

Postprint of a Meta-Analysis on the Effects of Protein Supplementation on Muscle Mass, Strength, and Physical Function in Frail Older Adults

Authors: Guo Yinning, Miao Xueyi, Jiang Xiaoman, Xu Ting, Xu Qin

Date: 2022-10-13T00:00:00+00:00

Abstract

Background: Frailty increases the risk of health-related adverse outcomes in older adults, and protein supplementation may be an effective approach to improve frailty; however, current evidence regarding the effects of protein supplementation on frailty remains controversial.

Objective: To systematically evaluate the effects of protein supplementation on muscle mass, strength, and physical function in frail older adults.

Methods: A comprehensive systematic search was conducted across CNKI, Wanfang, VIP, PubMed, Web of Science, Embase, Cochrane Library, CINAHL, and Medline databases for randomized controlled trials published from inception to June 2022 investigating the effects of protein supplementation on muscle mass, strength, and physical function in frail older adults. Literature was screened, and studies meeting the inclusion and exclusion criteria underwent quality assessment and data extraction. Meta-analysis was performed using Review Manager 5.4.

Results: A total of 12 studies involving 833 older adults were included, with 422 in the protein supplementation group and 411 in the control group. Meta-analysis results showed that protein supplementation could improve walking speed (MD=0.03, 95%CI 0.00~0.06, P=0.05), but demonstrated no significant advantages in improving muscle mass (appendicular lean mass), muscle strength (hand grip strength), other physical functions [balance ability, timed up and go test, Short Physical Performance Battery (SPPB)], or frailty scores, with pooled effects showing no statistical significance (P>0.05).

Conclusion: Protein supplementation can improve walking speed in frail older adults, but shows no significant advantages in improving their muscle mass,

strength, or other physical functions. Further large-scale studies with longer intervention durations are needed for validation.

Full Text

Preamble

Effects of Protein Supplementation on Muscle Mass, Strength, and Physical Function in Frail Older Adults: A Meta-Analysis

GUO Yinning, MIAO Xueyi, JIANG Xiaoman, XU Ting, XU Qin*

School of Nursing, Nanjing Medical University, Nanjing 211166, China

*Corresponding author: Xu Qin, Professor, Dean of the School of Nursing; E-mail: qinxu@njmu.edu.cn

Funding: National Natural Science Foundation of China “Exploring the Frailty Trajectory and Designing Intervention Programs for Gastric Cancer Survivors from the Perspective of Health Ecology” (Project No.: 82073407)

Abstract

Background: Frailty increases the risk of negative health-related outcomes in older adults, and protein supplementation may be an effective approach to improve frailty, but current evidence regarding its effects remains controversial. **Objective:** To systematically evaluate the effects of protein supplementation on muscle mass, strength, and physical function in frail older adults. **Methods:** We conducted a comprehensive systematic search of randomized controlled trials examining the effects of protein supplementation on muscle mass, strength, and physical function in frail older adults across CNKI, Wanfang, VIP, PubMed, Web of Science, Embase, Cochrane Library, CINAHL, and Medline databases from inception to June 2022. Literature screening, quality assessment, and data extraction were performed for eligible studies, with meta-analysis conducted using Review Manager 5.4. **Results:** Twelve articles comprising 833 older adults were included (422 in protein supplementation group, 411 in control group). Meta-analysis results showed that protein supplementation improved walking speed (MD=0.03, 95%CI 0.00~0.06, P=0.05), but demonstrated no significant advantages in improving muscle mass (appendicular lean mass), muscle strength (grip strength), other physical functions [balance ability, timed up and go test, Short Physical Performance Battery (SPPB)], or frailty scores, with no statistically significant pooled effects (P>0.05). **Conclusion:** Protein supplementation can improve walking speed in frail older adults but shows no clear advantage in improving muscle mass, strength, or other physical functions. Further large-scale studies with longer intervention periods are needed for validation.

Keywords: frailty; protein; elderly; muscle mass; muscle strength; physical function; meta-analysis

Introduction

The “Healthy China 2030” Planning Outline emphasizes strengthening health services for key populations to ensure comprehensive, lifelong health coverage for all citizens, with particular attention to “addressing health issues among key groups such as women, children, the elderly, people with disabilities, and low-income populations” [1]. In recent years, China’s modernization has accelerated, and population aging has become increasingly severe, with 264 million people aged 60 and above accounting for approximately 18.7% of the total population [2]. This substantial demographic shift makes addressing elderly health issues a central priority for achieving Healthy China 2030.

