

## ADEQUATE DIETARY PROTEIN IS ASSOCIATED WITH BETTER PHYSICAL PERFORMANCE AMONG POST-MENOPAUSAL WOMEN 60-90 YEARS

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**Abstract:** *Objectives:* Sarcopenia, the involuntary loss of skeletal muscle with age, affects up to one-quarter of older adults. Evidence indicates a positive association between dietary protein intake and lean muscle mass and strength among older persons, but information on dietary protein's effect on physical performance in older adults has received less attention. *Design:* Cross-sectional observational analysis of the relationship of dietary protein on body composition and physical performance. *Setting:* Clinical research center. *Participants:* 387 healthy women aged 60 – 90 years (mean 72.7 ± 7.0 y). *Measurements:* Measures included body composition (fat-free mass, appendicular skeletal mass and fat mass) via dual x-ray absorptiometry (DXA), physical performance (Physical Performance Test [PPT] and Short Physical Performance Battery [SPPB]), handgrip strength, Physical Activity Scale in the Elderly (PASE), quality of life measure (SF-8), falls, fractures, nutrient and macromolecule intake (four-day food record). Independent samples t-tests determined mean differences between the above or below RDA protein groups. *Statistical Analysis:* Analysis of covariance was used to control for body mass index (BMI) between groups when assessing physical performance, physical activity and health-related quality of life. *Results:* The subjects consumed an average of 72.2 g protein/day representing 1.1 g protein/kg body weight/day. Subjects were categorized as below the recommended daily allowance (RDA) for protein (defined as less than 0.8 g protein/kg) or at or above the RDA (equal to or higher than 0.8 g protein/kg). Ninety-seven subjects (25%) were in the low protein group, and 290 (75%) were in the higher protein group. Women in the higher protein group had lower body mass, including fat and lean mass, and fat-to-lean ratio than those in the lower-protein group (p <0.001). Composite scores of upper and lower extremity strength were impaired in the group with low protein intake; SPPB score was 9.9±1.9 compared to 10.6±1.6 in those with higher protein intake and PPT was 19.8± 2.9 compared to 20.9± 2.1 in the low and higher protein groups, respectively. The results were attenuated by correction for BMI, but remained significant. The physical component of the SF-8 was also lower in the low protein group but did not remain significant when controlling for BMI. No significant differences were found in hand grip strength or reported physical activity. *Conclusion:* Healthy, older postmenopausal women consumed, on average, 1.1 g/kg/d protein, although 25% consumed less than the RDA. Those in the low protein group had higher body fat and fat-to-lean ratio than those who consumed the higher protein diet. Upper and lower extremity function was impaired in those who consumed a low protein diet compared to those with a higher protein intake. Protein intake should be considered when evaluating the multi-factorial loss of physical function in older women.

**Key words:** Protein, body composition, physical performance, frailty.

### Introduction

Sarcopenia, the degenerative and involuntary process of skeletal muscle loss, is characteristic of aging and may affect as many as one-quarter of older adults (1). It is commonly associated with other diagnoses related to aging such as type II diabetes, hypertension, obesity and osteoporosis. Sarcopenia is an important reason for the reduced strength and functional capacity of older adults. Sarcopenia leads to an increase incidence of falls and fractures and loss of independence among the elderly (2-4). Undernutrition is thought to promote skeletal muscle loss, while nutritional intervention may offer an opportunity to prevent or forestall sarcopenia.

Considerable evidence indicates a positive association between protein intake and lean muscle mass, strength and vitality among older persons (5-8). Borsheim et al. found that short-term protein supplementation improves lean body mass,

strength and physical performance (i.e. walking speed) in older subjects (5). Houston et al. reported that subjects in the highest quintile of protein intake lost approximately 40% less lean muscle mass over the course of three years than those in the lowest quintile of intake (6). Available data indicate that dietary protein supplementation will induce muscle anabolism, even in older adults (9-12). Munger et al. found that dietary protein intake and the incidence of hip fracture are inversely related (13), potentially through an impact on changes in muscle strength. Similarly, Tkatch et al. showed that oral protein supplementation given to elderly patients hospitalized after hip fractures resulted in shorter hospital stays and lower rates of complications and mortality than those on a control supplement (14).

