is also important for development of brain and bone tissue and reduction in  
66 IGF-1 in older adults is associated with cognitive decline (Ceda et al., 2005), and lower bone  
67 mass (Ohlsson et al., 2011).

68           Bovine colostrum is, by definition, the first milk secreted by cows immediately following  
69 parturition (Larson, Heary, & Devery, 1980). Bovine colostrum contains essential amino acids  
70 and peptide components including whey and casein, and many bioactive components such as

71 lactoferrin, immunoglobulins, and various growth factors (Klagsbrun & Neumann, 1979;  
72 Korhonen, 1977; Larson et al., 1980). Insulin-like growth factor-1 (IGF-1) is the most abundant  
73 and well-characterized growth factor in bovine colostrum and is homologous to human IGF-1  
74 (Francis, Upton, Ballard, & McNeil, 1988; Marcotty, Frankenne, van Beeumen, Maghuin-  
75 Rogister, & Hennen, 1991). Bioactive components of colostrum are known to stimulate DNA  
76 synthesis, protein synthesis, and cellular growth in neonatal and newborn animals (Burrin et al.,  
77 1997; Francis et al., 1988) but it is unclear whether this anabolic effect applies to adult humans.

78         Bovine colostrum increases anti-inflammatory cytokines (Shing, Peake, Suzuki, Jenkins,  
79 & Coombes, 2007). Athletes have used bovine colostrum as a nutritional supplement during  
80 training to reduce upper respiratory tract infection, although no effects on either saliva or plasma  
81 immunoglobulin levels were found (Brinkworth & Buckley, 2003; Crooks, Cross, Wall, & Ali,  
82 2010); however, respiratory tract symptoms, often experienced during periods of heavy training,  
83 may actually result from inflammation rather than suppressed immune function (Bachert, van  
84 Kempen, Höpken, Holtappels, & Wagenmann, 2001). Exercise training with bovine colostrum  
85 supplementation is also beneficial for improving exercise performance (Buckley, Brinkworth, &  
86 Abbott, 2003; Hofman, Smeets, Verlaan, v.d. Lugt, & Verstappen, 2002) and increasing lean  
87 tissue mass (Antonio et al., 2001), but the effects on strength remain unclear (Shing, Hunter, &  
88 Stevenson, 2009). It is possible beneficial effects of bovine colostrum during exercise training  
89 may be from decreased inflammation.

90         The purpose of this study was to determine the effect of bovine colostrum  
91 supplementation during a resistance training program on inflammatory status, serum IGF-1  
92 levels, lean tissue mass, strength, cognitive function, and bone turnover in men and women 50y  
93 and older. It was hypothesized that bovine colostrum supplementation during resistance training

94 would prevent inflammation, increase IGF-1 levels, lean tissue mass, strength, and cognitive  
95 function, and reduce bone turnover.

96

## 97 **Methods**

### 98 *Participants*

99         Forty participants (15 males  $59.1 \pm 5.4$  y; 25 females,  $59.0 \pm 6.7$  y) were recruited via an  
100 advertisement in a local newspaper. The sample size was based on studies of young individuals  
101 where change in lean tissue mass with bovine colostrum supplementation was 1.5-2 kg compared  
102 to 0-1.2 kg with whey protein with a standard deviation for this change of 0.5 to 1.0 kg (Antonio  
103 et al., 2001; Kerksick et al., 2007), an alpha of 0.05 and power of 0.8. This sample size  
104 calculation indicated 10 participants per group (i.e. 20 in total) were required. The sample size  
105 was doubled because older individuals have greater variability in their physiological  
106 measurements (i.e. muscle mass and strength) compared to younger individuals (Candow &  
107 Chilibeck, 2005). The study was approved by The University of Saskatchewan's Research Ethics  
108 Board and participants gave informed consent for the study. Participants completed the Physical  
109 Activity Readiness Questionnaire (Thomas, Reading, & Shephard, 1992) prior to baseline testing  
110 to ensure there were no contra-indications to exercise participation.

