Systematic review and meta-analysis of protein intake to support muscle mass and function in healthy adults

Everson A Nunes[1,2], Lauren Colenso-Semple[1], Sean R McKellar[1], Thomas Yau[1], Muhammad Usman Ali[3], Donna Fitzpatrick-Lewis[3], Diana Sherifali[4], Claire Gaudichon[5], Daniel Tomé[5], Philip J Atherton[6], Maria Camprubi Robles[7], Sandra Naranjo-Modad[8], Michelle Braun[9], Francesco Landi[10] and Stuart M Phillips[1*]

[1] Exercise Metabolism Research Group, Department of Kinesiology, McMaster University, Hamilton, L8S 4L8, ON, Canada.

[2]Laboratory of Investigation of Chronic Diseases, Department of Physiological Sciences, Federal University of Santa Catarina, Florianópolis, 88040-900, SC, Brazil.

[3]McMaster Evidence Review and Synthesis Centre, McMaster University, Hamilton, L8S 4L8, ON, Canada.

[4]School of Nursing, Faculty of Health Sciences, McMaster University, Hamilton, L8S 4L8, ON, Canada.

[5]Université Paris-Saclay, AgroParisTech, INRAE, UMR PNCA, 75005 Paris, France

[6]MRC Versus Arthritis Centre of Excellence for Musculoskeletal Ageing Research (CMAR), NIHR Biomedical Research Centre, School of Medicine, University of Nottingham, Nottingham, NG7 2RD, England.

[7]Abbott Nutrition, Research and Development, Granada, Spain

[8]Givaudan, Research and Development, Avignon, France

[9] International Flavors & Fragrances, Research and Development, St. Louis, MO, USA

[10]Geriatric Internal Medicine Unit of the Geriatrics Department of the A. Gemelli University Hospital, Rome, IT

* Corresponding author: S. M. Phillips, phillis@mcmaster.ca and publications@ilsieurope.be; Tel.: +1-905-525-9140 (ext. 24465). Mailing address: Department of Kinesiology, Ivor Wynne Centre, McMaster University, 1280 Main St. West, Hamilton, ON L8S 4L8.

Supplementary Table 2 – GRADE evidence profile rating for lean body mass changes in studies testing additional dietary protein intervention in healthy subjects.

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
Overall effects of ingesting additional protein on lean or muscle mass gain									
66	randomised trials	serious [a,b]	not serious	not serious [c]	not serious [d]	publication bias strongly suspected [e]	SMD **0.22 SD higher** (0.15 higher to 0.29 higher)	⨁⨁⨁◯ MODERATE	CRITICAL
Overall effects of ingesting additional protein and RE on lean or muscle mass gain in studies reporting protein ingestion									
51	randomised trials	serious [f,g]	not serious [h]	not serious	not serious	publication bias strongly suspected [i]	SMD **0.19 SD higher** (0.11 higher to 0.28 higher)	⨁⨁⨁◯ MODERATE	CRITICAL
Lean Body Mass gain by protein ingestion and RE - Ingestion of <1.2g of protein/kg/day									
4	randomised trials	serious [j]	not serious	not serious	serious [k]	none	SMD **0.14 lower** (0.56 lower to 0.27 higher)	⨁◯◯◯ VERY LOW	IMPORTANT
Lean Body Mass gain by protein ingestion and RE - Ingestion of 1.2 - 1.59g of protein/kg/day									
24	randomised trials	serious [a,l]	not serious	not serious	not serious	none [i]	SMD **0.17 higher** (0.06 higher to 0.28 higher)	⨁⨁⨁◯ MODERATE	CRITICAL
Lean Body Mass gain by protein ingestion and RE - Ingestion of 1.6g of protein/kg/day or higher									
23	randomised trials	serious [f,l]	not serious	not serious	not serious	none [i]	SMD **0.29 SD higher** (0.13 higher to 0.45 higher)	⨁⨁⨁◯ MODERATE	CRITICAL
Lean body or muscle mass gain with protein supplementation and without resistance exercise									

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
6	randomised trials	serious [l]	not serious	not serious	serious [k]	none	SMD **0.21 higher** (0.05 lower to 0.48 higher)	⨁⨁◯◯ LOW	CRITICAL

Lean body or muscle mass gain with protein supplementation and resistance exercise

62	randomised trials	serious [a,g]	not serious	not serious	not serious	none [e]	SMD **0.22 SD higher** (0.14 higher to 0.3 higher)	⨁⨁⨁◯ MODERATE	CRITICAL

Lean Body Mass by Age by RE - < 65 years old

48	randomised trials	serious [f,p]	not serious	not serious	not serious	none [i]	SMD **0.25 SD higher** (0.16 higher to 0.35 higher)	⨁⨁⨁◯ MODERATE	IMPORTANT

