

1 *Supplementary Materials*

2 **Dietary Protein and Muscle in Aging People: The Potential 3 Role of the Gut Microbiome**

4 **Mary Ni Lochlainn ^{1,2,*}, Ruth C. E. Bowyer ¹ and Claire J. Steves ^{1,2},**

5 **1. Supplementary Table 1: Measures of Muscle Mass**

Muscle Mass	
Measure	Studies
DXA measurements [<i>lean body mass, leg lean mass, total lean mass, appendicular lean mass</i>]	(Ferrando <i>et al.</i> , 1996; Bunout <i>et al.</i> , 2001; Binder <i>et al.</i> , 2005; Cuthbertson <i>et al.</i> , 2005; Kortbein <i>et al.</i> , 2007; Houston <i>et al.</i> , 2008; Verhoeven, 2009; Dillon <i>et al.</i> , 2009; Kemmler <i>et al.</i> , 2010; Tieland <i>et al.</i> , 2012; Stout <i>et al.</i> , 2013; Deutz <i>et al.</i> , 2013; Gregorio <i>et al.</i> , 2014; Kerstetter <i>et al.</i> , 2015; Tanner <i>et al.</i> , 2015; Zhu <i>et al.</i> , 2015; Isanejad <i>et al.</i> , 2016; Norton <i>et al.</i> , 2016; Farsijani <i>et al.</i> , 2016; Mitchell <i>et al.</i> , 2017; Daly <i>et al.</i> , 2017; Dulac <i>et al.</i> , 2018)
Bioimpedance analysis	(Björkman, Finne-Soveri and Tilvis, 2012; Kim <i>et al.</i> , 2012; Okada <i>et al.</i> , 2013; Bo <i>et al.</i> , 2018)
Calf circumference	(F Landi <i>et al.</i> , 2017; Francesco Landi <i>et al.</i> , 2017; Marzetti <i>et al.</i> , 2018)
Mid-arm muscle circumference	(Ferrie <i>et al.</i> , 2016; F Landi <i>et al.</i> , 2017; Francesco Landi <i>et al.</i> , 2017)
Thigh muscle CSA at 50% femur length of the dominant leg (CT)	(Campbell <i>et al.</i> , 2001; Daly <i>et al.</i> , 2017; Mitchell <i>et al.</i> , 2017)
Quadriceps CSA (MRI)	(Verhoeven, 2009; Kilgour <i>et al.</i> , 2013; Drummond <i>et al.</i> , 2014)
Quadriceps volume (MRI)	(Greig <i>et al.</i> , 2011)
Forearm muscle thickness (US)	(Ferrie <i>et al.</i> , 2016)
Rectus femoris CSA (US)	(Ferrie <i>et al.</i> , 2016)
Calf muscle CSA (CT)	(Zhu <i>et al.</i> , 2015)
Free fat mass using labelled water	(Bonnefoy <i>et al.</i> , 2003)
Body weight minus fat mass (skin folds)	(Rydwik <i>et al.</i> , 2008)
Femur muscle density	(Daly <i>et al.</i> , 2015)
Type I and type II muscle fibre CSA (muscle biopsy)	(Cermak <i>et al.</i> , 2012)

7 Abbreviations: DXA: Dual-energy X-ray absorptiometry; CSA: cross sectional area; CT: Computed
8 Tomography; MRI: Magnetic Resonance Imaging; US: ultrasound

