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Protein for Optimal Health in Aging Humans: Current Controversies

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Abstract

Although protein requirements as measured by nitrogen balance do not change with age, the elderly are at the greatest risk of deficiency (much more so than children). There is great interest whether the quantity and quality of their protein intakes can influence their quality of life in terms of overall mobility and general well-being. This presentation will attempt to make sense of an often diverse and complicated literature.

Glossary of Abbreviations

aLM: Appendicular Lean Mass
BMD: Bone Mineral Density
BMI: Body Mass Index
IGF-1: Insulin-Like Growth Factor 1
MD: Metabolic Demand
N: Nitrogen
PE Requirement: Protein-Energy Ratio (Ratio of requirements for protein expressed as energy)
PPU: Postprandial Protein Utilization
RCT: Randomized Controlled Trials
WHO: World Health Organization

Introduction

In discussing optimal intakes of protein in the human diet more than 10 years ago,[1] I noted that "progress is slow in defining quantifiable indicators of adequacy other than balance and growth" and that for the elderly "there is no evidence of an increased requirement or benefit from increased intakes, except possibly for bone health." Elsewhere in discussing sarcopenia in the context of aging and protein requirements,[2] I had argued that "the main determinant appears to be the decline in resistance-type physical activities," and "information on the extent of any nutritional influence on this decline is not currently available."

Since writing this, a new report on protein requirements has been published by WHO,[3] which has reviewed both the protein requirements and the beneficial and adverse influences of protein intakes on health. Important new research findings have been published leading to considerable advocacy that protein intakes above the recommended dietary allowances may benefit the elderly population.[4] My task here is to examine the justification of this advocacy in the context of the science base.

Protein Homeostasis and Aging

We can ask whether there are any *a priori* reasons to expect a change in the protein requirement with age based on our understanding of age-related changes in protein turnover and homeostasis. In fact, although a change with age in protein turnover would be clearly important in relation to the organism's ability to remodel itself, any such change need not necessarily relate to the dietary protein requirement. The protein requirement is best described in terms of metabolic demand (MD), a function of postabsorptive net catabolism, and efficiency of dietary protein utilization, which can influence the response to protein feeding.[3,5] Changes that occur with age in these aspects of protein homeostasis are the important question.

We need to examine within the daily cycle of feeding and fasting, the changes in protein synthesis, proteolysis and amino-acid oxidation that mediate fed-state gains and postabsorptive losses of body protein. Even from this more specific standpoint, it is still a difficult question because in subjects in overall nitrogen balance, the amplitude of fed-state anabolism and postabsorptive net catabolism is a function of the habitual protein intake.[5] This means there is no single measure in terms of protein synthesis, proteolysis or amino-acid oxidation that is completely independent of habitual protein intake that can be examined as a function of age, and this partly explains the divergence of reported findings.[2]

Key Words

Aging
Amino Acids
Bone Health
Muscle
Protein Intakes
Protein Requirements
Protein Synthesis
Sarcopenia

What we do know is that for metabolic demand, i.e, the magnitude of postabsorptive losses, there is a small but significant fall with age as indicated in ^{13}C leucine kinetic studies of leucine oxidation[2,6,7] (see Table 1). This occurs because of a greater decline in postabsorptive proteolysis (per unit of FFM) compared with protein synthesis.[2] Not only is there a small decline in the capacity of the organism to remodel itself, but

Table 1. Changes with Age in Metabolic Demands, Efficiency of Protein Utilization and the Apparent Protein Requirement for Nitrogen Balance

		Metabolic Demand (g protein/kg per d)†		PPU‡		Apparent Protein Requirement (g protein/kg per d)§	
	n	Mean	SD	Mean	SD	Mean	SD
Young Adult (F)	5	0·83	0·14a	0·99	0·07a	0·79	0·15a
Young Adult (M)	5	0·90	0·06a	1·05	0·10a	0·80	0·07a
Middle-Aged (M)	5	0·87	0·18a	1·01	0·05a	0·79	0·18a
Elderly (F)	5	0·52	0·14b	0·92	0·16a	0·57	0·20b
Elderly (M)	5	0·58	0·16c	1·05	0·13a	0·53	0·15b
All	5	0·74	0·21	1·00	0·12a	0·70	0·20