### 111 *Intervention*

112         After completion of baseline testing (described below) participants were randomly  
113 assigned, by use of a computerized random number generator, to either bovine colostrum (N =  
114 12 females, 7 males) treatment or the control group (whey protein; N = 13 females, 8 males). The  
115 study was double blinded: researchers, participants, and all individuals conducting outcome  
116 assessments were unaware of group assignments. Both groups were provided a 4 kg container

117 and consumed 3 doses of 20g per day (60g/day total) colostrum or whey protein complex  
118 (containing about 38g of protein per 60g of complex) measured with a scoop provided. This dose  
119 was chosen because it is effective in young individuals for increasing lean tissue mass (Antonio  
120 et al., 2001; Kerksick et al., 2007). The bovine colostrum used in this study was a heat-treated  
121 spray-dried >25% IgG commercially available product (trade-named Eterna Gold<sup>TM</sup>  
122 manufactured and marketed by the Saskatoon Colostrum Co. Ltd., Saskatoon, Canada). The  
123 product is derived from first day post partum excess colostrum collected from Canadian dairy  
124 cows and is licensed by Health Canada as a natural health product for immune system and  
125 athletic support (Natural Health Product Number 80035324; full details on the product can be  
126 viewed at: [http://www.saskatooncolostrum.com/english/Article/Details/4779\\_Eterna-Gold-  
127 -Colostrum-For-People.html](http://www.saskatooncolostrum.com/english/Article/Details/4779_Eterna-Gold-<br/>127 -Colostrum-For-People.html)). Whey protein was used as the placebo because it matches bovine  
128 colostrum for protein content but does not have substantial effects on muscle size and strength or  
129 bone resorption in older adults (Candow, Chilibeck, Facci, Abeysekara, & Zello, 2006). The  
130 whey placebo was purchased commercially from Cereal By Products Co., Mt. Prospect, Illinois  
131 and was selected to match to the colostrum in overall nutritional composition (Table 1). The  
132 composition of the bovine colostrum and whey protein supplements was verified by an  
133 independent laboratory (SunWest Food Laboratory Ltd., Saskatoon SK, Canada). On exercise  
134 days participants were instructed to take one dose within 30 minutes before and another dose  
135 within 30 minutes after their exercise session with a third dose at their discretion; on non-  
136 exercise days all doses were taken at the participant's discretion. Participants mixed supplement  
137 with liquid of choice (e.g. water, juice, or milk) in a provided blender. All participants were  
138 assigned a full body resistance program of 12 machine-based exercises. Participants were  
139 required to attend an orientation session to be familiarized with the machines and exercises prior

140 to starting their program. Following orientation, participants were instructed to complete three  
141 sets of 8-12 repetitions (working to fatigue) for each exercise, under supervision on three  
142 separate days, again for familiarization with the exercises and to reduce any “learning” effects  
143 prior to strength testing. Participants were then tested for 1-RM strength. The exercise  
144 intervention was conducted three non-consecutive days per week and included three sets of 8-12  
145 repetitions on Lever machines (Pulse Fitness Systems; Winnipeg, Manitoba, Canada) (with  
146 exception of abdominal crunches) for the following exercises: bench press, iso-lateral lat  
147 pulldown, shoulder press, biceps curl, triceps extension, leg press, leg flexion and extension,  
148 back extension, and hip adduction and abduction. All sets were performed to fatigue and  
149 resistance was progressively increased once a participant could complete 12 repetitions with  
150 good form. All exercise sessions were supervised by Canadian Society for Exercise Physiology-  
151 Certified Exercise Physiologists to ensure proper form and resistance; this ensured compliance to  
152 each prescribed exercise and the appropriate sets and repetitions. In addition to tracking  
153 workouts and recording supplement compliance in logs, participants were required to sign an  
154 attendance sheet at each visit. Adverse events during the study were recorded on adverse event  
155 forms.

156       Following the intervention participants were asked which supplement they thought they  
157 were receiving (to test if blinding was effective) and asked to return remaining supplement to be  
158 weighed as confirmation of supplement compliance.

159

### 160 ***Outcome Measures***

161       All variables were assessed at baseline and after the eight-week intervention. Variables  
162 assessed included muscle thickness of the elbow flexors and knee extensors by ultrasound, IGF-

163 1, and C-reactive protein (CRP; as a marker of inflammation) from blood samples, urinary cross-  
164 linked N-telopeptides of Type 1 collagen (i.e. Ntx; bone resorption), body composition by dual  
165 energy X-ray absorptiometry (DXA), strength by determination of 1-repetition maximum (1-RM)  
166 on bench press and leg press, and cognitive function with the Telephone Interview of Cognitive  
167 Status (TICS; de Jager, Budge, & Clarke, 2003) questionnaire. Strength testing was always done  
168 last so as not to influence muscle thickness or body composition testing because of muscle  
169 swelling. Measurement techniques are described in detail below.

170 **Body Composition.** Body composition was assessed with DXA in array mode (QDR Discovery  
171 Wi, Hologic, Inc., Bedford, Md.) using QDR software for Windows XP (QDR Discovery). Lean  
172 tissue mass, fat mass, and bone mineral content were assessed from whole-body scans. The  
173 coefficients of variation for these measurements are 0.5%, 3%, and 0.5% respectively (Chilibeck  
174 et al., 2013).