Due to aging, older adults frequently experience health problems related to frailty, including weight loss, fatigue, and leg weakness. Frailty is a multidimensional geriatric syndrome characterized by multisystem physiological dysregulation, reduced physiological reserve, increased vulnerability, and diminished stress resistance [3]. Surveys indicate that the prevalence of frailty among community-dwelling older adults aged 60 and above in China is approximately 10% [4], while among hospitalized older adults it reaches about 30% [5]. Frailty not only increases the risk of negative health outcomes such as falls, functional decline, disability, and mortality, but also intensifies family and social caregiving burdens and healthcare resource consumption [6]. Therefore, actively exploring interventions for geriatric frailty is urgently needed.

Nutrition is considered a key factor influencing frailty in older adults [7], with protein being the primary underlying driver of the nutrition-frailty relationship due to its effects on muscle mass, strength, and function [8]. Protein supplementation may therefore be beneficial for frail older adults. However, current evidence on the effects of protein supplementation in this population remains inconsistent. Some studies [9, 10] have found that protein intake can prevent and improve frailty while enhancing physical function, whereas others [11, 12] have reported no effect or even that excessive protein intake may increase renal burden and produce adverse effects. Given these conflicting findings, this meta-analysis aims to comprehensively and objectively examine whether protein supplementation has positive effects on frail older adults and determine the required duration of supplementation to produce effects, thereby providing evidence for preventive care and clinical practice.

Methods

1.1 Literature Search

We conducted a comprehensive systematic search of relevant literature from database inception to June 2022 across CNKI, Wanfang, VIP, PubMed, Web of Science, The Cochrane Library, CINAHL, and Medline. MeSH terms and free-text keywords were combined to construct search strategies for both Chinese and English databases. Chinese search terms included “衰弱/衰弱综合征” (frailty/frailty syndrome), “老人/老年/老年人” (elderly/older adults), and “蛋白质/氨基酸” (protein/amino acid). English search terms included “frail elderly/frailty/frailty syndrome/frail*“, “aged/older/elderly”, and “protein/amino acid/leucine/HMB/beta-hydroxy-beta-methyl butyrate” .

1.2 Inclusion and Exclusion Criteria

Inclusion criteria: (1) Randomized controlled trials published in Chinese or English; (2) Participants aged \geq 60 years diagnosed as frail or pre-frail using established frailty assessment criteria; (3) Intervention: protein supplementation in the experimental group versus placebo or no intervention (maintaining habitual diet) in the control group; (4) Outcome measures: at least one of the following variables—muscle mass (appendicular lean mass), muscle strength (grip strength), physical function [gait speed, balance test, timed up and go test, Short Physical Performance Battery (SPPB)], or frailty score.

Exclusion criteria: (1) Duplicate publications; (2) Studies with unextractable data or incomplete statistical reporting, including unpublished full texts, conference proceedings, commentaries, and letters; (3) Participants with acute illness, cancer, or severe chronic diseases; (4) Participants engaged in weight-loss diets or other concurrent interventions; (5) Poor-quality articles rated as grade C.

1.3 Literature Screening and Data Extraction

Two researchers independently screened titles and abstracts according to the predefined inclusion and exclusion criteria, excluded duplicates and obviously irrelevant studies, then performed full-text reading to identify eligible studies. Data extraction was conducted independently and blindly, with content including author, year, study design, participant characteristics, intervention details, and outcome measures (appendicular lean mass, grip strength, gait speed, balance test, timed up and go test, SPPB score, frailty score). Disagreements were resolved through discussion with a third researcher.

1.4 Quality Assessment

Two researchers independently assessed the quality of the 12 included randomized controlled trials using the Cochrane Handbook version 5.1.0 risk of bias assessment tool [13]. The evaluation covered seven domains: random sequence generation, allocation concealment, blinding of participants and personnel, blinding

of outcome assessors, completeness of outcome data, selective reporting, and other sources of bias. Studies with low risk of bias in all domains were rated as quality grade “A” ; those with low risk in some domains as grade “B” ; and those with high risk in all domains as grade “C” .