9

10

11

2. Supplementary Table 2: Measures of Muscle Strength

Muscle Strength	
Measure	Studies
Handgrip strength	(Scognamiglio <i>et al.</i> , 2005; Björkman, Finne-Soveri and Tilvis, 2012; Tieland <i>et al.</i> , 2012, 2017; Stout <i>et al.</i> , 2013; Gregorio <i>et al.</i> , 2014; Zhu <i>et al.</i> , 2015; Ferrie <i>et al.</i> , 2016; Isanejad <i>et al.</i> , 2016; F Landi <i>et al.</i> , 2017; Francesco Landi <i>et al.</i> , 2017; Bo <i>et al.</i> , 2018; Dulac <i>et al.</i> , 2018; Marzetti <i>et al.</i> , 2018)
Grip strength/body mass	(Isanejad <i>et al.</i> , 2016)
1-RM bicep curl	(Dillon <i>et al.</i> , 2009)
1-RM tricep extension	(Dillon <i>et al.</i> , 2009)
Knee extension	(Binder <i>et al.</i> , 2005; Kortebéin <i>et al.</i> , 2007; Björkman, Finne-Soveri and Tilvis, 2012; Kim <i>et al.</i> , 2012; Deutz <i>et al.</i> , 2013; Kilgour <i>et al.</i> , 2013; Drummond <i>et al.</i> , 2014; Isanejad <i>et al.</i> , 2016; Mitchell <i>et al.</i> , 2017)
1-RM leg extension strength	(Dillon <i>et al.</i> , 2009; Verhoeven, 2009; Tieland <i>et al.</i> , 2012, 2017)
Isometric leg extension	(Kemmler <i>et al.</i> , 2010)
Isometric contraction of knee extensors and/or plantar flexors	(Deutz <i>et al.</i> , 2013; Daly <i>et al.</i> , 2017; Guizelini <i>et al.</i> , 2018)
Quadriceps strength	(Bunout <i>et al.</i> , 2001)
1-RM leg press strength (kg)	(Verhoeven, 2009; Cermak <i>et al.</i> , 2012; Tieland <i>et al.</i> , 2012, 2017; Deutz <i>et al.</i> , 2013; Daly <i>et al.</i> , 2014; Dulac <i>et al.</i> , 2018)
3-RM leg press strength	(Daly <i>et al.</i> , 2014, 2015)
Leg press (unspecified)	(Rydwik <i>et al.</i> , 2008)
1-RM leg curl	(Dillon <i>et al.</i> , 2009)
Isokinetic leg strength (unspecified)	(Stout <i>et al.</i> , 2013)
Rapid force capacity	(Guizelini <i>et al.</i> , 2018)

12

Abbreviations: RM: repetition maximum

13

14

3. Supplementary Table 3: Measures of Physical Function

Physical Function	
Measure	Studies
Timed up and go	(Rydwik <i>et al.</i> , 2008; Kemmler <i>et al.</i> , 2010; Del Favero <i>et al.</i> , 2012; Deutz <i>et al.</i> , 2013; Stout <i>et al.</i> , 2013; Daly <i>et al.</i> , 2014, 2015; Zhu <i>et al.</i> , 2015; Mitchell <i>et al.</i> , 2017; Bo <i>et al.</i> , 2018)
Chair stand test	(Daly <i>et al.</i> , 2014, 2015; F Landi <i>et al.</i> , 2017; Francesco Landi <i>et al.</i> , 2017; Bo <i>et al.</i> , 2018)
Chair risers	(Bonnefoy <i>et al.</i> , 2003; Del Favero <i>et al.</i> , 2012; Isanejad <i>et al.</i> , 2016)
Floor transfer test	(Ferrando <i>et al.</i> , 2010)
Squat	(Isanejad <i>et al.</i> , 2016)
Gait speed (<i>measured using: 6m gait speed test, 10m walking speed, maximum walking speed, 4m walk, 12 minute walk, self-selected gait speed, 6 minute walking distance</i>)	(Bunout <i>et al.</i> , 2001; Bonnefoy <i>et al.</i> , 2003; Scognamiglio <i>et al.</i> , 2005; Drummond <i>et al.</i> , 2014; Daly <i>et al.</i> , 2015; Isanejad <i>et al.</i> , 2016; Kim <i>et al.</i> , 2016; Bo <i>et al.</i> , 2018)
Tandem walk for six-metres	(Isanejad <i>et al.</i> , 2016)
Incremental walking test	(Del Favero <i>et al.</i> , 2012)
Stair ascent test	(Ferrando <i>et al.</i> , 2010; Drummond <i>et al.</i> , 2014)
Stair descent test	(Ferrando <i>et al.</i> , 2010)
Time to limit of tolerance	(Del Favero <i>et al.</i> , 2012)
Balance (<i>measured using four square step test, one leg stance</i>)	(Daly <i>et al.</i> , 2014; Isanejad <i>et al.</i> , 2016)
Short physical performance battery (<i>includes: chair risers, 4m walk [gait speed] and ability to stand for ten seconds with feet in 3 different positions [balance]</i>)	(Tieland <i>et al.</i> , 2012; Deutz <i>et al.</i> , 2013; Gregorio <i>et al.</i> , 2014; Isanejad <i>et al.</i> , 2016; Mitchell <i>et al.</i> , 2017; Dulac <i>et al.</i> , 2018)
Senior Fitness Test (<i>includes: chair stand test, arm curl test, 2 minute step test, chair sit-and-reach test, back scratch test, eight-foot up-and-go test</i>)	(Szcześniak <i>et al.</i> , 2014)
Modified Physical Performance Test (<i>nine item test</i>)	(Drummond <i>et al.</i> , 2014; Gregorio <i>et al.</i> , 2014)
Physical activity scale for the elderly questionnaire	(Drummond <i>et al.</i> , 2014; Gregorio <i>et al.</i> , 2014)
Fatigue score [Chalder scale]	(Ferrie <i>et al.</i> , 2016)

