SUPPLEMENATL MATERIAL

References of studies not included in the systemic review and meta-analysis

- **Not a randomized controlled trial (n=5)**

Borsheim E, Bui QU, Tissier S, Kobayashi H, Ferrando AA, Wolfe RR. Effect of amino acid supplementation on muscle mass, strength and physical function in elderly. Clin Nutr. 2008;27:189–195.

Casperson SL, Sheffield-Moore M, Hewlings SJ, Paddon-Jones D. Leucine supplementation chronically improves muscle protein synthesis in older adults consuming the RDA for protein. Clin Nutr. 2012 Aug;31(4):512-9.

Katsanos CS, Kobayashi H, Sheffield-Moore M, Aarsland A, Wolfe RR. Aging is associated with diminished accretion of muscle proteins after the ingestion of a small bolus of essential amino acids. Am J Clin Nutr. 2005;82:1065–1073.

Paddon-Jones D, Sheffield-Moore M, Zhang XJ, et al. Amino acid ingestion improves muscle protein synthesis in the young and elderly. Am J Physiol Endocrinol Metab. 2004; 286:E321–E328.

Rieu I, Balage M, Sornet C, etal. Leucine supplementation improves muscle protein synthesis in elderly men independently of hyperaminoacidaemia. J Physiol. 2006;575:305–315.

- **The population included non-elderly people (n=13)**

Churchward-Venne TA, Breen L, Di Donato DM, Hector AJ, Mitchell CJ, Moore DR, Stellingwerff T, Breuille D, Offord EA, Baker SK, Phillips SM. Leucine supplementation of a low-protein mixed macronutrient beverage enhances myofibrillar protein synthesis in young men: a double-blind, randomized trial. Am J Clin Nutr. 2014 Feb;99(2):276-86.

Churchward-Venne TA, Burd NA, Mitchell CJ, West DW, Philp A, Marcotte GR, Baker SK, Baar K, Phillips SM (2012) Supplementation of a suboptimal protein dose with leucine or essential amino acids: effects on myofibrillar protein synthesis at rest and following resistance exercise in men. J Physiol. 2012 Jun 1;590(Pt 11):2751-65.

Coburn JW, Housh DJ, Housh TJ, Malek MH, Beck TW, Cramer JT, Johnson GO, Donlin PE. Effects of leucine and whey protein supplementation during eight weeks of unilateral resistance training. J Strength Cond Res 2006;20:284–91.

Deldicque L, De Bock K, Maris M, Ramaekers M, Nielens H, Francaux M, Hespel P. Increased p70s6k phosphorylation during intake of a protein-carbohydrate drink following resistance exercise in the fasted state. Eur J Appl Physiol. 2010 Mar;108(4):791-800.

Dreyer HC, Drummond M, Pennings B, Fujita S, Glynn E, Chinkes D, Dhanani S, Volpi E, Rasmussen B. Leucine-enriched essential amino acid and carbohydrate ingestion following resistance exercise enhances mTOR signaling and protein synthesis in human muscle. Am J Physiol Endocrinol Metab 2008;294:E392–400.

Frestedt JL, Zenk JL, Kuskowski MA, Ward LS, Bastian ED. A whey-protein supplement increases fat loss and spares lean muscle in obese subjects: a randomized human clinical study. Nutr Metab (Lond). 2008 Mar 27;5:8. doi: 10.1186/1743-7075-5-8.

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. J Strength Cond Res. 2013 Nov;27(11):3091-100.

Mielke M, Housh TJ, Malek MH, Beck T, Schmidt RJ, Johnson GO, Housh DJ. The effects of whey protein and leucine supplementation on strength, muscular endurance, and body composition during resistance training. J Exerc Physiol Online 2009;12:39–50.

Pasiakos SM, McClung HL, McClung JP, Margolis LM, Andersen NE, Cloutier GJ, Pikosky MA, Rood JC, Fielding RA, Young AJ. Leucine-enriched essential amino

acid supplementation during moderate steady state exercise enhances postexercise muscle protein synthesis. Am J Clin Nutr. 2011 Sep;94(3):809-18.

Stock MS, Young JC, Golding LA, Kruskall LJ, Tandy RD, Conway-Klaassen JM, Beck TW. The effects of adding leucine to pre and postexercise carbohydrate beverages on acute muscle recovery from resistance training. J Strength Cond Res. 2010 Aug;24(8):2211-9.