Lean Body Mass by Age by RE - > 65 years old

14	randomised trials	serious [m]	not serious	not serious	not serious	none	SMD **0.13 SD higher** (0.00 lower to 0.28 higher)	⨁⨁◯◯ LOW	IMPORTANT

Lean Body Mass gain by protein ingestion and RE - Ingestion of 1.2 - 1.59g of protein/kg/day <65 years old

15	randomised trials	serious [a,g]	not serious	not serious	not serious	none	SMD **0.15 SD higher** (0.02 lower to 0.31 higher)	⨁⨁◯◯ LOW	IMPORTANT

Lean Body Mass gain by protein ingestion and RE - Ingestion of 1.2 - 1.59g of protein/kg/day ≥ 65 years old

9	randomised trials	serious [a,g]	not serious	not serious	not serious [k]	none	SMD **0.20 SD higher** (0.02 higher to 0.37 higher)	⨁⨁◯◯ LOW	IMPORTANT

Lean Body Mass gain by protein ingestion and RE - 1.6g of protein/kg/day or higher <65 years old

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
23	randomised trials	serious [a,g]	not serious	not serious	not serious	none	SMD **0.30 SD higher** (0.17 lower to 0.43 higher)	⨁⨁⨁◯ MODERATE	IMPORTANT

CI: Confidence interval; **SMD:** Standardised mean difference

Explanations

a. At least 30% of the studies presented a potential unclear or high risk of bias for using no blinding or single-blinded study designs.
b. Seven studies present a potential high or unclear risk of bias for randomization and allocation.
c. Low heterogeneity
d. Both intervention and place/control groups have >1200 subjects each.
e. Taylor 2016 and Willoughby 2007 are outside CI 95% during visual funnel plot analysis
f. At least 40% of the studies present a potential unclear or high risk of bias due to improper description of randomization and blinding of research subjects or staff
g. Some studies present a potential high or unclear risk of bias for randomization and allocation.
h. Overall heterogeneity is low. However, heterogeneity between subgroups comparison is significantly high.
i. Taylor 2016 and Willoughby 2007 are outside CI 95% during visual funnel plot analysis. Both studies tested subjects <65 years old submitted to resistance exercise training.
j. 40-50% of the studies show a potential high or unclear risk of bias for blinding of research subjects and staff.
k. A low number of studies with less than 200 subjects in each treatment group.
l. Two studies present a potential high or unclear risk of bias for blinding of participants and research staff.
m. At least 50% of the studies present a potentially unclear or high risk of bias for blinding of research participants and research staff.
n. Two studies present a potential unclear or high risk for selection bias.
o. Three studies a potential unclear risk for selection bias.
p. Five studies present a potential high or unclear risk of bias for randomization and allocation.

Supplementary Table 3 – GRADE evidence profile rating for muscle strength changes in studies testing additional dietary protein intervention in healthy subjects.

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
Bench Press strength in protein supplemented subjects overall effect									
34	randomised trials	serious [a]	serious [b]	not serious	not serious	none [c]	SMD **0.20 higher** (0.06 higher to 0.34 higher)	⨁⨁◯◯ LOW	CRITICAL
Bench Press strength in protein supplemented subjects with RE									
33	randomised trials	serious [a]	serious [b]	not serious	not serious	none [c]	SMD **0.18 higher** (0.04 higher to 0.32 higher)	⨁⨁◯◯ LOW	CRITICAL
Bench Press strength in protein supplemented subjects in studies reporting daily protein ingestion with RE									
16	randomised trials	serious [a]	serious [d]	not serious	not serious	none	SMD **0.18 higher** (0.07 lower to 0.42 higher)	⨁⨁◯◯ LOW	IMPORTANT
Bench Press strength in protein supplemented subjects with RE - Ingestion of 1.2 - 1.59g of protein/kg/day									
14	randomised trials	serious [a]	serious [d]	not serious	not serious	none	SMD **0.17 higher** (0.01 lower to 0.35 higher)	⨁⨁◯◯ LOW	IMPORTANT
Bench Press strength in protein supplemented subjects with RE - Ingestion of 1.6g of protein/kg/day or higher									
16	randomised trials	serious [a]	serious [d]	not serious	not serious	none	SMD **0.13 higher** (0.15 lower to 0.41 higher)	⨁⨁◯◯ LOW	IMPORTANT
Bench Press strength in protein supplemented subjects with RE by Age - <65 years old									

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
33	randomised trials	serious [a]	serious [b]	not serious	not serious	none	SMD **0.18 higher** (0.03 higher to 0.33 higher)	⨁⨁◯◯ LOW	CRITICAL

Lower body strength in protein supplemented subjects overall effect

50	randomised trials	serious [f]	serious [b]	not serious	not serious	none [g]	SMD **0.20 higher** (0.08 higher to 0.33 higher)	⨁⨁◯◯ LOW	CRITICAL

Lower body strength in protein supplemented subjects in studies reporting protein ingestion with RE