These studies suggest high dietary protein and/or supplementation offer effective ways of preventing, delaying or

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slowing the progression of sarcopenia in older adults. However, the current protein Recommended Daily Allowance (RDA) for adults, 0.8 g protein/kg body weight/day, may be insufficient to adequately meet the metabolic and physiological needs of the elderly (15-17). Defining nutritional requirements of older adults is complicated by the impact of chronic diseases, multiple medications, and disabilities (18-21). Dietary protein declines with age, whether expressed as total g per day or as g per kg. Fulgoni estimated that the mean daily protein intake of US adults over age 71 was  $66 \pm 17$  g/d ( $1.0 \pm 0.3$  g/kg ideal body weight), far less than the mean daily protein of  $91 \pm 22$  g/d ( $1.3 \pm 0.4$  g/kg ideal body weight) in persons aged 19-30 (22). Given the steady aging of the American population and the public health implications if the prevalence of sarcopenia increases, it is important to clarify the relationship of protein intake to muscle mass and physical performance.

We examine the relationship of dietary protein to body composition and physical performance among older, community-dwelling, independent post-menopausal women. In particular, we hypothesized that physical function, assessed by direct observation, would be greater in subjects who consume a higher protein diet (above or equal to the RDA) when compared to those who consume less.

### Methods

#### Subjects

The analysis is based on baseline measurements of 387 healthy women aged 60-90 years (mean age  $72.7 \pm 7.0$  y) recruited from central Connecticut to participate in one of three intervention trials (23-25). The first two studies evaluated 1) dehydroepiandrosterone (DHEA) with gentle aerobic or yogic exercise and 2) 1.2g fish oil or placebo supplementation. Volunteers enrolled in the third study from August 2007 to November 2010 at the University of Connecticut Health Center (UCHC, Farmington, CT) and Yale University (New Haven, CT). For all studies, women being treated for osteoporosis, diseases or medications known to affect bone metabolism, or life expectancy less than two years were excluded. In the first two studies, women were selected for some level of frailty and in the third study; selection was based on reports of lower protein intake. There were no criteria for selection based on body mass index, medication or illness except those stated that may affect bone metabolism. The goal of combining the studies was to provide a large number of women with variability in protein intake and physical function. Written informed consent was obtained for all subjects and approved by the Institutional Review Boards.

#### Body Composition

Body composition was assessed using dual x-ray absorptiometry (DXA). Lunar Prodigy (Lunar Radiation Inc., Madison, WI) was used at UCHC and a Hologic 4500W (Hologic, Bedford, MA) was used at Yale. Lean body mass

including total lean mass and appendicular skeletal mass (ASM) and fat mass were assessed from the whole body scan.

#### Strength and Function

Physical function was determined using the Physical Performance Test (PPT) and the Short Physical Performance Battery (SPPB). The PPT measures upper extremity strength, fine and course motor function, mobility and coordination and includes simple functional tasks. Scores range from 0-28; higher scores indicate better performance. SPPB measures three individual measures of lower extremity function including measures of balance, walking speed, and strength. Each test is scored on a scale of 0-4 with the total score ranging from 0-12. Higher scores on the SPPB indicate greater physical function. A score of 11 or 12 on the SPPB is considered normal, while a score of 9 or less is indicative of frailty. A Jamar hand-held dynamometer was used to measure handgrip strength.

#### Questionnaires

For 2 studies (23, 25) the Physical Activity Scale in the Elderly (PASE), was used to assess physical activity (26); a higher score indicates more activity. The Medical Outcomes Survey Short-form 8 (MOS SF-8) was administered to assess participants' health related quality of life; higher scores indicate better well-being. Subjects also reported falls, fractures and medical diagnoses. Subjects met with a registered dietitian to receive instructions (including estimation) on completing a four-day food record. Subjects were instructed to record 4 consecutive days including 1 weekend day from estimated food/beverage consumption. The dietary food log was analyzed for daily nutrient intake using Food Processor software (ESHA Research, Salem OR, version 10.1.0).