175 **Strength.** Strength (1-RM) was assessed during the bench press and leg press exercises, which  
176 were chosen as representative exercises for upper- and lower-body strength. We have previously  
177 described these assessments elsewhere (Chrusch, Chilibeck, Chad, Davison, & Burke, 2001).  
178 The coefficients of variation for these measurements are 3.0% and 3.6% for leg press and bench  
179 press, respectively (Chrusch et al., 2001).

180 **Muscle Thickness.** Ultrasound was used to assess muscle thickness of the elbow flexors and  
181 knee extensors of the dominant limb prior to 1RM testing. We have described these methods in  
182 detail elsewhere (Farthing & Chilibeck, 2003; Candow & Chilibeck, 2005). The coefficients of  
183 variation (CVs) for muscle thickness measurements are 2.5% for elbow flexors and 2.1% for  
184 knee extensors (Candow & Chilibeck, 2005).

185 **Serum Assessment.** Blood samples were drawn from an antecubital vein, centrifuged and plasma  
186 harvested and separated into aliquots, which were frozen at -80 degrees C. Samples were thawed  
187 and analyzed for: a) IGF-1 using ELISA (Enzo Life Sciences) (intra-assay CV = 3.6%) and b)  
188 CRP as a marker of inflammation using ELISA (ALPCO Diagnostics) (intra-assay CV = 3.8%).  
189 Samples from all time points for each individual were analyzed in the same assay to eliminate  
190 between-assay variability.

191 **Bone Resorption.** Participants were instructed to collect 24-hour urine samples as previously  
192 described (Pinkoski et al., 2006). Baseline urine collection was completed prior to starting the  
193 study and post-intervention urine collection was completed in the 3 days after the exercise  
194 intervention. Participants continued consuming the supplement during these 3 days. Alcohol and  
195 intense exercise was prohibited during the 24 hours of collection and the 24 hours prior.  
196 Returned urine containers were measured for urine volume, and aliquots were removed and  
197 frozen at -80 degrees C prior to being thawed and analyzed in duplicate within the same assay by  
198 ELISA (Osteomark Ntx test, Ostex International, Inc., Seattle, WA) (intra-assay CV = 6.7%) for  
199 bone resorption via Ntx. The concentration of Ntx in urine samples [expressed as bone collagen  
200 equivalents (BCE)] was corrected for urinary creatinine and multiplied by 24-h urine volume to  
201 produce a value for daily Ntx excretion relative to daily creatinine excretion. Creatinine was  
202 assessed by a commercially available colorimetric kit (Cayman Chemical Co., Ann Arbor, MI)  
203 (intra-assay CV = 6.0%). The concentration was multiplied by 24-hr urine volume to determine  
204 the amount excreted over 24 hr.

205 **Cognitive Function.** Cognitive function was assessed via the TICS (de Jager et al., 2003)  
206 questionnaire, administered in person. TICS assesses four domains: a) orientation, b)  
207 registration, recent memory and delayed recall (memory), c) attention and calculation, and d)

208 semantic memory, comprehension and repetition (language). TICS was originally developed as a  
209 dementia screen, but is useful for tracking cognitive function over time (de Jager et al., 2003).

### 210 ***Diet and Physical Activity Monitoring***

211 Participants completed the Godin Leisure-Time Exercise Questionnaire (Godin &  
212 Shephard, 1985) during baseline testing and at the end of the intervention, and were asked to  
213 include only physical activities outside of the intervention. Participants were told at the start of  
214 the study not to change their diets substantially during the study. Participants were given two 3-  
215 day food logs; one to be completed prior to starting the supplement and one during the last week  
216 of the intervention to ensure diets remained consistent throughout the intervention. Food logs  
217 were entered and analyzed via United States Department of Agriculture Center for Nutrition  
218 Policy and Promotion (USDA, Alexandria, VA) online food tracker SuperTracker.

### 219 **Data Analysis**

220 An independent t-test was used to assess baseline characteristics and to compare  
221 compliance between groups. Repeated-measures ANOVA with within-factor defined as time and  
222 between-factors defined as gender and group was used to assess all dependent variables. The  
223 following assumptions were tested and met: a) independence of observations, b) normality, and  
224 c) sphericity. All analyses were done using IBM SPSS (Statistics version 20, Chicago). To  
225 ensure statistical results for the leg press strength measurement were not due to differences  
226 between baseline means, we also ran an analysis of covariance for this variable, testing for  
227 differences between groups at 8 weeks, using baseline strength as a covariate. Analysis was done  
228 on an intent-to-treat basis. Results are expressed as means and standard deviations. Significance  
229 was accepted when  $p \leq 0.05$ .