15

Abbreviations: m: metres

16

17 **4. Supplementary Table 4: Animal Models**

Animal Model	References
Mice	(Faith <i>et al.</i> , 2011; Kar <i>et al.</i> , 2017; van Dijk <i>et al.</i> , 2017)
Rats	(Le Leu <i>et al.</i> , 2007; Marzani <i>et al.</i> , 2008; An <i>et al.</i> , 2014; Lee, Han and Yim, 2015)
Pigs	(Nunan, Sandercock and Brodie, 2010; Rist <i>et al.</i> , 2014; Zhou <i>et al.</i> , 2015, 2016; Lei <i>et al.</i> , 2018)
Golden Syrian hamsters	(Butteiger <i>et al.</i> , 2016)
Dogs	(Li <i>et al.</i> , 2017)
American pikas	(Kohl <i>et al.</i> , 2018)
Salmon	(Gajardo <i>et al.</i> , 2017)
Rainbow trout	(Rimoldi <i>et al.</i> , 2018)
Chicks	(Nakashima <i>et al.</i> , 2005)
Killifish	(Smith <i>et al.</i> , 2017)

18

19

20

21 **5. Supplementary Table 5: Measures of Dietary Protein Used**

Dietary Measure	References
Three-day food record	(Tieland <i>et al.</i> , 2012; Zhu <i>et al.</i> , 2015; Isanejad <i>et al.</i> , 2016; Beaumont <i>et al.</i> , 2017; Cardon-Thomas <i>et al.</i> , 2017; Mitchell <i>et al.</i> , 2017; Dulac <i>et al.</i> , 2018)
Food Frequency Questionnaire	(Ferrando <i>et al.</i> , 1996; Houston <i>et al.</i> , 2008; Wu <i>et al.</i> , 2011; Welch <i>et al.</i> , 2016)
The Healthy Food Diversity Index	(Claesson <i>et al.</i> , 2012; De Filippis <i>et al.</i> , 2016)
Four-day food record	(Gregorio <i>et al.</i> , 2014)
11-unit dietary score	(De Filippis <i>et al.</i> , 2016)
Seven-day weighed food diary	(De Filippis <i>et al.</i> , 2016)
'Recall' questionnaire	(Wu <i>et al.</i> , 2011)
Individual questionnaires, unique to that study	(F Landi <i>et al.</i> , 2017)
Telephone-facilitated 24-h dietary recalls	(Daly <i>et al.</i> , 2014; Farsijani <i>et al.</i> , 2016)
Diet History Questionnaire II ²	(David <i>et al.</i> , 2014)