#### Statistical Analysis

Using a cut-point of 0.8 g protein/kg body weight/day, subjects were divided into two groups, below or at and above the RDA for protein. Normal distribution of data was confirmed. Independent samples t-tests determined mean group differences between below or at the RDA protein groups. Due to differences in body composition between groups, analysis of covariance was used to evaluate differences in physical performance between groups, corrected for BMI. To confirm findings, linear regression analyses of physical function (SPPB/PPT) as dependent measures and intake of protein, carbohydrate and fat and the cutpoint for protein intake was performed. Significance was defined as p value < 0.05. SPSS version 19.0 (PASW Statistics, IBM, New York, NY) was used for the analysis.

### Results

A total of 387 older postmenopausal women participated; 7% of subjects reported heart disease, 39% had hypertension, 15% had osteoporosis and 27% had osteoarthritis. Ten percent

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of subjects reported a fall within the past six weeks, and 38.5% reported at least one fracture during their lifetime. Those in the low protein group had higher rates of hypertension, osteoarthritis and fracture. Caucasian women represented 95.5% of the study population, 3.4 % were African-American, and 1.1% were Asian. Forty-three percent of subjects were of normal body mass index (BMI), 33% were overweight and 23.5% met criteria for obesity.

On average, subjects consumed 72.2 g protein/day representing 1.1 g protein/kg body weight/day. The minimum protein intake reported was 0.31 g/kg/day and the maximum 3.16 g/kg/day. Ninety-seven subjects (25%) ate less than 0.8 g/kg/d, while 290 (75%) consumed at least that much (>0.8 g/kg/d). For the purposes of this study, these 2 groups will be referred to as low and high protein consumers.

**Table 1**

Demographics and Outcome measures in the below RDA and at /above RDA dietary protein subgroups

Variable	Mean ± SD		p value
	< .8 g/kg/d	≥ .8 g/kg/d	
Demographics	N = 97	N = 290	
Age, years	73.5 ± 6.7	72.5 ± 7.0	.182
Height, cm	160.8 ± 5.9	159.9 ± 6.7	.23
Weight, kg	75.5 ± 12.3	65.7 ± 12.1	< .001
BMI, kg/m2	29.2 ± 4.4	25.7 ± 4.8	< .001
Osteoporosis (%)	15	15	.478
Heart disease (%)	4	8	.228
Hypertension (%)	46	36	.050
Osteoarthritis (%)	40	22	.001
Reported Falls (%)	11.3	9.7	.380
Reported Fractures (%)*	48.9	35.0	.014
Body Composition			
Fat mass, kg	32.2 ± 8.4	25.3 ± 8.8	< .001
Lean mass, kg	40.7 ± 4.8	38.2 ± 4.6	< .001
ASM, kg	17.0 ± 2.3	15.9 ± 2.4	< .001
ASM/height2, kg/m2	10.5 ± 1.2	9.9 ± 1.3	< .001
Fat-to-Lean Ratio	0.79± .18	0.66 ± 0.20	< .001
Diet			
Calories, kcal	1245 ± 351	1778 ± 554	< .001
Calories from fat, kcal	392.1 ± 145.9	581.3 ± 274.8	< .001
Protein, g	49.7 ± 12.1	79.7 ± 24.3	< .001
Carbohydrates, g	159.4 ± 44.2	220.6 ± 76.7	< .001

Analysis by ANOVA or Chi square (for dichotomous variable); \* Fracture difference reported as ANCOVA adjusted for BMI; BMI-body mass index; ASM – appendicular skeletal mass

The average age of women was not different between the two protein groups (Table 1). Women in the higher protein group weighed less and had lower BMIs than the low protein group. Further, those in the high protein group had lower fat and lean mass, including the ratio of fat/lean mass. Dietary intake of calories, protein, fat and carbohydrates were greater in the high protein group consistent with the notion that they eat a more complete diet. There was no difference in the incidence of falls, but those in the low protein group reported more fractures and that difference persisted after adjusted for BMI.