Volek JS, Volk BM, Gómez AL, Kunces LJ, Kupchak BR, Freidenreich DJ, Aristizabal JC, Saenz C, Dunn-Lewis C, Ballard KD, Quann EE, Kawiecki DL, Flanagan SD, Comstock BA, Fragala MS, Earp JE, Fernandez ML, Bruno RS, Ptolemy AS, Kellogg MD, Maresh CM, Kraemer WJ. Whey protein supplementation during resistance training augments lean body mass. J Am Coll Nutr. 2013;32(2):122-35.

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. Int J Sport Nutr Exerc Metab 2010;20:409–17.

Zemel MB, Bruckbauer A. Effects of a leucine and pyridoxine-containing nutraceutical on body weight and composition in obese subjects. Diabetes Metab Syndr Obes. 2013 Aug 23;6:309-15.

- **Leucine dosing unclear (n=3)**

Chale A, Cloutier GJ, Hau C et al. (2013) Efficacy of whey protein supplementation on resistance exercise-induced changes in lean mass, muscle strength, and physical function in mobility-limited older adults. J Gerontol A Biol Sci Med Sci 68, 682-690.

Tieland M, Dirks ML, van der Zwaluw N et al. (2012) Protein supplementation increases muscle mass gain during prolonged resistance-type exercise training in frail elderly people: a randomized, double-blind, placebo-controlled trial. J Am Med Dir Assoc 13, 713-719.

Tieland M, van de Rest O, Dirks ML et al. (2012) Protein supplementation improves physical performance in frail elderly people: a randomized, double-blind, placebo-controlled trial. J Am Med Dir Assoc 13, 720-726.

- **Did not include an outcome of interest** (n=1)

Jonker R, Deutz NE, Erbland ML, Anderson PJ, Engelen MP. Hydrolyzed casein and whey protein meals comparably stimulate net whole-body protein synthesis in COPD patients with nutritional depletion without an additional effect of leucine co-ingestion. Clin Nutr. 2013 Jul 1. pii: S0261-5614(13)00186-6.

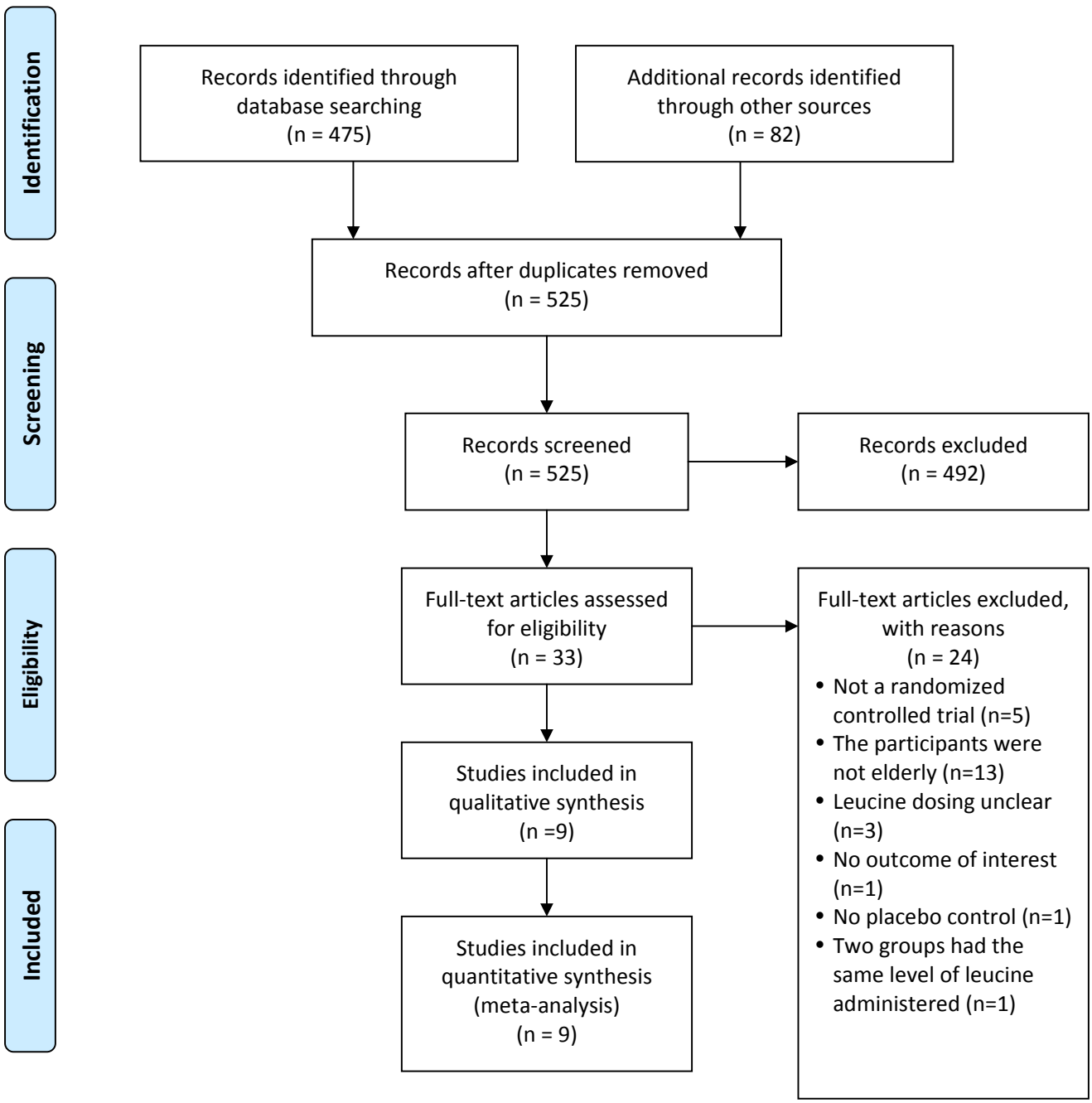
- **No placebo control (n=1)**:

Kim HK, Suzuki T, Saito K, Yoshida H, Kobayashi H, Kato H, Katayama M. Effects of exercise and amino acid supplementation on body composition and physical function in community-dwelling elderly Japanese sarcopenic women: a randomized controlled trial. J Am Geriatr Soc. 2012 Jan;60(1):16-23.

- **Treatment groups received the same amount of leucine** (n=1):

Volpi E, Kobayashi H, Sheffield-Moore M, Mittendorfer B, Wolfe RR. Essential amino acids are primarily responsible for the amino acid stimulation of muscle protein anabolism in healthy elderly adults. Am J Clin Nutr. 2003;78:250–258.

PRISMA 2009 Flow Diagram



From: Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). *P*referred *R*eporting *I*tems for *S*ystematic Reviews and *M*eta-*A*nalyses: The PRISMA Statement. PLoS Med 6(6): e1000097. doi:10.1371/journal.pmed1000097

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PRISMA 2009 Checklist

Section/topic	#	Checklist item	Reported on page #
TITLE			
Title	1	Identify the report as a systematic review, meta-analysis, or both.	1
ABSTRACT			
Structured summary	2	Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number.	2
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of what is already known.	3,4
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	4
METHODS			
Protocol and registration	5	Indicate if a review protocol exists, if and where it can be accessed (e.g., Web address), and, if available, provide registration information including registration number.	NA
Eligibility criteria	6	Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.	4,5
Information sources	7	Describe all information sources (e.g., databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.	4,5
Search	8	Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.	4,5
Study selection	9	State the process for selecting studies (i.e., screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).	4,5
Data collection process	10	Describe method of data extraction from reports (e.g., piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.	5
Data items	11	List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.	5
Risk of bias in individual studies	12	Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis.	5
Summary measures	13	State the principal summary measures (e.g., risk ratio, difference in means).	5
Synthesis of results	14	Describe the methods of handling data and combining results of studies, if done, including measures of consistency (e.g., I^2) for each meta-analysis.	5,6

PRISMA 2009 Checklist

Section/topic	#	Checklist item	Reported on page #
Risk of bias across studies	15	Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies).	5
Additional analyses	16	Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified.	5,6
RESULTS			
Study selection	17	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.	6, Figure 1
Study characteristics	18	For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations.	6, Table 1
Risk of bias within studies	19	Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12).	7, Figure 2
Results of individual studies	20	For all outcomes considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect estimates and confidence intervals, ideally with a forest plot.	8, 9, Table 1-2, Figure 4-6
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency.	8, 9, Figure 4-6
Risk of bias across studies	22	Present results of any assessment of risk of bias across studies (see Item 15).	7, Figure 3
Additional analysis	23	Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression [see Item 16]).	9, Figure 7
DISCUSSION			
Summary of evidence	24	Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers).	9, 10
Limitations	25	Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias).	10-14
Conclusions	26	Provide a general interpretation of the results in the context of other evidence, and implications for future research.	14
FUNDING			
Funding	27	Describe sources of funding for the systematic review and other support (e.g., supply of data); role of funders for the systematic review.	15

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