22

23

24 **References**

- 25 An, C. *et al.* (2014) 'Caecal fermentation, putrefaction and microbiotas in rats fed milk casein, soy protein or fish
26 meal', *Applied Microbiology and Biotechnology*, 98(6), pp. 2779–2787. doi: 10.1007/s00253-013-5271-5.
- 27 Beaumont, M. *et al.* (2017) 'Quantity and source of dietary protein influence metabolite production by gut
28 microbiota and rectal mucosa gene expression: A randomized, parallel, double-blind trial in overweight
29 humans', *American Journal of Clinical Nutrition*, 106(4), pp. 1005–1019. doi: 10.3945/ajcn.117.158816.
- 30 Binder, E. F. *et al.* (2005) 'Effects of progressive resistance training on body composition in frail older adults:
31 Results of a randomized, controlled trial', *Journals of Gerontology - Series A Biological Sciences and Medical Sciences*,
32 60(11), pp. 1425–1431. doi: 10.1093/gerona/60.11.1425.
- 33 Björkman, M. P., Finne-Soveri, H. and Tilvis, R. S. (2012) 'Whey protein supplementation in nursing home
34 residents. A randomized controlled trial.', *Eur. Geriatr. Med*, 3, pp. 161–166. Available at: <http://www.em-consulte.com/en/article/728030>.
- 36 Bo, Y. *et al.* (2018) 'A high whey protein, vitamin D and E supplement preserves muscle mass, strength, and
37 quality of life in sarcopenic older adults: a double-blind randomized controlled trial', *Clinical Nutrition*.
38 European Society for Clinical Nutrition and Metabolism. doi: 10.1016/j.clnu.2017.12.020.
- 39 Bonnfoy, M. *et al.* (2003) 'The effects of exercise and protein-energy supplements on body composition and
40 muscle function in frail elderly individuals: a long-term controlled randomised study', *British Journal of
41 Nutrition*, 89(05), p. 731. doi: 10.1079/BJN2003836.
- 42 Bunout, D. *et al.* (2001) 'The impact of nutritional supplementation and resistance training on the health
43 functioning of free-living Chilean elders: results of 18 months of follow-up.', *The Journal of nutrition*, 131(9), p.
44 2441S–6S.
- 45 Butteiger, D. N. *et al.* (2016) 'Soy Protein Compared with Milk Protein in a Western Diet Increases Gut Microbial
46 Diversity and Reduces Serum Lipids in Golden Syrian Hamsters', *The Journal of Nutrition*, 146(4), pp. 697–705.
47 doi: 10.3945/jn.115.224196.The.
- 48 Campbell, W. W. *et al.* (2001) 'The recommended dietary allowance for protein may not be adequate for older
49 people to maintain skeletal muscle', *Journals of Gerontology - Series A Biological Sciences and Medical Sciences*, 56(6),
50 pp. M373–M380. doi: 10.1093/gerona/56.6.M373.
- 51 Cardon-Thomas, D. K. *et al.* (2017) 'Dietary protein in older adults: Adequate daily intake but potential for
52 improved distribution', *Nutrients*, 9(3), pp. 1–10. doi: 10.3390/nu9030184.
- 53 Cermak, N. M. *et al.* (2012) 'Protein supplementation augments the adaptive response of skeletal muscle to
54 resistance type exercise training a meta analysis', *American Journal of Clinical Nutrition*, 96(February), pp. 1454–
55 1464. doi: 10.3945/ajcn.112.037556.INTRODUCTION.
- 56 Claesson, M. J. *et al.* (2012) 'Gut microbiota composition correlates with diet and health in the elderly', *Nature*,
57 488(7410), pp. 178–184. doi: 10.1038/nature11319.