There were significant and meaningful differences in physical performance between women consuming a higher protein intake and those with a lower protein intake (Table 2). The differences in SPPB and PPT composite scores were significantly different. Reported meaningful change in SPPB score is 0.45 27; there are no reported meaningful change data for PPT score, but scores 21-28 denote independence and scores from 3-15 are associated with dependent function (D Reuben, personal communication). Women in the lower protein group performed less well on the single leg stance component of the SPPB; their average time was 11.2 seconds compared to 15.3 seconds for the higher protein group (p = 0.002). They also walked eight feet at a slower pace than those women in the higher protein group (p = 0.006). Although the difference in chair rise time was not statistically significant, it was in the direction that we predicted. The average handgrip strength for both groups was similar. Because differences in body size may confound the differences in physical performance, we adjusted the analysis controlling for BMI. Overall, results were attenuated, but composite scores for physical function and single leg stance time remained significantly different between protein groups. When controlling for ideal body weight, results did not change and when controlling for lean mass, significance in PPT was lost, SPPB results remained and chair rise time became significant (data not shown).

**Table 2**

Physical performance, physical activity and health-related quality of life measures in the below RDA and at /above RDA dietary protein subgroups

Variable	Mean ± SD		p value	Analysis adjusted for BMI; p value
	< .8 g/kg/d	≥ .8 g/kg/d		
	N = 97	N = 290		
PPT	19.8 ± 2.9	20.9 ± 2.1	.006	.022
SPPB	9.9 ± 1.9	10.6 ± 1.6	< .001	.021
Chair rise, sec	14.9 ± 5.9	13.5 ± 7.9	.095	.332
Single leg stance, sec	11.2 ± 10.2	15.3 ± 10.9	.002	.043
Timed 8 foot walk, m/sec	1.0 ± 0.2	1.1 ± 0.2	.006	.170
Avg. handgrip strength, kg	18.5 ± 6.0	19.1 ± 6.4	.404	.338
<i>Self-Reported Physical Activity and Health related Quality of Life</i>				
PASE score *	183 ± 111	206 ± 110	.196	.299
PASE kilocal *	831 ± 438	846 ± 571	.851	.189
MOS SF-8 physical	47.9 ± 9.6	49.8 ± 8.2	.050	.919
MOS SF-8 mental	54.9 ± 7.2	55.2 ± 7.1	.701	.438

\* Data available for 214 women (N= 62 for low protein; N=152 for high protein subgroups) for PASE score and PASE kilocal; PPT- physical performance test; SPPB – short physical performance battery ; PASE –Physical Activity Score in the Elderly ; MOS SF-8 – Medical Outcome Study Short Form -8

Self-reported physical function was not different between protein groups. Those with lower protein intake reported lower scores on the physical component of the MOS SF-8 (p = 0.050), though this result was no longer significant when evaluated with BMI (p=.919). There were no differences in the

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mental component of the MOS SF-8.

In linear regression analyses with SPPB and PPT as the dependent measures, results from models including fat intake, protein, carbohydrate intake and cut-point for protein revealed a significant contribution from the cut-point for low protein intake (Table 3).

**Table 3**

Linear regression analysis modeling contribution of dietary intake of fat, carbohydrate and protein (as continuous vs cutpoint of RDA) on physical performance measures

	PPT B	SPPB Significance	B	Significance
Protein intake	-0.006	0.373	-0.001	0.844
Fat intake	0.009	0.148	-0.002	0.612
Carbohydrates intake	-0.002	0.302	0.000	0.814
Protein intake Cutpoint*	-1.154	0.004	-0.746	0.001

\* below 0.8 g/kg/d vs at or above 0.8g/kg/d; PPT- physical performance test; SPPB – short physical performance battery

**Discussion**

In a cross sectional observation study of 387 older women, we investigated the relationship of dietary protein to body composition and physical performance. Subjects consuming at and above the RDA for protein performed better on many of the self-reported and observed functional tests compared to those who consumed less than the RDA for protein. The low protein group had lower performance on the PPT and the SPPB, composite scores of upper and lower physical performance, in comparison to the higher protein group. These differences were observed despite differences in body composition, including higher weight, BMI, lean and fat mass and fat/lean ratio in the low protein group.

Consistent with our findings of better physical performance in the higher protein group, Beasley et al. found a 32% lower risk of frailty (low physical function, exhaustion, low physical activity and weight loss) in older adults with a 20% increase in protein (8). Similarly, low nutrient intake including a low intake of protein was associated with increased odds of frailty in an epidemiologic study of older European men and women (28). Women (n=280) in the United Kingdom with lower protein intake walked more slowly, though similar findings were not found in men (n=321) (29). We found that BMI moderates the effect of protein on physical function. Poor physical performance is reported in those with sarcopenic obesity (30, 31), but the independent association of low protein intake remained even when controlling for BMI.