230

## 231 **Results**

232 Baseline characteristics were not significantly different between groups. Participants in the  
233 colostrum group were on average  $78.6 \pm 17.6$  kg,  $171 \pm 7$  cm, and  $61.8 \pm 4.8$  y as compared to  
234 participants in the whey group who were on average  $74.0 \pm 19.2$  kg,  $169 \pm 9$  cm, and  $57.5 \pm 6.3$  y.

235 **Compliance and Blinding.** One female participant from the whey group withdrew due to  
236 personal reasons and was lost to follow-up. Two participants from the whey group discontinued  
237 use of supplement due to gastrointestinal reflux the investigators classified as “definitely” related  
238 to the supplement, but continued with the exercise training. Exercise and supplement compliance  
239 was not significantly different between groups. Participants in the colostrum group were  $86 \pm$   
240  $20\%$  compliant to the exercise and  $97 \pm 12\%$  compliant to the supplement compared to  
241 participants in the whey group who were  $84 \pm 21\%$  compliant to the exercise and  $88 \pm 23\%$   
242 compliant to the supplement. Thirty-seven percent of the participants in the colostrum group and  
243  $25\%$  of participants in the whey protein group correctly guessed their group assignment.

244 **Gender Differences.** As expected, there were several gender-based effects. Males had greater leg  
245 press and bench press strength, IGF-1 levels, bone mineral content, lean tissue mass, elbow  
246 flexors and knee extensors muscle thickness, and kcal, protein, and carbohydrate intake  
247 compared to females ( $p < 0.05$ ). Males had lower percent body fat and cognitive scores compared  
248 to females ( $p < 0.01$ ).

249 **Body Composition.** Over time, there was a significant increase in lean tissue mass ( $p < 0.001$ ) and  
250 bone mineral content ( $p = 0.012$ ) and a significant decrease in percent fat ( $p < 0.01$ ) with no  
251 differences between groups (Table 2). There were no significant changes in fat mass (Table 2).

252 **Strength.** There was a significant group by time interaction for leg press strength, with strength  
253 increasing more in the colostrum group than the whey protein group ( $p = 0.026$ ; Figure 1).

254 Baseline and post-intervention strength measures for the colostrum group were  $121 \pm 40$  and  $145$   
255  $\pm 53$  kg, and for the whey protein group were  $143 \pm 51$  kg and  $151 \pm 58$  kg. An analysis of  
256 covariance with baseline leg press strength as the covariate indicated that adjusted means at the  
257 end of the intervention were significantly greater in the colostrum compared to the whey protein  
258 group ( $165 \pm 5$  kg vs.  $149 \pm 5$  kg;  $p=0.045$ ). There were no differences for changes in bench  
259 press strength between groups (Figure 2). There was a significant time main effect for bench  
260 press strength ( $p < 0.001$ ) with the colostrum group increasing from  $57 \pm 31$  to  $69 \pm 35$  kg and  
261 the whey protein group increasing from  $63 \pm 37$  to  $79 \pm 46$  kg. There was a gender by time  
262 interaction for leg press and bench press strength, ( $p < 0.05$ ), with males increasing more than  
263 females (data not shown).

264 **Muscle Size.** Muscle thickness of the knee extensors and elbow flexors increased over time  
265 ( $p < 0.001$ ) with no difference between groups (Table 2).

266 **Serum & Urine Measurements.** There was a group by time interaction for urinary Ntx, with the  
267 colostrum group decreasing more than the whey protein group ( $p = 0.024$ ; Table 2). There were  
268 no differences between groups over time, nor were there any time main effects for levels of CRP  
269 and IGF-1 (Table 2).

270 **Questionnaires.** There was a significant increase in cognitive function over time ( $p = 0.015$ ) with  
271 no differences between groups (Table 2). There were no significant differences over time or  
272 between groups for leisure time physical activity (Table 2).

273 **Nutrition.** Both groups decreased dietary protein intake (excluding the nutritional supplement)  
274 over time ( $p = 0.047$ ) (Table 3). There were no differences between groups over time for any  
275 nutritional variables (Table 3).