42	randomised trials	serious [f]	serious [d]	not serious	not serious	none [g]	SMD **0.22 higher** (0.08 higher to 0.35 higher)	⨁⨁◯◯ LOW	CRITICAL

Lower body strength in protein supplemented subjects with RE - Ingestion of <1.2g of protein/kg/day

1	randomised trials	very serious [h]	not serious	not serious	serious [e]	none	SMD **0.16 lower** (1.09 lower to 0.77 higher)	⨁◯◯◯ VERY LOW	IMPORTANT

Lower body strength in protein supplemented subjects with RE - Ingestion of 1.2 - 1.59g of protein/kg/day

20	randomised trials	serious [a]	serious [d]	not serious	not serious	none	SMD **0.08 higher** (0.10 lower to 0.27 higher)	⨁⨁◯◯ LOW	IMPORTANT

Lower body strength in protein supplemented subjects with RE - Ingestion of 1.6g of protein or higher /kg/day

19	randomised trials	serious [a]	serious [d]	not serious	not serious	none	SMD **0.40 higher** (0.19 higher to 0.56 higher)	⨁⨁◯◯ LOW	IMPORTANT

Lower body strength in protein supplemented subjects with RE - Ingestion of 1.6g of protein or higher /kg/day <65 years old

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
17	randomised trials	serious [a]	serious [d]	not serious	not serious	none	SMD 0.40 higher	⨁⨁◯◯ LOW	IMPORTANT

Lower body strength in protein supplemented subjects - No resistance exercise program

4	randomised trials	serious [a]	not serious	not serious	very serious [e]	none	SMD **0.14 higher** (0.36 lower to 0.64 higher)	⨁◯◯◯ VERY LOW	CRITICAL

Lower body strength in protein supplemented subjects - Resistance exercise training

47	randomised trials	serious [f]	serious [d]	not serious	not serious	none	SMD **0.21 higher** (0.08 higher to 0.34 higher)	⨁⨁◯◯ LOW	CRITICAL

Lower body strength in protein supplemented subjects with RE by Age - <65 years old

35	randomised trials	serious [f]	serious [b]	not serious	not serious	none	SMD **0.19 higher** (0.03 higher to 0.36 higher)	⨁⨁◯◯ LOW	CRITICAL

Lower body strength in protein supplemented subjects with RE by Age - >65 years old

12	randomised trials	serious [a]	serious [d]	not serious	not serious	none	SMD **0.22 higher** (0.01 higher to 0.48 higher)	⨁⨁◯◯ LOW	CRITICAL

CI: Confidence interval; **SMD:** Standardised mean difference

Explanations

a. At least 30% of the studies presented a potential unclear or high risk of bias using no blinding or single-blinded study designs.
b. Heterogeneity 50 - 60%
c. Funnel plots visual analysis shows Obradovic 2020, Vangsoe 2018 and Willoughby 2007 out of 95% CI lines
d. Heterogeneity 40 - 50%
e. Less than 300 subjects in the intervention or control/placebo group
f. At least 40% of the studies presented a potential unclear or high risk of bias using no blinding or single-blinded study designs.
g. Funnel plots visual analysis shows Aas 2020, Burke 2001 and Candow 2006 out of 95% CI lines
h. At least 60% of the studies presented a potential unclear or high risk of bias using no blinding or single-blinded study designs.

Supplementary Table 4 – GRADE evidence profile rating for handgrip strength changes in studies testing additional dietary protein intervention in healthy subjects.

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
Handgrip Strength									
10	randomised trials	serious [a,b]	not serious	serious [c]	serious [d]	none	SMD **0.15 higher** (0.03 lower to 0.32 higher)	⨁◯◯◯ VERY LOW	CRITICAL

CI: Confidence interval; **SMD:** Standardised mean difference

Explanations

a. At least 50% of the studies present a potentially unclear or high risk of performance and detection bias.
b. Two studies present a potentially unclear risk of selection bias.
c. Four studies with and six studies without resistance exercise training
d. Less than 300 subjects in the intervention or control/placebo group

Supplementary Table 5 – GRADE evidence profile rating for physical testing performance changes in studies providing additional dietary protein to healthy subjects.

Certainty assessment							Effect	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Absolute (95% CI)		
Additional protein and changes in physical testing performance.									
16	randomised trials	serious [a,b]	very serious [c]	not serious	not serious	none	SMD **0.15 higher** (0.00 higher to 0.29 higher)	⨁◯◯◯ VERY LOW	CRITICAL
Additional protein and changes in physical testing performance in studies with no resistance exercise program									
5	randomised trials	serious [a]	very serious	not serious	serious [d]	none	SMD **0.09 higher** (0.08 lower to 0.25 higher)	⨁◯◯◯ VERY LOW	CRITICAL
Additional protein and changes in physical testing performance in studies where subjects were submitted to resistance exercise									
11	randomised trials	very serious [c]	very serious [c]	not serious	not serious	none	SMD **0.17 higher** (0.03 lower to 0.37 higher)	⨁◯◯◯ VERY LOW	CRITICAL

CI: Confidence interval; **SMD:** Standardized mean difference

Explanations

a. At least 30% of the studies present a potentially unclear or right risk for blinding of research subjects and staff.
b. Two studies present an unclear risk of bias for allocation and randomization
c. Heterogeneity >50%
d. Total number of subjects in the intervention or controls is < 300.