1.5 Data Processing

Outcome measures were extracted and synthesized as mean (M) \pm standard deviation (SD). A few studies [11, 14] originally reported outcomes as mean with 95% confidence interval (CI), requiring conversion using the following formula [15] to calculate SD, where n represents sample size and the denominator t-value can be obtained in Excel by entering “=tinv(probability, degrees of freedom)” with probability=1-0.95 (for 95% CI) and degrees of freedom=n-1.

$$SD = \sqrt{n} \times \frac{(95\%CI_{upper} - 95\%CI_{lower})}{2t}$$

1.6 Statistical Analysis

Meta-analysis was performed using RevMan 5.4 software. Since all extracted outcomes were continuous variables, mean difference (MD) was used for data pooling when outcome measures employed identical methods and units; standardized mean difference (SMD) was used when measurement methods or units differed. All outcome analyses calculated 95% CI. Heterogeneity among studies was assessed using I^2 test ($\alpha=0.1$). If $I^2 < 50\%$ and $P \leq 0.1$, *indicating low heterogeneity, a fixed-effects model was adopted; otherwise, a random-effects model was used after conducting sensitivity analysis*

Funnel plots were generated for outcome indicators with high heterogeneity and sufficient studies ($n \geq 10$) to assess publication bias.

Results

2.1 General Characteristics of Included Studies

The initial search yielded 5,257 relevant articles. After preliminary screening, 44 articles were included for full-text review, ultimately resulting in 12 included articles. The literature screening process is illustrated in [Figure 1: see original paper].

2.2 Basic Characteristics of Included Studies

Twelve articles were included, with Caldo-Silva et al. publishing two related articles using identical data [16, 17], which were combined for descriptive purposes. All 12 articles were published in English, comprising 833 older adults (422 in experimental group, 411 in control group). Five articles [10, 12, 18-20] originated

from Asia, six [9, 14, 16, 17, 21, 22] from Europe, and one [11] from South America. Ten articles [9-12, 14, 16, 17, 19, 21, 22] used Fried frailty criteria or its revised version, one [20] used Frailty Working Group criteria, and one [18] used the Korean Cardiovascular Health Study (CHS) frailty criteria. All studies described intervention content and duration with clearly defined outcome measures. Detailed characteristics are presented in .

2.3 Quality Assessment of Included Studies

Quality assessment revealed three articles [18-20] rated as grade A and nine articles [9-12, 14, 16, 17, 21, 22] as grade B. Methodological quality assessment results are shown in .

2.4 Meta-Analysis Results

2.4.1 Muscle Mass (Appendicular Lean Mass) Three articles [9, 14, 21] reported effects of 12-week protein supplementation on appendicular lean mass, with significant heterogeneity ($I^2=77\%$). After removing Dirks et al. [9] using the one-study removal method, heterogeneity became zero ($I^2=0$), and a fixed-effects model was applied. Results indicated no significant effect of 12-week protein intervention on appendicular lean mass (MD=0.03, 95% CI -0.36 to 0.41, $P=0.89$). One article (with three subgroups) [11] compared 16-week protein intervention effects, with no heterogeneity ($I^2=0$), using a fixed-effects model. Results also showed no effect (MD=-0.56, 95% CI -1.59 to 0.47, $P=0.29$). Three articles [9, 14, 21] reported 24-week protein supplementation effects with substantial heterogeneity ($I^2=92\%$). After removing Tieland (1) et al. [21], heterogeneity disappeared ($I^2=0$), and a fixed-effects model revealed that 24-week protein intervention significantly increased appendicular lean mass (MD=1.99, 95% CI 1.19 to 2.79, $P<0.00001$). Overall, protein supplementation showed a non-significant positive trend for increasing appendicular lean mass in frail older adults (MD=0.3, 95% CI -0.03 to 0.63, $P=0.07$). See [Figure 2: see original paper].