276 **Adverse Events.** Five participants reported adverse events related to gastrointestinal problems.  
277 Two participants consuming colostrum reported adverse events classified as ‘mild’ in severity  
278 included bloating, nausea, diarrhea, and unsettled stomach. The researchers classified the adverse  
279 events as either ‘probable’ or ‘possible’. The two participants continued taking the colostrum  
280 supplement for the remainder of the study; however one reduced the dosage. The other three  
281 adverse events were also related to gastrointestinal problems in participants consuming whey.  
282 Two of these three adverse events were classified as ‘moderate’ in severity (gastro esophageal  
283 reflux). These adverse events were considered “definitely” related to the supplement based on  
284 cessation of symptoms upon stopping the supplement and reappearance of the adverse event  
285 upon re-introduction. Both participants discontinued the supplement. The other participant’s  
286 adverse event was ‘mild’ in severity (nausea), and considered ‘possibly’ related to the  
287 supplement. This participant continued taking the supplement.

288

## 289 **Discussion**

290 The present study is the first to examine the effects of bovine colostrum supplementation  
291 during a resistance training program in older adults. Colostrum supplementation promoted  
292 greater increases in leg press strength than did whey protein. Colostrum supplementation also  
293 reduced bone resorption compared to whey protein. Both colostrum and whey protein  
294 supplemented groups significantly increased bench press strength, muscle size, cognitive  
295 function, lean tissue mass, and bone mineral content over time. Males had greater baseline values  
296 for most outcome measures, which was to be expected and is supported by previous studies  
297 (Chilibeck, Stride, Farthing, & Burke, 2004). Despite this, male and female participants

298 responded equally to the supplementation (i.e. there were no supplement group x gender x time  
299 interactions).

300           The increase in leg press strength associated with colostrum supplementation is  
301 important because older adults lose strength in the lower body to a greater extent than in the  
302 upper body (Candow & Chilibeck, 2005; IWGS, 2011). The group supplemented with colostrum  
303 increased leg press strength by about 21% whereas the group supplemented with whey protein  
304 had a non-significant increase in leg press strength of about 5%. The increase in the colostrum  
305 group might be clinically significant because a 20% decline in leg press strength with aging is  
306 associated with increased functional limitations (Brill, Macera, Davis, Blair, & Gordon, 2000).  
307 The mechanism for the greater increase in leg press strength in the colostrum group is unclear  
308 because the groups did not differ in changes in lean tissue mass or knee extensor muscle  
309 thickness. A possible explanation as to the lack of increase in leg press strength in the whey  
310 group is they had slightly (but not statistically) higher baseline strength and therefore may have  
311 been closer to their physiological ceiling and had less room for improvement. The greater  
312 increase in leg press strength in the colostrum group could be due to statistical error (i.e. type I  
313 error with multiple statistical tests). Both groups increased equally in bench press strength. Males  
314 increased leg press and bench press strength more than females; this is supported by previous  
315 studies (Chilibeck et al., 2004). Further research is needed to determine if there is a true increase  
316 in strength due to colostrum supplementation or whether other factors such as an increase in  
317 muscle quality may be responsible for the apparent increase in strength.

318           The participants receiving bovine colostrum had a greater decrease in bone resorption  
319 (assessed by urinary Ntx) compared to participants consuming whey protein. This suggests  
320 bovine colostrum might have benefits for bone health. Previously Brinkworth, Buckley,

321 Slavotinek, & Kurmis (2004) showed a trend ( $p=0.06$ ) toward a greater increase in bone cross-  
322 sectional area in the trained upper arm of participants supplemented with colostrum compared to  
323 whey protein for eight weeks. A number of studies using animal models have also suggested a  
324 positive effect of bovine colostrum on bone. Supplementation with proteins extracted from  
325 bovine colostrum (i.e. osteopontin, lactoferrin, epidural growth factor, and IGF-2) increased  
326 mineral density, micro-architectural properties, and mechanical strength of bones from  
327 ovariectomized rats (a model for postmenopausal osteoporosis), and reduced markers of bone  
328 resorption and increased markers of bone formation in serum (Du et al., 2011; Hou, Xue, & Lin,  
329 2012). Bovine colostrum or proteins derived from colostrum (i.e. lactoferrin) increase the  
330 proliferation of osteoblasts (i.e. cells involved in bone formation) and the release of growth  
331 factors from osteoblasts derived from rats (Lee et al., 2008; Nakajima et al., 2011), and bovine  
332 colostrum reduces activity of osteoclasts (i.e. cells involved in bone resorption) derived from  
333 rabbits (Vidal, van den Broek, Lorget, & Donnet-Hughes, 2004).