- 58 Cuthbertson, D. *et al.* (2005) 'Anabolic signaling deficits underlie amino acid resistance of wasting, aging
59 muscle', *The FASEB Journal*, 19(3), pp. 422–424. doi: 10.1096/fj.04-2640fje.
- 60 Daly, R. . *et al.* (2017) 'DOES LEAN RED MEAT ENHANCE THE EFFECTS OF EXERCISE ON MUSCLE
61 HEALTH AND FUNCTION IN THE ELDERLY?', *Innovation in Aging*, 1(March), pp. 3–4.
- 62 Daly, R. M. *et al.* (2014) 'Protein-enriched diet, with the use of lean red meat, combined with progressive
63 resistance training enhances lean tissue mass and muscle strength and reduces circulating IL-6 concentrations
64 in elderly women: a cluster randomized controlled trial', *The American Journal of Clinical Nutrition*, (99), pp. 899–
65 910. doi: 10.3945/jn.113.064154.Sarcopenia.
- 66 Daly, R. M. *et al.* (2015) 'The effects of a protein enriched diet with lean red meat combined with a multi-modal
67 exercise program on muscle and cognitive health and function in older adults: Study protocol for a randomised
68 controlled trial', *Trials*. Trials, 16(1), pp. 1–16. doi: 10.1186/s13063-015-0884-x.
- 69 David, L. A. *et al.* (2014) 'Diet rapidly and reproducibly alters the human gut microbiome', *Nature*, 505(7484),
70 pp. 559–563. doi: 10.1038/nature12820.Diet.
- 71 Deutz, N. E. P. *et al.* (2013) 'Effect of β-hydroxy-β-methylbutyrate (HMB) on lean body mass during 10 days of
72 bed rest in older adults', *Clinical Nutrition*. Elsevier Ltd, 32(5), pp. 704–712. doi: 10.1016/j.clnu.2013.02.011.
- 73 van Dijk, M. *et al.* (2017) 'Sarcopenia in older mice is characterized by a decreased anabolic response to a protein
74 meal', *Archives of Gerontology and Geriatrics*. Elsevier Ireland Ltd, 69, pp. 134–143. doi:
75 10.1016/j.archger.2016.11.014.
- 76 Dillon, E. L. *et al.* (2009) 'Amino acid supplementation increases lean body mass, basal muscle protein synthesis,
77 and insulin-like growth factor-I expression in older women', *Journal of Clinical Endocrinology and Metabolism*,
78 94(5), pp. 1630–1637. doi: 10.1210/jc.2008-1564.
- 79 Drummond, M. J. *et al.* (2014) 'Downregulation of E3 ubiquitin ligases and mitophagy-related genes in skeletal
80 muscle of physically inactive, frail older women: A cross-sectional comparison', *Journals of Gerontology - Series
81 A Biological Sciences and Medical Sciences*, 69(8), pp. 1040–1048. doi: 10.1093/gerona/glu004.
- 82 Dulac, M. C. *et al.* (2018) 'Differences in muscle adaptation to a 12-week mixed power training in elderly men,
83 depending on usual protein intake', *Experimental Gerontology*. Elsevier Inc. doi: 10.1016/j.exger.2018.02.001.
- 84 Faith, J. J. *et al.* (2011) 'Predicting a human gut microbiota's response to diet in gnotobiotic mice', *Science*,
85 333(6038), pp. 101–104. doi: 10.1126/science.1206025.Predicting.
- 86 Farsijani, S. *et al.* (2016) 'Relation between mealtime distribution of protein intake and lean mass loss in free-
87 living older adults of the NuAge study', *The American Journal of Clinical Nutrition*. Oxford University Press,
88 104(3), pp. 694–703. doi: 10.3945/ajcn.116.130716.
- 89 Del Favero, S. *et al.* (2012) 'Beta-alanine (Carnosyn™) supplementation in elderly subjects (60-80 years): Effects
90 on muscle carnosine content and physical capacity', *Amino Acids*, 43(1), pp. 49–56. doi: 10.1007/s00726-011-1190-