2.4.2 Grip Strength Eleven articles [9-12, 14, 16, 18-22] reported intervention effects of 6, 12, 16, or 24-week protein supplementation on grip strength. Kang et al. [10] reported grip strength changes separately for males and females. Due to different measurement tools, standardized mean difference (SMD) was used. Significant heterogeneity existed ($I^2=48\%$, $P=0.008$), which reduced to zero ($I^2=0$, $P=0.89$) after removing Dirks et al. [9] (12 and 24-week data) and Tieland (1) et al. [21] (12-week data). The fixed-effects model showed no significant improvement in grip strength across different durations (SMD=-0.11, 95% CI -0.23 to 0.01, $P=0.07$). See [Figure 3: see original paper].

2.4.3 Physical Function

2.4.3.1 Walking Speed (m/s) Two articles [10, 18] reported the relationship between 6-week protein supplementation and gait speed, with no heterogeneity ($I^2=0$, $P=0.99$), using a fixed-effects model. Results showed no significant effect (MD=0.00, 95% CI -0.05 to 0.06, $P=0.88$). Four articles [10, 12, 18, 20] compared 12-week protein supplementation effects, also with no heterogeneity ($I^2=0$, $P=0.44$). Meta-analysis revealed that 12-week protein supplementation significantly improved gait speed (MD=0.04, 95% CI 0.00 to 0.08, $P=0.03$). Overall, protein supplementation improved walking speed in frail older adults (MD=0.03, 95% CI 0.00 to 0.06, $P=0.05$). See [Figure 4: see original paper].

2.4.3.2 Balance Test (sec) Three articles [10, 12, 18] reported effects of 6-week and 12-week protein supplementation on balance ability. Kang et al. [10] presented results as ordinal data that could not be pooled with the other two studies. No heterogeneity was found ($I^2=0$, $P=0.52$), and the fixed-effects model showed no significant difference in balance ability improvement (MD=0.04, 95% CI -0.02 to 0.09, $P=0.19$). Kang et al. [10] also found no effect of 12-week protein supplementation on balance. See [Figure 5: see original paper].

2.4.3.3 Timed Up and Go Test (sec) Four articles [11, 18-20] compared effects of 6, 12, or 16-week protein supplementation on timed up and go test performance. No heterogeneity existed ($I^2=0$, $P=0.56$), and the fixed-effects model indicated no significant effect in reducing timed up and go duration (MD=0.09, 95% CI -0.22 to 0.40, $P=0.57$). See [Figure 6: see original paper].

2.4.3.4 SPPB Score Five articles [9, 12, 14, 18, 20] examined the relationship between 6, 12, or 24-week protein supplementation and SPPB scores. Due to potential differences in SPPB assessment standards, SMD was used. Heterogeneity was substantial ($I^2=48\%$, $P=0.04$) but decreased after removing Dirks et al. [9] 24-week data ($I^2=4\%$, $P=0.40$). The fixed-effects model showed no significant improvement in SPPB scores across durations (SMD=-0.02, 95% CI -0.17 to 0.13, $P=0.82$). See [Figure 7: see original paper].

2.4.4 Frailty Score Two articles [17, 18] reported the relationship between protein supplementation and overall frailty scores. Due to different assessment tools, SMD was used with no heterogeneity ($I^2=0$, $P=0.98$) and a fixed-effects model. Meta-analysis showed no significant difference in frailty score reduction with 6, 12, or 16-week protein supplementation compared to control (SMD=-0.02, 95% CI -0.23 to 0.19, $P=0.85$). See [Figure 8: see original paper].

2.5 Publication Bias

Funnel plots for grip strength and SPPB score showed generally symmetrical scatter plots, indicating no obvious publication bias and suggesting credible meta-analysis results. See [Figure 9: see original paper] and [Figure 10: see original paper].

Discussion

Inadequate nutritional intake is considered a crucial component of frailty [23], potentially triggering muscle atrophy and dysfunction in older adults. Research indicates that muscle mass maintenance depends on the dynamic balance between muscle protein synthesis and breakdown [24]. With aging, muscle protein synthesis decreases while catabolism increases, disrupting this balance and causing degenerative changes and systemic loss of muscle mass and function, thereby inducing sarcopenia [25], which represents the core pathological basis of frailty [7]. Protein serves as the primary regulator of muscle protein metabolism, stimulating synthesis and inhibiting breakdown [26] to rebalance these processes and maintain muscle mass and function, thus preventing or improving frailty. Therefore, protein supplementation may be beneficial for frail older adults. Based on this rationale, we conducted this meta-analysis to quantify the effects of protein supplementation on muscle mass, strength, and physical function in frail older adults.