334       Bovine colostrum contains substantial amounts of IGF-1 (Marcotty et al., 1991). IGF-1 is  
335 the major mediator of growth hormone (GH) and is linked to muscle hypertrophy (Allen &  
336 Boxhorn, 1989). Participants in this study had no increase in serum IGF-1 levels with colostrum  
337 supplementation. While Mero et al. (2002) showed that levels of plasma IGF-1 increased after 2  
338 weeks of bovine colostrum supplementation and training in male and female athletes, most other  
339 studies have shown levels of plasma IGF-1 did not increase after bovine colostrum  
340 supplementation and training (Buckley et al., 2003; Buckley, Abbott, Brinkworth, & Whyte,  
341 2002; Shing et al., 2009). It is theorized that the increase in IGF-1 in the Mero et al. (2002) study  
342 may have been transient due to the short supplementation period, whereas studies that have  
343 longer supplementation periods may allow enough time for the body to facilitate a negative

344 feedback and return the plasma concentrations to normal (Buckley et al., 2003). It should also be  
345 noted that Mero et al. (2002) used carbohydrate as a control; whereas other studies used protein  
346 as a control. This may also account for differences in IGF-1 responses between studies.

347 We found no significant differences between colostrum and whey protein groups for  
348 changes in systemic inflammation (i.e. C-reactive protein). Similarly Crooks et al. (2010) found  
349 no significant differences in C-reactive protein after 10 weeks of daily 50g supplementation of  
350 either colostrum or skim-milk powder during intense swim training (both in water and on-land).

351 It was hypothesized that bovine colostrum would improve cognitive function in older  
352 adults by increasing IGF-1 levels. There was an increase in cognitive function over time for both  
353 groups. This indicates the exercise training itself may have increased cognitive function. Physical  
354 activity is well known to enhance cognitive function (Forte et al., 2013) and reduce risk of  
355 cognitive decline and dementia (de Bruijn et al., 2013). It is also possible that there was a  
356 learning effect with our specific cognitive test, as participants may have strategized for the word  
357 recall portion of the TICS questionnaire administered at the end of the intervention.

358 The decrease in protein intake over time for both groups is likely due to participants  
359 compensating for the additional protein provided by the supplement by reducing protein  
360 consumption elsewhere in their diet. As the food logs analyzed did not include the supplement,  
361 the post values for protein consumption do not include the 38 grams of protein provided by the  
362 colostrum or the whey.

### 363 ***Conclusion***

364 Bovine colostrum may have benefits over whey protein for increasing lower body  
365 strength and reducing bone resorption in older adults but had no effect beyond those seen with  
366 whey supplementation and resistance training on measurements of upper body strength, IGF-1,

367 inflammation, or body composition. Our finding that short-term colostrum supplementation  
368 decreased bone resorption compared to whey protein suggests that the long-term effects of  
369 bovine colostrum on clinically relevant measures of bone health (i.e. hip or lumbar spine bone  
370 mineral density) should be investigated.