References:

1. Aas SN, Seynnes O, Benestad HB, Raastad T. Strength training and protein supplementation improve muscle mass, strength, and function in mobility-limited older adults: a randomized controlled trial. Aging Clinical and Experimental Research. 2020;32:605-16. doi:10.1007/s40520-019-01234-2
2. Aleman-Mateo H, Carreon VR, Macias L, Astiazaran-Garcia H, Gallegos-Aguilar AC, Enriquez JRR. Nutrient-rich dairy proteins improve appendicular skeletal muscle mass and physical performance, and attenuate the loss of muscle strength in older men and women subjects: a single-blind randomized clinical trial. Clinical interventions in aging. 2014;9:1517-25. doi:10.2147/CIA.S67449
3. Amasene M, Besga A, Echeverria I, Urquiza M, Ruiz JR, Rodriguez-Larrad A, et al. Effects of Leucine-Enriched Whey Protein Supplementation on Physical Function in Post-Hospitalized Older Adults Participating in 12-Weeks of Resistance Training Program: A Randomized Controlled Trial. Nutrients. 2019;11:doi:10.3390/nu11102337
4. Antonio J, Peacock CA, Ellerbroek A, Fromhoff B, Silver T. The effects of consuming a high protein diet (4.4 g/kg/d) on body composition in resistance-trained individuals. Journal of the International Society of Sports Nutrition. 2014;11:19. doi:10.1186/1550-2783-11-19
5. Antonio J, Ellerbroek A, Silver T, Orris S, Scheiner M, Gonzalez A, et al. A high protein diet (3.4 g/kg/d) combined with a heavy resistance training program improves body composition in healthy trained men and women - a follow-up investigation. Journal of the International Society of Sports Nutrition. 2015;12:39. doi:10.1186/s12970-015-0100-0
6. Aristizabal JC, Freidenreich DJ, Volk BM, Kupchak BR, Saenz C, Maresh CM, et al. Effect of resistance training on resting metabolic rate and its estimation by a dual-energy X-ray absorptiometry metabolic map. Eur J Clin Nutr. 2015;69:831-6. doi:10.1038/ejcn.2014.216
7. Arnarson A, Geirsdottir OG, Ramel A, Briem K, Jonsson PV, Thorsdottir I. Effects of whey proteins and carbohydrates on the efficacy of resistance training in elderly people: double blind, randomised controlled trial. European Journal of Clinical Nutrition. 2013;67:821-6. doi:10.1038/ejcn.2013.40
8. Babault N, Deley G, Le Ruyet P, Morgan F, Allaert FA. Effects of soluble milk protein or casein supplementation on muscle fatigue following resistance training program: a randomized, double-blind, and placebo-controlled study. Journal of the International Society of Sports Nutrition. 2014;11:36. doi:10.1186/1550-2783-11-36
9. Bartholomae E, Incollingo A, Vizcaino M, Wharton C, Johnston CS. Mung Bean Protein Supplement Improves Muscular Strength in Healthy, Underactive Vegetarian Adults. Nutrients. 2019;11:doi:10.3390/nu11102423
10. Bridge A, Brown J, Snider H, Nasato M, Ward WE, Roy BD, et al. Greek Yogurt and 12 Weeks of Exercise Training on Strength, Muscle Thickness and Body Composition in Lean, Untrained, University-Aged Males. Frontiers in Nutrition. 2019;6:55. doi:10.3389/fnut.2019.00055
11. Brown EC, DiSilvestro RA, Babaknia A, Devor ST. Soy versus whey protein bars: Effects on exercise training impact on lean body mass and antioxidant status. Nutrition Journal. 2004;3:22. doi:10.1186/1475-2891-3-22
12. Burke DG, Chilibeck PD, Davidson KS, ow DG, Farthing J, Smith-Palmer T. The effect of whey protein supplementation with and without creatine monohydrate combined with resistance training on lean tissue mass and muscle strength. International journal of sport nutrition and exercise metabolism. 2001;11:349-64. doi:10.1123/ijsnem.11.3.349
13. Campbell BI, Aguilar D, Conlin L, Vargas A, Schoenfeld BJ, Corson A, et al. Effects of High Versus Low Protein Intake on Body Composition and Maximal Strength in Aspiring Female Physique Athletes Engaging in an 8-Week Resistance Training Program. International Journal of Sport Nutrition and Exercise Metabolism. 2018;28:580-5. doi:10.1123/ijsnem.2017-0389