Regarding muscle mass and strength, this study found that protein supplementation had no significant effect on preserving appendicular lean mass (muscle mass) or grip strength (muscle strength) in frail older adults, consistent with Tieland et al. [27]. This may relate to intervention duration, as included studies ranged from 6 to 24 weeks. Although the effect on muscle mass and strength loss remains unclear, our results show a trend toward larger differences between protein and control groups with longer supplementation, particularly for lean mass, which showed significant differences at 24 weeks. This suggests protein supplementation may influence muscle mass and strength in frail older adults, but current intervention durations may be insufficient to detect measurable changes. Research indicates that older adults lose approximately 1% muscle mass annually (about 0.6 kg) [28], translating to an expected 0.07-0.28 kg loss in control groups during 6-24 week interventions. Given measurement errors from dual-energy X-ray absorptiometry (DXA) and bioelectrical impedance analysis (BIA), differences between groups may be too small to detect. Extending supplementation to 3 years would yield an estimated 1.8 kg muscle loss in control groups, creating larger between-group differences more likely detectable by DXA and BIA. Houston et al. [29] confirmed this, showing that higher protein intake prevents age-related muscle mass loss over 3 years. Therefore, investigating long-term protein supplementation effects on muscle mass and strength in frail older adults may be more meaningful.

For physical function, this study demonstrated that protein supplementation significantly improved gait speed but showed no clear advantages in balance ability (balance test), walking/reaction capacity (timed up and go test), or overall physical performance (SPPB score). Gait speed is a component of Fried frailty criteria. While 6-week supplementation showed no significant effect, 12-week supplementation significantly improved gait speed, consistent with Matsuzawa

et al. [30]. This may relate to protein supplementation promoting whole-body and limb muscle mass. Although earlier meta-analyses showed no significant effects on muscle mass and strength, this is not contradictory, as 12 weeks may be insufficient to manifest overt changes in these parameters while still producing subtle improvements that enhance gait speed. Additionally, since leg muscles constitute the largest and most powerful muscle group [31], they may benefit first from protein supplementation, explaining why protein supplementation improves gait speed but not balance, walking/reaction capacity, or overall physical function. Future studies with longer interventions should verify effects on other physical function domains.

For overall impact on frailty, this study used frailty scores as a comprehensive measure. Meta-analysis showed no significant effect on frailty score reduction, though effect sizes increased with longer supplementation duration, suggesting that sufficiently prolonged protein supplementation may yield significant frailty score reductions and symptom improvement, requiring validation through longer interventions.

Several factors may have influenced our results, including frailty assessment tools, protein supplementation quantity, and type. Most studies used the frailty phenotype (FP), but two employed FWG and CHS criteria, and participants included both pre-frail and frail states, creating variability in frailty severity that may affect outcomes. Future research should standardize assessment tools to enhance comparability. Protein intake quantity also matters. Included studies provided 6-30g protein supplementation. Rahi et al. [32] found high protein intake (\$ 1.0 g/kg/d) reduced frailty prevalence by 59%, while Ruth Teh et al. [33] found low protein intake increased frailty risk and impaired physical function. Coelho-Junior et al. [8] observed lower frailty prevalence at mean intake of 1.28 g/kg/d. Thus, different supplementation amounts may yield different results, though optimal protein intake for frail older adults remains undefined.

Furthermore, our study included various protein types (soy, whey, protein beverages, powder, essential amino acids [EAA], branched-chain amino acids [BCAA]). Research shows plant-based proteins like soy and wheat elicit lower muscle protein synthesis (MPS) responses than equivalent animal proteins [34]. Yang et al. [35] reported that neither 20g nor 40g soy protein stimulated MPS in older adults at rest, whereas whey protein significantly increased MPS. This anabolic response difference stems primarily from two factors [36]: (1) digestibility—animal-based proteins show 90% digestibility versus typically <50% for plant proteins; (2) EAA content, particularly BCAAs (isoleucine, leucine, valine), with animal proteins containing higher EAA and BCAA levels [37], thus producing greater MPS. We speculate that protein type may have influenced our results, warranting further investigation.