Large epidemiologic studies assessing dietary protein report a consistent decline in protein intake with aging across a broad age range (32), though in our smaller age range we did not find an association. It is well established that adequate caloric intake is required to optimally utilize dietary protein to maintain muscle function rather than as energy source (33). It is curious

that those with low protein intake had a higher body mass and fat mass. The cause is uncertain, but could represent underreporting of total calorie and fat intake in those with higher BMI (34).

We selected 0.8 g protein/kg as the cut-off point in our study. The RDA for dietary protein is based on that which was required to achieve nitrogen equilibrium; it was originally 1.0 g/kg until the 1980 RDAs and it was subsequently decreased to 0.8 g/kg. There is significant evidence suggesting that the current recommendations for dietary protein in the elderly population (≥65) are inadequate for optimal muscle and bone health (15, 35-37). The attainment of nitrogen equilibrium alone (where nitrogen input is equal to output), as the primary outcome or end point has received considerable criticism (38), primarily because it has little relationship to physical or muscular function; it is simply a crude measure of nitrogen input and output. Our data suggest that simple measures of physical performance may also be important outcomes of adequate dietary protein in addition to bone and muscular health. Interestingly, the women in our low protein group reported significantly more fractures than the high protein group.

Data from epidemiologic studies found an association between higher lean mass and higher protein consumption in healthy men and women (6, 39), although this has not universally observed (40). We did not find an association between lean mass and protein intake, but found that those with higher BMI and higher fat/lean ratio were more likely to consume protein below the RDA. Studies of weight loss found that subjects on adequate protein diets are more likely to lose fat mass than lean mass (41, 42); corollaries may be made to decline in lean mass that accompanies unintentional weight loss or to the well recognized fall in lean and increase in fat mass in aging adults. The possibility exists that these body composition changes could be attenuated in elderly individuals consuming an adequate protein diet, and has recently been tested in a randomized trial in which skeletal muscle mass did not change but physical performance improved in 65 frail older adults receiving 15 g of protein at breakfast and lunch (43). Nonetheless it is known that higher fat mass is associated with increased insulin resistance which is a precursor to frailty (44) and is associated with muscle wasting and impaired protein metabolism (45). Older, non-diabetic individuals with evidence of insulin resistance have poor quadriceps muscle performance (46). In fact, our subjects had other evidence of physiologic decline associated with increased weight including higher rates of osteoarthritis and hypertension.

This study has limitations. Previous research indicates that protein from different sources may have different effects on energy balance, muscle protein synthesis and body composition and therefore muscle mass, strength and physical performance. Animal proteins, particularly those derived from dairy, might support muscle protein synthesis better than plant proteins (47). In this study we did not evaluate the source of the protein

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consumed by subjects. In addition, our study was comprised primarily of community-dwelling, independent, affluent, Caucasian women and the results may not reflect the interaction of protein and physical function in other populations. The cross sectional nature of our study does not allow us to claim causation only association. It may be that lower function with lower energy output decreases appetite and begins the cycle of poor function and low protein intake (48). Further, dietary intake was collected as self-report. The dietary record method provides quantitatively accurate information on food consumed during the recording period, though the reported energy and protein intakes may underestimate in the range of 4-37% compared to energy expenditure as measured by doubly labeled water or urinary nitrogen (49-52). All methods of dietary records have limitations, including 24 hour recall in an older population that may have poor recall or longer and more tedious food frequency questionnaires (51). Our study also had significant strengths. It rigorously evaluated dietary intake using a validated instrument and interviews by registered dietitians and we used a range of self-reported and observed measures to assess physical function.

### Conclusion

In conclusion, we found that healthy, older postmenopausal women consumed, on average, 1.1 g/kg/d protein, although 25% consumed less than the RDA. Though the women were not selected for impaired physical function or protein malnutrition, we were able to demonstrate an association between dietary protein on walking speed and single leg stance time. These measures were impaired in those who consumed protein below the RDA.

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