14. Candow DG, Burke NC, Smith-Palmer T, Burke DG. Effect of whey and soy protein supplementation combined with resistance training in young adults. International Journal of Sport Nutrition and Exercise Metabolism. 2006;16:233-44.
15. Candow DG, Chilibeck PD, Facci M, Abeysekara S, Zello GA. Protein supplementation before and after resistance training in older men. European Journal of Applied Physiology. 2006;97:548-56.
16. Chale A, Cloutier GJ, Hau C, Phillips EM, Dallal GE, Fielding RA. Efficacy of Whey Protein Supplementation on Resistance ExerciseInduced Changes in Lean Mass, Muscle Strength, and Physical Function in Mobility-Limited Older Adults. Journals of Gerontology Series a-Biological Sciences and Medical Sciences. 2013;68:682-90.
17. Coburn JW, Housh DJ, Housh TJ, Malek MH, Beck TW, Cramer JT, et al. Effects of leucine and whey protein supplementation during eight weeks of unilateral resistance training. Journal of Strength and Conditioning Research. 2006;20:284-91. doi:10.1519/R-17925.1
18. Deibert P, Solleder F, Konig D, Vitolins MZ, Dickhuth HH, Gollhofer A, et al. Soy protein based supplementation supports metabolic effects of resistance training in previously untrained middle aged males. Aging Male. 2011;14:273-9. doi:10.3109/13685538.2011.565091
19. Dulac MC, Pion CH, Lemieux FC, Carvalho LP, El Hajj Boutros G, Belanger M, et al. EFFECTS OF SLOW VERSUS FAST-DIGESTED PROTEIN SUPPLEMENTATION COMBINED WITH MIXED POWER TRAINING ON MUSCLE FUNCTION AND FUNCTIONAL CAPACITIES IN OLDER MEN. The British journal of nutrition. 2020;1-40. doi:10.1017/S0007114520001932
20. Eliot KA, Knehans AW, Bemben DA, Witten MS, Carter J, Bemben MG. 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 & Aging. 2008;12:208-12. doi:10.1007/BF02982622
21. Erskine RM, Fletcher G, Hanson B, Foll, P. J. Whey Protein Does Not Enhance the Adaptations to Elbow Flexor Resistance Training. Medicine and Science in Sports and Exercise. 2012;44:1791-800.
22. Farup J, Rahbek SK, Vendelbo MH, Matzon A, Hindhede J, Bejder A, et al. Whey protein hydrolysate augments tendon and muscle hypertrophy independent of resistance exercise contraction mode. Scandinavian journal of medicine & science in sports. 2014;24:788-98. doi:10.1111/sms.12083
23. Gryson C, Ratel S, Rance M, Pen, o S, Bonhomme C, et al. Four-Month Course of Soluble Milk Proteins Interacts With Exercise to Improve Muscle Strength and Delay Fatigue in Elderly Participants. Journal of the American Medical Directors Association. 2014;15:958 e1-9. doi:10.1016/j.jamda.2014.09.011
24. Hartman JW, Tang JE, Wilkinson SB, Tarnopolsky MA, Lawrence RL, Fullerton AV, et al. Consumption of fat-free fluid milk after resistance exercise promotes greater lean mass accretion than does consumption of soy or carbohydrate in young, novice, male weightlifters. American Journal of Clinical Nutrition. 2007;86:373-81.
25. Haun CT, Mobley CB, Vann CG, Romero MA, Roberson PA, Mumford PW, et al. Soy protein supplementation is not androgenic or estrogenic in college-aged men when combined with resistance exercise training (vol 8, pg 11151, 2018). Scientific Reports. 2018;8:
26. Herda AA, Herda TJ, Costa PB, Ryan ED, Stout JR, Cramer JT. MUSCLE PERFORMANCE, SIZE, AND SAFETY RESPONSES AFTER EIGHT WEEKS OF RESISTANCE TRAINING AND PROTEIN SUPPLEMENTATION: A RANDOMIZED, DOUBLE-BLINDED, PLACEBO-CONTROLLED CLINICAL TRIAL. Journal of Strength and Conditioning Research. 2013;27:3091-100. doi:10.1519/JSC.0b013e31828c289f
27. Hida A, Hasegawa Y, Mekata Y, Usuda M, Masuda Y, Kawano H, et al. Effects of Egg White Protein Supplementation on Muscle Strength and Serum Free Amino Acid Concentrations. Nutrients. 2012;4:1504-17. doi:10.3390/nu4101504
28. Hoffman JR, Ratamess NA, Kang J, Falvo MJ, Faigenbaum AD. Effects of protein supplementation on muscular performance and resting hormonal changes in college football players. J Sports Sci Med. 2007;6:85-92.
29. Hoffman JR, Ratamess NA, Tranchina CP, Rashti SL, Kang J, Faigenbaum AD. Effect of Protein-Supplement Timing on Strength, Power, and Body-Composition Changes in Resistance-Trained