This study has several limitations: (1) Lack of prospective large-sample studies; (2) Exclusion of conference proceedings, dissertations, and non-Chinese/English literature, potentially introducing publication bias; (3) Variability in participants' frailty severity and intervention duration, content, and frequency, which

may affect pooled results; (4) Some meta-analyses included only 2 studies, requiring cautious interpretation.

In summary, protein supplementation significantly improves gait speed in frail older adults, but its effects on muscle mass, strength, and other physical functions remain unclear. Future large-scale studies with more rigorous, standardized interventions and longer durations are needed to explore optimal protein supplementation patterns (duration, type, and amount) for frailty management.

Author Contributions

Guo Yinning: manuscript writing, data collection and analysis; Guo Yinning and Miao Xueyi: statistical analysis, results interpretation; Jiang Xiaoman and Xu Ting: manuscript revision; Xu Qin: quality control, final approval, overall responsibility, and supervision.

Conflict of Interest: The authors declare no conflicts of interest.

References

- [1] Xinhua News Agency. CPC Central Committee and State Council Issue the “Healthy China 2030” Planning Outline [EB/OL]. (2016-10-25). [2022-07-12]. http://www.gov.cn/xinwen/2016-10/25/content_{5124174}.htm
- [2] National Bureau of Statistics. Communique of the Seventh National Population Census (No. 5) [EB/OL]. (2021-05-11). [2022-07-10]. http://www.stats.gov.cn/zjtj/zdtjgz/zgrkpc/dqcrkpc/ggl/202105/t20210519_{1817698}.html
- [3] Clegg A, Young J, Iliffe S, et al. Frailty in elderly people [J]. *Lancet*, 2013, 381(9868): 752-62. DOI: 10.1016/s0140-6736(12)62167-9
- [4] Ma L, Tang Z, Zhang L, et al. Prevalence of Frailty and Associated Factors in the Community-Dwelling Population of China [J]. *Journal of the American Geriatrics Society*, 2018, 66(3): 559-64. DOI: 10.1111/jgs.15214
- [5] Liang Y D, Zhang Y N, Li Y M, et al. Identification of Frailty and Its Risk Factors in Elderly Hospitalized Patients from Different Wards: A Cross-Sectional Study in China [J]. *Clinical interventions in aging*, 2019, 14: 2249-59. DOI: 10.2147/cia.S225149
- [6] Ma L N, Chen B. Research status and development trend of frailty syndrome in the elderly [J]. *Chinese Journal of Geriatrics*, 2020, 39: 369-72. DOI: 10.3760/cma.j.issn.0254-9026.2020.04.001
- [7] Ni Lochlainn M, Cox N J, Wilson T, et al. Nutrition and Frailty: Opportunities for Prevention and Treatment [J]. *Nutrients*, 2021, 13(7). DOI: 10.3390/nu13072349