- 91 x.
- 92 Ferrando, A. *et al.* (2010) 'EAA supplementation to increase nitrogen intake improves muscle function during
93 bed rest in the elderly.', *Clin Nutr*, 29(1), pp. 18–23.
- 94 Ferrando, A. A. *et al.* (1996) 'Prolonged bed rest decreases skeletal muscle and whole body protein synthesis.',
95 *The American journal of physiology*. American Physiological Society Bethesda, MD , 270(4 Pt 1), pp. E627-33. doi:
96 10.1152/ajpendo.1996.270.4.E627.
- 97 Ferrie, S. *et al.* (2016) 'Protein Requirements in the Critically Ill: A Randomized Controlled Trial Using Parenteral
98 Nutrition', *Journal of Parenteral and Enteral Nutrition*, 40(6), pp. 795–805. doi: 10.1177/0148607115618449.
- 99 De Filippis, F. *et al.* (2016) 'High-level adherence to a Mediterranean diet beneficially impacts the gut microbiota
100 and associated metabolome', *Gut*, 65(11), pp. 1812–1821. doi: 10.1136/gutjnl-2015-309957.
- 101 Gajardo, K. *et al.* (2017) 'Alternative Protein Sources in the Diet Modulate Microbiota and Functionality in the
102 Distal Intestine of Atlantic Salmon (*Salmo salar*)', *Applied and Environmental Microbiology*, 83(5), pp. 1–16. doi:
103 10.1128/AEM.02615-16.
- 104 Gregorio, L. *et al.* (2014) 'ADEQUATE DIETARY PROTEIN IS ASSOCIATED WITH BETTER PHYSICAL
105 PERFORMANCE AMONG POST-MENOPAUSAL WOMEN 60–90 YEARS', *J Nutr Health Aging*, 18(2), pp. 155–
106 160. doi: 10.1007/s12603-013-0391-2.ADEQUATE.
- 107 Greig, C. A. *et al.* (2011) 'Blunting of adaptive responses to resistance exercise training in women over 75 y',
108 *Experimental Gerontology*. Pergamon, 46(11), pp. 884–890. doi: 10.1016/J.EXGER.2011.07.010.
- 109 Guizelini, P. C. *et al.* (2018) 'Effect of resistance training on muscle strength and rate of force development in
110 healthy older adults: A systematic review and meta-analysis', *Experimental Gerontology*. Elsevier, 102(June 2017),
111 pp. 51–58. doi: 10.1016/j.exger.2017.11.020.
- 112 Houston, D. K. *et al.* (2008) 'Dietary protein intake is associated with lean mass change in older , community-
113 dwelling adults : the Health , Aging , and Body Composition (Health ABC) Study', *Am J Clin Nutr* 2008;87:150–
114 5., 87, pp. 150–155.
- 115 Isanejad, M. *et al.* (2016) 'Dietary protein intake is associated with better physical function and muscle strength
116 among elderly women', *British Journal of Nutrition*, 115(7), pp. 1281–1291. doi: 10.1017/S000711451600012X.
- 117 Kar, S. K. *et al.* (2017) 'Dietary protein sources differentially affect microbiota, mTOR activity and transcription
118 of mTOR signaling pathways in the small intestine', *PLoS ONE*, 12(11), pp. 1–19. doi:
119 10.1371/journal.pone.0188282.
- 120 Kemmler, W. *et al.* (2010) 'Exercise, body composition, and functional ability: a randomized controlled trial.',
121 *Am J Prev Med*, 38(3), pp. 279–87. Available at: <https://www.ncbi.nlm.nih.gov/pubmed/20171529>.
- 122 Kerstetter, J. E. *et al.* (2015) 'The effect of a whey protein supplement on bone mass in older Caucasian adults',
123 *Journal of Clinical Endocrinology and Metabolism*, 100(6), pp. 2214–2222. doi: 10.1210/jc.2014-3792.

- 124 Kilgour, A. H. M. *et al.* (2013) 'Increased skeletal muscle 11 β HSD1 mRNA is associated with lower muscle
125 strength in ageing', *PLoS ONE*, 8(12), pp. 8–13. doi: 10.1371/journal.pone.0084057.
- 126 Kim, H. K. *et al.* (2012) 'Effects of Exercise and Amino Acid Supplementation on Body Composition and Physical
127 Function in Community-Dwelling Elderly Japanese Sarcopenic Women: A Randomized Controlled Trial',
128 *Journal of the American Geriatrics Society*. Wiley/Blackwell (10.1111/j.1532-
129 5415.2011.03776.x).
- 130 Kim, J. E. *et al.* (2016) 'Effects of dietary protein intake on body composition changes after weight loss in older
131 adults: A systematic review and meta-analysis', *Nutrition Reviews*, 74(3), pp. 210–224. doi:
132 10.1093/nutrit/nuv065.
- 133 Kohl, K. D. *et al.* (2018) 'Gut microbial communities of American pikas (*Ochotona princeps*): Evidence for
134 phylosymbiosis and adaptations to novel diets', *Journal of Animal Ecology*, 87(2), pp. 323–330. doi: 10.1111/1365-
135 2656.12692.
- 136 Kortebein, P. *et al.* (2007) 'Effect of 10 Days of Bed Rest on Skeletal Muscle in Healthy Older Adults', *JAMA*.
137 American Medical Association, 297(16), p. 1769. doi: 10.1001/jama.297.16.1772-b.
- 138 Landi, F. *et al.* (2017) 'Age-Related Variations of Muscle Mass, Strength, and Physical Performance in
139 Community-Dwellers: Results From the Milan EXPO Survey', *Journal of the American Medical Directors
140 Association*. Elsevier Inc., 18(1), p. 88.e17-88.e24. doi: 10.1016/j.jamda.2016.10.007.
- 141 Landi, F. *et al.* (2017) 'ANIMAL-DERIVED PROTEIN CONSUMPTION IS ASSOCIATED WITH MUSCLE
142 MASS AND STRENGTH IN COMMUNITY-DWELLERS: RESULTS FROM THE MILAN EXPO SURVEY', *J
143 Nutr Health Aging*, 21(9), pp. 1050–1056.
- 144 Lee, S.-M., Han, H. W. and Yim, S. Y. (2015) 'Beneficial effects of soy milk and fiber on high cholesterol diet-
145 induced alteration of gut microbiota and inflammatory gene expression in rats', *Food Funct*. Royal Society of
146 Chemistry, 6(2), pp. 492–500. doi: 10.1039/C4FO00731J.
- 147 Lei, X. J. *et al.* (2018) 'Effects of levan-type fructan on growth performance, nutrient digestibility, diarrhoea
148 scores, faecal shedding of total lactic acid bacteria and coliform bacteria, and faecal gas emission in weaning
149 pigs', *Journal of the Science of Food and Agriculture*, 98(4), pp. 1539–1544. doi: 10.1002/jsfa.8625.
- 150 Le Leu, R. K. *et al.* (2007) 'Effect of dietary resistant starch and protein on colonic fermentation and intestinal
151 tumourigenesis in rats', *Carcinogenesis*, 28(2), pp. 240–245. doi: 10.1093/carcin/bgl245.
- 152 Li, Q. *et al.* (2017) 'Effects of the dietary protein and carbohydrate ratio on gut microbiomes in dogs of different
153 body conditions', *mBio*, 8(1), pp. 1–14. doi: 10.1128/mBio.01703-16.
- 154 Marzani, B. *et al.* (2008) 'Antioxidant Supplementation Restores Defective Leucine Stimulation of Protein
155 Synthesis in Skeletal Muscle from Old Rats', *J. Nutr*, 138, pp. 2205–2211. doi: 10.3945/jn.108.094029.
- 156 Marzetti, E. *et al.* (2018) 'Age-related changes of skeletal muscle mass and strength among Italian and Taiwanese