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526 **Figure Captions**

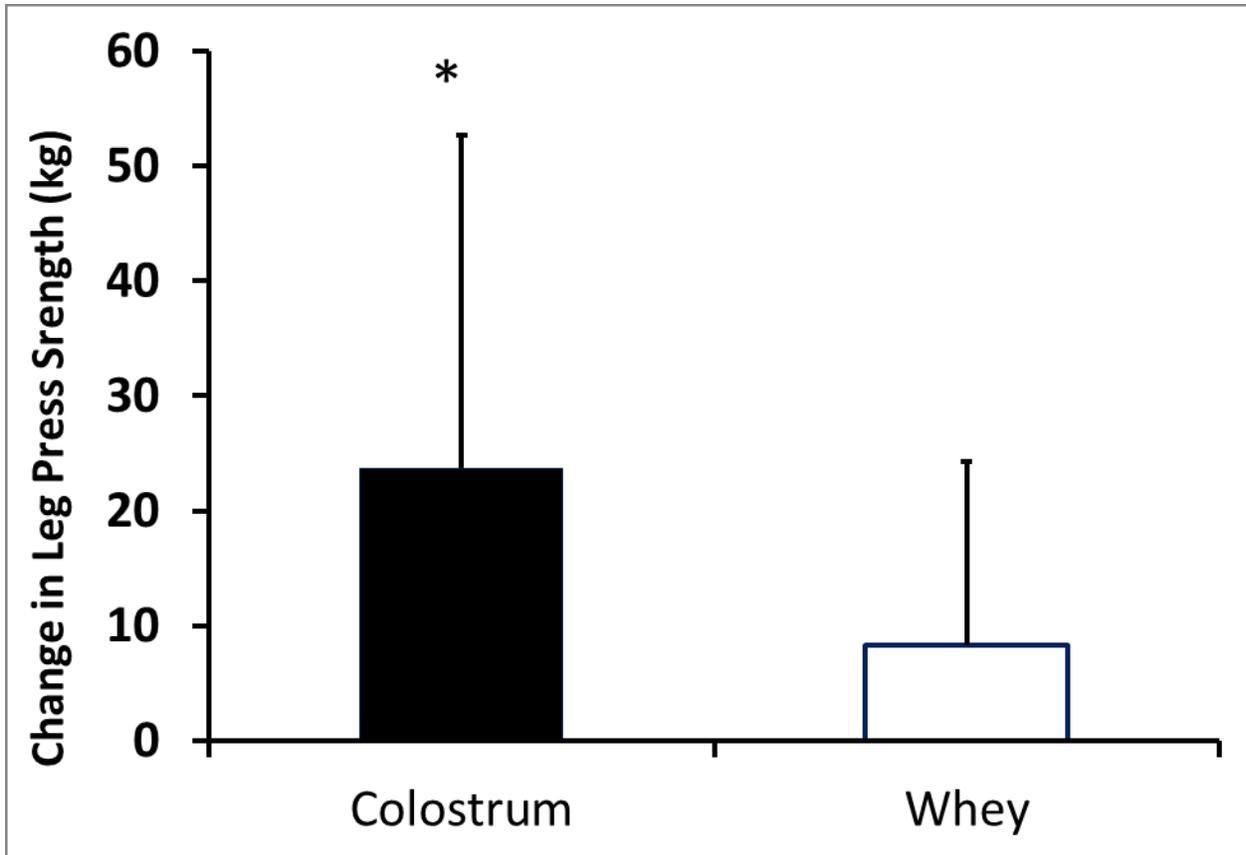
527

528 Figure 1. Change in leg press strength for colostrum and whey-protein treatment groups. Data are  
529 means and SD. \*Difference between the colostrum vs. the whey protein group ( $p < 0.05$ ).

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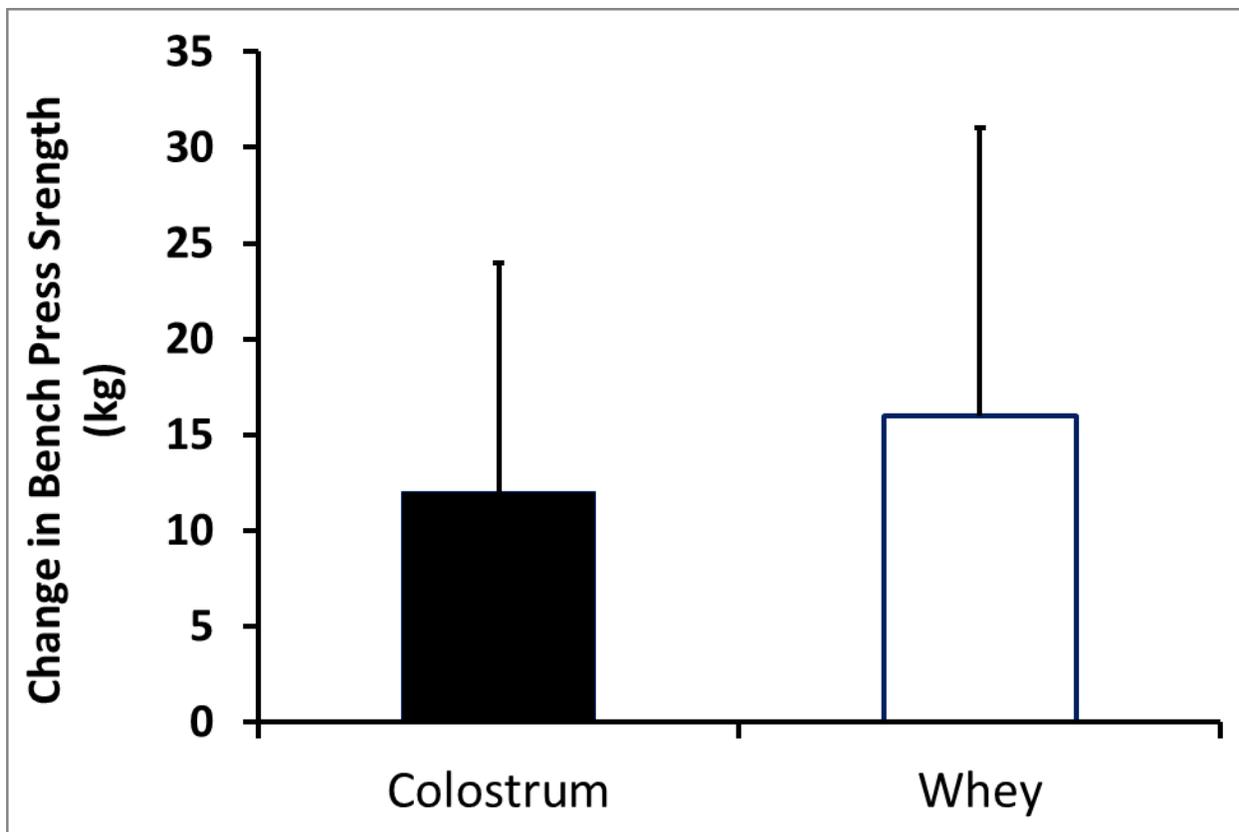
531 Figure 2. Change in bench press strength for colostrum and whey-protein groups. Data are means  
532 and SD. \*Time main effect ( $p < 0.05$ ).