- [8] Coelho-Junior H J, Marzetti E, Picca A, et al. Protein Intake and Frailty: A Matter of Quantity, Quality, and Timing [J]. *Nutrients*, 2020, 12(10). DOI: 10.3390/nu12102915
- [9] Dirks M L, Tieland M, Verdijk L B, et al. Protein Supplementation Augments Muscle Fiber Hypertrophy but Does Not Modulate Satellite Cell Content During Prolonged Resistance-Type Exercise Training in Frail Elderly [J]. *Journal of the American Medical Directors Association*, 2017, 18(7): 608-15. DOI: 10.1016/j.jamda.2017.02.006
- [10] Kang L, Gao Y, Liu X H, et al. Effects of whey protein nutritional supplement on muscle function among community-dwelling frail older people: A multicenter study in China [J]. *Archives of gerontology and geriatrics*, 2019, 83: 7-12. DOI: 10.1016/j.archger.2019.03.012
- [11] Roschel H, Hayashi A P, Fernandes A L, et al. Supplement-based nutritional strategies to tackle frailty: A multifactorial, double-blind, randomized placebo-controlled trial [J]. *Clinical nutrition (Edinburgh, Scotland)*, 2021, 40(8): 4849-58. DOI: 10.1016/j.clnu.2021.06.024
- [12] Peng L N, Cheng Y C, Yu P C, et al. Oral Nutritional Supplement with beta-hydroxy-beta-methylbutyrate (HMB) Improves Nutrition, Physical Performance and Ameliorates Intramuscular Adiposity in Pre-Frail Older Adults: A Randomized Controlled Trial [J]. *Journal of Nutrition Health & Aging*, 2021, 25(6): 767-73. DOI: 10.1007/s12603-021-1637-9
- [13] Higgins J, Green S. *Cochrane handbook for systematic reviews of interventions version 5.1.0*[EB/OL]. (2011-03). [2022-07-13]. <https://training.cochrane.org/handbook/archive/v5.1/>
- [14] Tieland M, Dirks M L, van der Zwaluw N, et al. Protein Supplementation Increases Muscle Mass Gain During Prolonged Resistance-Type Exercise Training in Frail Elderly People: A Randomized, Double-Blind, Placebo-Controlled Trial [J]. *Journal of the American Medical Directors Association*, 2012, 13(8): 713-9. DOI: 10.1016/j.jamda.2012.05.020
- [15] Liu G J, Wu T X. Correct application of statistical measures in evidence-based medicine [J]. *Chinese Journal of Tissue Engineering Research*, 2003, 7: 359-62,65. DOI: 10.3321/j.issn:1673-8225.2003.03.003
- [16] Caldo-Silva A, Furtado G E, Chupel M U, et al. Effect of a 40-weeks multi-component exercise program and branched chain amino acids supplementation on functional fitness and mental health in frail older persons [J]. *Experimental gerontology*, 2021, 155: 111592. DOI: 10.1016/j.exger.2021.111592
- [17] Caldo-Silva A, Furtado G E, Chupel M U, et al. Effect of Training-Detraining Phases of Multicomponent Exercises and BCAA Supplementation on Inflammatory Markers and Albumin Levels in Frail Older Persons [J]. *Nutrients*, 2021, 13(4). DOI: 10.3390/nu13041106
- [18] Park Y, Choi J-E, Hwang H-S. Protein supplementation improves muscle mass and physical performance in undernourished prefrail and frail elderly sub-

- jects: a randomized, double-blind, placebo-controlled trial [J]. *American Journal of Clinical Nutrition*, 2018, 108(5): 1026-33. DOI: 10.1093/ajcn/nqy214
- [19] Kim H, Suzuki T, Kim M, et al. Effects of exercise and milk fat globule membrane (MFGM) supplementation on body composition, physical function, and hematological parameters in community-dwelling frail Japanese women: a randomized double blind, placebo-controlled, follow-up trial [J]. *PloS one*, 2015, 10(2): e0116256. DOI: 10.1371/journal.pone.0116256
- [20] Kim C-O, Lee K-R. Preventive effect of protein-energy supplementation on the functional decline of frail older adults with low socioeconomic status: a community-based randomized controlled study [J]. *The journals of gerontology Series A, Biological sciences and medical sciences*, 2013, 68(3): 309-16. DOI: 10.1093/gerona/gls167
- [21] Tieland M, van de Rest O, Dirks M L, et al. Protein Supplementation Improves Physical Performance in Frail Elderly People: A Randomized, Double-Blind, Placebo-Controlled Trial [J]. *Journal of the American Medical Directors Association*, 2012, 13(8): 720-6. DOI: 10.1016/j.jamda.2012.07.005
- [22] Biesek S, Wojciechowski A S, Melo J, et al. Effects of Exergames and Protein Supplementation on Body Composition and Musculoskeletal Function of Prefrail Community-Dwelling Older Women: A Randomized, Controlled Clinical Trial [J]. *International journal of environmental research and public health*, 2021, 18(17). DOI: 10.3390/ijerph18179324
- [23] Cruz-Jentoft A J, Kiesswetter E, Drey M, et al. Nutrition, frailty, and sarcopenia [J]. *Aging clinical and experimental research*, 2017, 29(1): 43-8. DOI: 10.1007/s40520-016-0709-0
- [24] Breen L, Phillips S M. Skeletal muscle protein metabolism in the elderly: Interventions to counteract the ‘anabolic resistance’ of ageing [J]. *Nutr Metab (Lond)*, 2011, 8: 68. DOI: 10.1186/1743-7075-8-68
- [25] Yang L, Du J, Liu R Y, et al. Current status and prospects of the etiology research of sarcopenia [J]. *Chinese Journal of Osteoporosis*, 2020, 26: 1689-93. DOI: 10.3969/j.issn.1006-7108.2020.11.024
- [26] Stokes T, Hector A J, Morton R W, et al. Recent Perspectives Regarding the Role of Dietary Protein for the Promotion of Muscle Hypertrophy with Resistance Exercise Training [J]. *Nutrients*, 2018, 10(2). DOI: 10.3390/nu10020180
- [27] Tieland M, Franssen R, Dullemeijer C, et al. The Impact of Dietary Protein or Amino Acid Supplementation on Muscle Mass and Strength in Elderly People: Individual Participant Data and Meta-Analysis of RCT’ s [J]. *The journal of nutrition, health & aging*, 2017, 21(9): 994-1001. DOI: 10.1007/s12603-017-0896-1
- [28] Wilkinson D J, Piasecki M, Atherton P J. The age-related loss of skeletal muscle mass and function: Measurement and physiology of muscle fibre atrophy