- 157 older people: Results from the Milan EXPO 2015 survey and the I-Lan Longitudinal Aging Study', *Experimental*
 158 *Gerontology*. Elsevier, 102(October 2017), pp. 76–80. doi: 10.1016/j.exger.2017.12.008.
- 159 Mitchell, C. J. et al. (2017) 'The effects of dietary protein intake on appendicular lean mass and muscle function
 160 in elderly men: a 10-wk randomized controlled trial', *The American journal of clinical nutrition*, 106(6), pp. 1375–
 161 1383. doi: 10.3945/ajcn.117.160325.
- 162 Nakashima, K. et al. (2005) 'Leucine suppresses myofibrillar proteolysis by down-regulating ubiquitin-
 163 proteasome pathway in chick skeletal muscles', *Biochemical and Biophysical Research Communications*, 336(2), pp.
 164 660–666. doi: 10.1016/j.bbrc.2005.08.138.
- 165 Norton, C. et al. (2016) 'Protein Supplementation at Breakfast and Lunch for 24 Weeks beyond Habitual Intakes
 166 Increases Whole-Body Lean Tissue Mass in Healthy Older Adults', *The Journal of Nutrition*. Oxford University
 167 Press, 146(1), pp. 65–69. doi: 10.3945/jn.115.219022.
- 168 Nunan, D., Sandercock, G. R. and Brodie, D. A. (2010) 'A quantitative systematic review of normal values for
 169 short-term heart rate variability in healthy adults', *Pacing Clin Electrophysiol*. 2010/07/29, 33(11), pp. 1407–1417.
 170 doi: 10.1111/j.1540-8159.2010.02841.x.
- 171 Okada, H. C. et al. (2013) 'Facial changes caused by smoking: A comparison between smoking and nonsmoking
 172 identical twins', *Plastic and Reconstructive Surgery*, 132(5), pp. 1085–1092. doi: 10.1097/PRS.0b013e3182a4c20a.
- 173 Rimoldi, S. et al. (2018) 'Next generation sequencing for gut microbiome characterization in rainbow trout
 174 (*Oncorhynchus mykiss*) fed animal by-product meals as an alternative to fishmeal protein sources', pp. 1–29.
 175 doi: 10.1371/journal.pone.0193652.
- 176 Rist, V. T. S. et al. (2014) 'Effect of dietary protein supply originating from soybean meal orcasein on the
 177 intestinal microbiota of piglets', *Anaerobe*. Elsevier Ltd, 25, pp. 72–79. doi: 10.1016/j.anaerobe.2013.10.003.
- 178 Rydwik, E. et al. (2008) 'Effects of a physical and nutritional intervention program for frail elderly people over
 179 age 75. A randomized controlled pilot treatment trial', *Aging Clin Exp Res*, 20(2), pp. 159–70.
- 180 Scognamiglio, R. et al. (2005) 'Oral amino acids in elderly subjects: effect on myocardial function and walking
 181 capacity.', *Gerontology*, 51(5), pp. 302–8. Available at: <https://www.ncbi.nlm.nih.gov/pubmed/16110231>.
- 182 Smith, P. et al. (2017) 'Regulation of Life Span by the Gut Microbiota in The Short-Lived African Turquoise
 183 Killifish', *bioRxiv*. Cold Spring Harbor Laboratory, p. 120980. doi: 10.1101/120980.
- 184 Stout, J. R. et al. (2013) 'Effect of calcium β-hydroxy-β-methylbutyrate (CaHMB) with and without resistance
 185 training in men and women 65+ yrs: A randomized, double-blind pilot trial', *Experimental Gerontology*. The
 186 Authors, 48(11), pp. 1303–1310. doi: 10.1016/j.exger.2013.08.007.
- 187 Szczęśniak, D. et al. (2014) 'Anserine and carnosine supplementation in the elderly: Effects on cognitive
 188 functioning and physical capacity.', *Archives of Gerontology and Geriatrics*, 59(2), pp. 485–490. Available at:
 189 <https://www.sciencedirect.com/science/article/pii/S0167494314000545?via%3Dihub>.