Men. International Journal of Sport Nutrition and Exercise Metabolism. 2009;19:172-85. doi:10.1123/ijsnem.19.2.172
30. Iglay HB, Apolzan JW, Gerrard DE, Eash JK, Anderson JC, Campbell WW. Moderately increased protein intake predominately from egg sources does not influence whole body, regional, or muscle composition responses to resistance training in older people. Journal of Nutrition Health & Aging. 2009;13:108-14.
31. Kerksick CM, Rasmussen CJ, Lancaster SL, Magu B, Smith P, Melton C, et al. The effects of protein and amino acid supplementation on performance and training adaptations during ten weeks of resistance training. Journal of Strength and Conditioning Research. 2006;20:643-53. doi:10.1519/R-17695.1
32. Kim HH, Kim YJ, Lee SY, Jeong DW, Lee JG, Yi YH, et al. Interactive effects of an isocaloric high-protein diet and resistance exercise on body composition, ghrelin, and metabolic and hormonal parameters in untrained young men: A randomized clinical trial. J Diabetes Investig. 2014;5:242-7. doi:10.1111/jdi.12148
33. Kim D, Park Y. Amount of Protein Required to Improve Muscle Mass in Older Adults. Nutrients. 2020;12:1700. doi:10.3390/nu12061700
34. Kirmse M, Oertzen-Hagemann V, de Marees M, Bloch W, Platen P. Prolonged collagen peptide supplementation and resistance exercise training affects body composition in recreationally active men. Nutrients. 2019;11:1154. doi:10.3390/nu11051154
35. Leenders M, Verdijk LB, Van der Hoeven L, Van Kranenburg J, Nilwik R, Wodzig W, et al. Protein Supplementation during Resistance-Type Exercise Training in the Elderly. Medicine and Science in Sports and Exercise. 2013;45:542-52. doi:10.1249/MSS.0b013e318272fcdb
36. Lockwood CM, Roberts MD, Dalbo VJ, Smith-Ryan AE, Kendall KL, Moon JR, et al. Effects of Hydrolyzed Whey versus Other Whey Protein Supplements on the Physiological Response to 8 Weeks of Resistance Exercise in College-Aged Males. Journal of the American College of Nutrition. 2017;36:16-27. doi:10.1080/07315724.2016.1140094
37. Maesta N, Nahas EAP, Nahas-Neto J, Orsatti FL, Fern, es CE, et al. Effects of soy protein and resistance exercise on body composition and blood lipids in postmenopausal women. Maturitas. 2007;56:350-8. doi:10.1016/j.maturitas.2006.10.001
38. Mobley CB, Haun CT, Roberson PA, Mumford PW, Romero MA, Kephart WC, et al. Effects of Whey, Soy or Leucine Supplementation with 12 Weeks of Resistance Training on Strength, Body Composition, and Skeletal Muscle and Adipose Tissue Histological Attributes in College-Aged Males. Nutrients. 2017;9:doi:10.3390/nu9090972
39. Mori H, Tokuda Y. Effect of whey protein supplementation after resistance exercise on the muscle mass and physical function of healthy older women: A randomized controlled trial. Geriatrics & Gerontology International. 2018;18:1398-404. doi:10.1111/ggi.13499
40. Nabuco HCG, Tomeleri CM, Sugihara P, Fern, es RR, Cavalcante EF, et al. Effects of Whey Protein Supplementation Pre- or Post-Resistance Training on Muscle Mass, Muscular Strength, and Functional Capacity in Pre-Conditioned Older Women: A Randomized Clinical Trial. Nutrients. 2018;10:doi:10.3390/nu10050563
41. Nabuco HCG, Tomeleri CM, Sugihara P, Fern, es RR, Cavalcante EF, et al. Effects of pre- or post-exercise whey protein supplementation on body fat and metabolic and inflammatory profile in pre-conditioned older women: A randomized, double-blind, placebo-controlled trial. Nutrition Metabolism and Cardiovascular Diseases. 2019;29:290-300. doi:10.1016/j.numecd.2018.11.007
42. Nabuco HCG, Tomeleri CM, Sugihara P, Fern, es RR, Cavalcante EF, et al. Effect of whey protein supplementation combined with resistance training on cellular health in pre-conditioned older women: A randomized, double-blind, placebo-controlled trial. Archives of Gerontology and Geriatrics. 2019;82:232-7. doi:10.1016/j.archger.2019.03.007
43. Naclerio F, Seijo-Bujia M, Larumbe-Zabala E, Earnest CP. Carbohydrates Alone or Mixing With Beef or Whey Protein Promote Similar Training Outcomes in Resistance Training Males: A Double-