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Table 1

*Colostrum versus Whey Nutritional Breakdown*

	Colostrum	Whey
Crude Protein (%)	62.4	64.6
Crude Fat (%)	13.9	14.7
Carbohydrates (g/100g)	13.5	12.5
Calories/100g	429	441

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542 Table 2

543 *Body Composition, Muscle Thickness, Serum, Urine, Leisure Time Activity and Cognitive Function Results*

	<u>Colostrum</u>		<u>Whey</u>	
	Baseline	Post	Baseline	Post
BMC (kg)*	2.44 ± 0.39	2.47 ± 0.40	2.41 ± 0.59	2.42 ± 0.61
Fat mass (kg)	27.5 ± 13.0	27.4 ± 13.1	25.0 ± 9.3	24.8 ± 9.0
Lean tissue mass (kg)*	47.8 ± 9.4	48.5 ± 9.0	46.3 ± 12.7	46.8 ± 12.8
Total mass (kg)	77.7 ± 18.0	78.4 ± 17.5	73.7 ± 19.3	74.1 ± 19.1
Fat (%)*	34.1 ± 10.5	33.7 ± 10.6	33.7 ± 7.9	33.3 ± 7.8
Biceps (cm)*	2.64 ± 0.75	2.91 ± 0.76	2.54 ± 0.59	2.81 ± 0.65
Quadriceps (cm)*	2.73 ± 0.53	2.95 ± 0.65	2.57 ± 0.47	2.78 ± 0.53
CRP (mg/l)	2.3 ± 2.6	2.4 ± 3.2	2.1 ± 2.9	2.5 ± 3.5
IGF-1 (ng/ml)	155.3 ± 35.4	156.1 ± 36.1	162.3 ± 44.1	159.0 ± 42.0
Ntx (nmol BCE/mmol Crn)	1085 ± 585	770 ± 359**	1074 ± 614	1172 ± 762
LTEQ	29 ± 30	29 ± 25	33 ± 22	29 ± 21
TICS*	28 ± 2	29 ± 2	28 ± 3	30 ± 4

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545 Data are means ± standard deviation; BMC = bone mineral content; CRP = C-reactive protein; IGF-1 = Insulin-like

546 growth factor-1, Ntx = cross-linked n-telopeptides of type I collagen; BCE = bone collagen equivalents; Crn=

547 creatinine; TICS = Telephone Interview of Cognitive Status

548 \*Time main effect (p&lt;0.05).

549 \*\*The change in the colostrum group was greater than the whey protein group (p&lt;0.05)

550

551 Table 3

552 *Nutrition*

	<u>Colostrum</u>		<u>Whey</u>	
	Baseline	Post	Baseline	Post
Calories (g/day)	2001 ± 472	1886 ± 292	1726 ± 506	1673 ± 522
Fat (g/day)	68 ± 20	67 ± 18	62 ± 24	58 ± 32
Carbohydrates (g/day)	256 ± 68	234 ± 44	215 ± 53	212 ± 59
Protein (g/day) without supplement *	85 ± 20	81 ± 23	73 ± 22	63 ± 21
Protein (g/day) with supplement		119 ± 23		98 ± 31
Protein (g/kg) without supplement	1.16 ± 0.44	1.08 ± 0.38	0.91 ± 0.39	0.79 ± 0.36
Protein (g/kg) with supplement		1.59 ± 0.47		1.34 ± 0.36

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554 Data are means ±standard deviations

555 \*Time main effect (p&lt;0.05)

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