- 190 Tanner, R. E. *et al.* (2015) 'Age-related differences in lean mass, protein synthesis and skeletal muscle markers
191 of proteolysis after bed rest and exercise rehabilitation.', *The Journal of physiology*. Wiley-Blackwell, 593(18), pp.
192 4259–73. doi: 10.1111/jp270699.
- 193 Tieland, M. *et al.* (2012) 'Protein Supplementation Improves Physical Performance in Frail Elderly People: A
194 Randomized, Double-Blind, Placebo-Controlled Trial', *Journal of the American Medical Directors Association*.
195 Elsevier Ltd, 13(8), pp. 720–726. doi: 10.1016/j.jamda.2012.07.005.
- 196 Tieland, M. *et al.* (2017) 'The impact of dietary protein or amino acid supplementation on muscle mass and
197 strength in elderly people: Individual participant data and meta-analysis of RCT's', *The journal of nutrition, health
198 & aging*, 21(9), pp. 994–1001. doi: 10.1007/s12603-017-0896-1.
- 199 Verhoeven, S. (2009) 'Long-term leucine supplementation does not increase muscle mass or strength in healthy
200 elderly men', *The American journal ...*, (April), pp. 1468–1475. doi: 10.3945/ajcn.2008.26668.1.
- 201 Welch, A. A. *et al.* (2016) 'Dietary Magnesium Is Positively Associated With Skeletal Muscle Power and Indices
202 of Muscle Mass and May Attenuate the Association Between Circulating C-Reactive Protein and Muscle Mass
203 in Women', *J Bone Miner Res*, 31(2), pp. 317–325. doi: 10.1002/jbmr.2692.
- 204 Wu, G. D. *et al.* (2011) 'Linking Long-Term Dietary Patterns with Gut Microbial Enterotypes', *Science*, 334(6052),
205 pp. 105–108. doi: 10.1126/science.1208344.Linking.
- 206 Zhou, L. *et al.* (2016) 'Effects of the dietary protein level on the microbial composition and metabolomic profile
207 in the hindgut of the pig', *Anaerobe*. Elsevier Ltd, 38, pp. 61–69. doi: 10.1016/j.anaerobe.2015.12.009.
- 208 Zhou, P. *et al.* (2015) 'Effects of dietary crude protein levels and cysteamine supplementation on protein
209 synthetic and degradative signaling in skeletal muscle of finishing pigs', *PLoS ONE*, 10(9), pp. 1–16. doi:
210 10.1371/journal.pone.0139393.
- 211 Zhu, K. *et al.* (2015) 'Two-Year Whey Protein Supplementation Did Not Enhance Muscle Mass and Physical
212 Function in Well-Nourished Healthy Older Postmenopausal Women', *Journal of Nutrition*, 145(11), pp. 2520–
213 2526. doi: 10.3945/jn.115.218297.
- 214