Blind, Randomized Controlled Clinical Trial. International Journal of Sport Nutrition and Exercise Metabolism. 2017;27:408-20. doi:10.1123/ijsnem.2017-0003
44. Naclerio F, Larumbe-Zabala E, Ashrafi N, Seijo M, Nielsen B, Allgrove J, et al. Effects of protein-carbohydrate supplementation on immunity and resistance training outcomes: a double-blind, randomized, controlled clinical trial. European Journal of Applied Physiology. 2017;117:267-77. doi:10.1007/s00421-016-3520-x
45. Nahas PC, Rossato LT, Martins FM, Souza AP, de Branco FMS, Carneiro MAS, et al. Moderate Increase in Protein Intake Promotes a Small Additional Improvement in Functional Capacity, But Not in Muscle Strength and Lean Mass Quality, in Postmenopausal Women Following Resistance Exercise: A Randomized Clinical Trial. Nutrients. 2019;11:
46. Nakayama K, Saito Y, Sanbongi C, Murata K, Urashima T. Effects of low-dose milk protein supplementation following low-to-moderate intensity exercise training on muscle mass in healthy older adults: a randomized placebo-controlled trial. European Journal of Nutrition. 2020;doi:10.1007/s00394-020-02302-4
47. Negro M, oni M, Ottobrini S, Codrons E, Correale L, Buonocore D, et al. Protein Supplementation with Low Fat Meat after Resistance Training: Effects on Body Composition and Strength. Nutrients. 2014;6:3040-9. doi:10.3390/nu6083040
48. Norton C, Toomey C, McCormack WG, Francis P, Saunders J, Kerin E, et al. Protein Supplementation at Breakfast and Lunch for 24 Weeks beyond Habitual Intakes Increases Whole-Body Lean Tissue Mass in Healthy Older Adults. J Nutr. 2016;146:65-9. doi:10.3945/jn.115.219022
49. Obradovic J, Vukadinovic Jurisic M, Rakonjac D. The effects of leucine and whey protein supplementation with eight weeks of resistance training on strength and body composition. J Sports Med Phys Fitness. 2020;60:864-9. doi:10.23736/S0022-4707.20.09742-X
50. Oertzen-Hagemann V, Kirmse M, Eggers B, Pfeiffer K, Marcus K, de Marees M, et al. Effects of 12 Weeks of Hypertrophy Resistance Exercise Training Combined with Collagen Peptide Supplementation on the Skeletal Muscle Proteome in Recreationally Active Men. Nutrients. 2019;11:doi:10.3390/nu11051072
51. Ormsbee MJ, Willingham BD, Marchant T, Binkley TL, Specker BL, Vukovich MD. Protein Supplementation During a 6-Month Concurrent Training Program: Effect on Body Composition and Muscular Strength in Sedentary Individuals. International Journal of Sport Nutrition and Exercise Metabolism. 2018;28:619-28. doi:10.1123/ijsnem.2018-0036
52. Orsatti FL, Maesta N, de Oliveira EP, Nahas Neto J, Burini RC, Nunes PRP, et al. Adding Soy Protein to Milk Enhances the Effect of Resistance Training on Muscle Strength in Postmenopausal Women. Journal of dietary supplements. 2018;15:140-52. doi:10.1080/19390211.2017.1330794
53. Ottestad I, Lovstad AT, Gjevestad GO, Hamarsl, H., Benth JS, et al. Intake of a protein-enriched milk and effects on muscle mass and strength. A 12-week randomized placebo controlled trial among community-dwelling older adults. Journal of Nutrition Health & Aging. 2017;21:1160-9.
54. Ozan M, Buzdagli Y, Siktar E, Ucan I. The Effect of Protein and Carbohydrate Consumption during 10-Week Strength Training on Maximal Strength and Body Composition. International Journal of Applied Exercise Physiology. 2020;9:161-71.
55. Paoli A, Pacelli QF, Neri M, Toniolo L, Cancellara P, Canato M, et al. Protein supplementation increases postexercise plasma myostatin concentration after 8 weeks of resistance training in young physically active subjects. Journal of medicinal food. 2015;18:137-43. doi:10.1089/jmf.2014.0004
56. Pihoker AA, Peterjohn AM, Trexler ET, Hirsch KR, Blue MNM, Anderson KC, et al. The effects of nutrient timing on training adaptations in resistance-trained females. Journal of Science and Medicine in Sport. 2019;22:472-7. doi:10.1016/j.jsams.2018.09.236
57. Planella-Farrugia C, Comas F, Sabater-Masdeu M, Moreno M, Moreno-Navarrete JM, Rovira O, et al. Circulating Irisin and Myostatin as Markers of Muscle Strength and Physical Condition in Elderly Subjects. Frontiers in Physiology. 2019;10:

58. Rankin JW, Goldman LP, Puglisi MJ, Nickols-Richardson SM, Earthman CP, Gwazdauskas FC. Effect of post-exercise supplement consumption on adaptations to resistance training. Journal of the American College of Nutrition. 2004;23:322-30. doi:10.1080/07315724.2004.10719375
59. Reidy PT, Borack MS, Markofski MM, Dickinson JM, Deer RR, Husaini SH, et al. Protein Supplementation Has Minimal Effects on Muscle Adaptations during Resistance Exercise Training in Young Men: A Double-Blind Randomized Clinical Trial. Journal of Nutrition. 2016;146:1660-9.
60. Rossato LT, Nahas PC, de Branco FMS, Martins FM, Souza AP, Carneiro MAS, et al. Higher Protein Intake Does Not Improve Lean Mass Gain When Compared with RDA Recommendation in Postmenopausal Women Following Resistance Exercise Protocol: A Randomized Clinical Trial. Nutrients. 2017;9:
61. Rozenek R, Ward P, Long S, Garhammer J. Effects of high-calorie supplements on body composition and muscular strength following resistance training. J Sports Med Phys Fitness. 2002;42:340-7.
62. Sharp MH, Lowery RP, Shields KA, Lane JR, Gray JL, Partl JM, et al. THE EFFECTS OF BEEF, CHICKEN, OR WHEY PROTEIN AFTER WORKOUT ON BODY COMPOSITION AND MUSCLE PERFORMANCE. Journal of Strength and Conditioning Research. 2018;32:2233-42. doi:10.1519/JSC.0000000000001936
63. Snijders T, Res PT, Smeets JSJ, van Vliet S, van Kranenburg J, Maase K, et al. Protein Ingestion before Sleep Increases Muscle Mass and Strength Gains during Prolonged Resistance-Type Exercise Training in Healthy Young Men. Journal of Nutrition. 2015;145:1178-84.
64. Spillane M, Willoughby DS. Daily Overfeeding from Protein and/or Carbohydrate Supplementation for Eight Weeks in Conjunction with Resistance Training Does not Improve Body Composition and Muscle Strength or Increase Markers Indicative of Muscle Protein Synthesis and Myogenesis in Resistance-Trained Males. Journal of Sports Science and Medicine. 2016;15:17-25.
65. Taylor LW, Wilborn C, Roberts MD, White A, Dugan K. Eight weeks of pre- and postexercise whey protein supplementation increases lean body mass and improves performance in Division III collegiate female basketball players. Applied Physiology Nutrition and Metabolism. 2016;41:249-54. doi:10.1139/apnm-2015-0463
66. van Dongen EJI, Haveman-Nies A, Doets EL, Dorhout BG, de Groot L. Effectiveness of a Diet and Resistance Exercise Intervention on Muscle Health in Older Adults: ProMuscle in Practice. Journal of the American Medical Directors Association. 2020;21:1065-+. doi:10.1016/j.jamda.2019.11.026
67. Vangsoe MT, Joergensen MS, Heckmann LHL, Hansen M. Effects of Insect Protein Supplementation during Resistance Training on Changes in Muscle Mass and Strength in Young Men. Nutrients. 2018;10:doi:10.3390/nu10030335
68. Verdijk LB, Jonkers RAM, Gleeson BG, Beelen M, Meijer K, Savelberg H, et al. Protein supplementation before and after exercise does not further augment skeletal muscle hypertrophy after resistance training in elderly men. American Journal of Clinical Nutrition. 2009;89:608-16.
69. Volek JS, Volk BM, Gomez AL, Kunces LJ, Kupchak BR, Freidenreich DJ, et al. Whey Protein Supplementation During Resistance Training Augments Lean Body Mass. Journal of the American College of Nutrition. 2013;32:122-35. doi:10.1080/07315724.2013.793580
70. Walker TB, Smith J, Herrera M, Lebegue B, Pinchak A, Fischer J. The Influence of 8 Weeks of Whey-Protein and Leucine Supplementation on Physical and Cognitive Performance. International Journal of Sport Nutrition and Exercise Metabolism. 2010;20:409-17. doi:10.1123/ijsnem.20.5.409
71. Watanabe K, Holobar A, Mita Y, Kouzaki M, Ogawa M, Akima H, et al. Effect of Resistance Training and Fish Protein Intake on Motor Unit Firing Pattern and Motor Function of Elderly. Frontiers in Physiology. 2018;9:1733. doi:10.3389/fphys.2018.01733
72. Weisgarber KD, Candow DG, Vogt ESM. Whey Protein Before and During Resistance Exercise Has No Effect on Muscle Mass and Strength in Untrained Young Adults. International Journal of Sport Nutrition and Exercise Metabolism. 2012;22:463-9.

73. Willoughby DS, Stout JR, Wilborn CD. Effects of resistance training and protein plus amino acid supplementation on muscle anabolism, mass, and strength. Amino Acids. 2007;32:467-77. doi:10.1007/s00726-006-0398-7
74. Zhu K, Kerr DA, Meng XQ, Devine A, Solah V, Binns CW, et al. Two-Year Whey Protein Supplementation Did Not Enhance Muscle Mass and Physical Function in Well-Nourished Healthy Older Postmenopausal Women. Journal of Nutrition. 2015;145:2520-6.