and muscle fibre loss in humans [J]. *Ageing research reviews*, 2018, 47: 123-32. DOI: 10.1016/j.arr.2018.07.005

[29] Houston D K, Nicklas B J, Ding J, et al. Dietary protein intake is associated with lean mass change in older, community-dwelling adults: the Health, Aging, and Body Composition (Health ABC) Study [J]. *The American journal of clinical nutrition*, 2008, 87(1): 150-5. DOI: 10.1093/ajcn/87.1.150

[30] Matsuzawa R, Yamamoto S, Suzuki Y, et al. The effects of amino acid/protein supplementation in patients undergoing hemodialysis: A systematic review and meta-analysis of randomized controlled trials [J]. *Clinical nutrition ESPEN*, 2021, 44: 114-21. DOI: 10.1016/j.clnesp.2021.04.027

[31] Zhao J. Research on the leg training of men' s 70 kg amateur bodybuilders in Shandong Province [D]. Shandong Sport University, 2020

[32] Rahi B, Colombet Z, Gonzalez-Colaço Harmand M, et al. Higher Protein but Not Energy Intake Is Associated With a Lower Prevalence of Frailty Among Community-Dwelling Older Adults in the French Three-City Cohort [J]. *Journal of the American Medical Directors Association*, 2016, 17(7): 672.e7-.e11. DOI: 10.1016/j.jamda.2016.05.005

[33] Teh R, Mendonça N, Muru-Lanning M, et al. Dietary Protein Intake and Transition between Frailty States in Octogenarians Living in New Zealand [J]. *Nutrients*, 2021, 13(8). DOI: 10.3390/nu13082843

[34] Pinckaers P J M, Trommelen J, Snijders T, et al. The Anabolic Response to Plant-Based Protein Ingestion [J]. *Sports medicine (Auckland, NZ)*, 2021, 51(Suppl 1): 59-74. DOI: 10.1007/s40279-021-01540-8

[35] Yang Y, Churchward-Venne T A, Burd N A, et al. Myofibrillar protein synthesis following ingestion of soy protein isolate at rest and after resistance exercise in elderly men [J]. *Nutr Metab (Lond)*, 2012, 9(1): 57. DOI: 10.1186/1743-7075-9-57

[36] Berrazaga I, Micard V, Gueugneau M, et al. The Role of the Anabolic Properties of Plant- versus Animal-Based Protein Sources in Supporting Muscle Mass Maintenance: A Critical Review [J]. *Nutrients*, 2019, 11(8). DOI: 10.3390/nu11081825

[37] Gwin J A, Carbone J W, Rodriguez N R, et al. Physiological Limitations of Protein Foods Ounce Equivalents and the Underappreciated Role of Essential Amino Acid Density in Healthy Dietary Patterns [J]. *The Journal of nutrition*, 2021, 151(11): 3276-83. DOI: 10.1093/jn/nxab262

Note: Figure translations are in progress. See original paper for figures.

Source: ChinaXiv – Machine translation. Verify with original.