PPU, postprandial protein utilization; F, female; M, male.
Studies involved changes in N balance calculated from [1-13C] leucine balance during the transition from a low- to a high-protein intake in subjects fed repeated small milk-based meals.[5]
a,b,c Mean values within a column with unlike superscript letters were significantly different (P <0·05).
† Calculated from postabsorptive leucine losses scaled to 24 h assuming that leucine oxidation represents an equivalent loss of tissue protein nitrogen at 4·77mg leucine/g N and that the total amino acid–N conversion factor is 7·31.
‡ Fractional efficiency of protein utilization (utilization/intake) calculated from leucine balance/leucine intake.
§ Apparent dietary requirement for daily balance, calculated as the metabolic demand/PPU.

also an increasing restraint in postabsorptive proteolysis that limits net catabolism in the period after the protein intake from food has been absorbed and utilized. Whether these changes reflect an adaptive fall with age mediated by lower habitual protein intakes or a fundamental feature of aging is currently unknown, but they do mean that this component of the protein requirement is lower rather than higher in the elderly.

As for the response to feeding, this is much more difficult to assess and few reports have quantified this. In multilevel nitrogen (N) balance studies, the slope of the regression of N balance on protein intake is a measure of the efficiency of protein utilization, and the recent N balance studies of younger and older men and women[8] indicated no significant age (or gender) effects, especially when calculated on the basis of fat-free mass. However, as argued elsewhere[5] in N balance studies, the apparent efficiency of protein utilization (the slope of balance versus intake) at 50% or less is much lower than would be expected with the high-quality protein sources that are usually used in the balance studies.

The most likely explanation of this is incomplete adaptation to the low-protein diets fed in these multilevel N-balance studies that results in a lower utilization than occurs when adaptation is complete.[5] However, when studied with ^{13}C leucine balance in the fasted and fed state with milk or animal-sourced food-derived protein meals, no change with age in efficiency of protein utilization is observed[2,5,6] (see Table 1). The efficiency of protein utilization measured as N gain/N intake calculated from leucine gain/leucine intake is close to 100% efficient, i.e., postprandial protein utilization (PPU) is close to 1 in all age groups. These studies suggest a fall in age in the protein requirement, at least as measured in the laboratory under standardized conditions.

Many will find these results surprising, especially the lack of change in postprandial protein utilization, in light of experimental studies of the stimulation of protein synthesis in skeletal muscle by amino acids. We showed some three decades ago[9] that human muscle protein synthesis is stimulated 1.5 to 2 fold by a protein meal, and this effect is now known to be mediated by a combination of amino acids[10,11] and insulin.[14] In the case of amino acids, it appears that leucine, especially in the extracellular compartment, plays an important role in initiating a signaling cascade[12,13] activating protein synthesis, while the insulin response to feeding mediates a fall in proteolysis.[14]

Importantly, in elderly men there appears to be an anabolic resistance, a feature of aging in which amino acid supply is less able to elevate muscle protein synthesis,[15] and insulin is less able to decrease proteolysis of muscle protein[14] under conditions in which it would be expected that muscle anabolic processes would be stimulated, e.g., with feeding and after exercise. Anabolic resistance appears to be induced by decreased physical activity[16] so that age and decreased physical activity have a double-edged effect in decreasing the anabolic processes of muscle maintenance.

What is not known is the practical implication of this anabolic resistance within the daily gains and losses of tissue protein in muscle and elsewhere. On one hand, the rate of muscle wasting in the elderly exhibiting sarcopenia must be very slow on a daily basis because the deficit of muscle mass develops slowly over many years,[17] so that it is unlikely to be detectable within the overall whole body anabolic response to eating studied in Table 1.

On the other hand, the maximization of meal protein utilization for the elderly may require higher meal protein intakes than younger subjects. To some extent this has been examined in the daily feeding pattern of large meals versus multiple small meals.[18] If there is an anabolic resistance, then it might be assumed that the greater postprandial hyperaminoacidemia after a large protein meal would be more effective in mediating protein deposition than several smaller meals. This has been tested in meal feeding studies in subjects adapted over 14 days to specific meal size regimes.[18] When the protein intake was

mainly limited to one large meal, i.e., 80% of the daily protein intake fed at midday rather than spread throughout four meals, subjects did exhibit a more positive nitrogen balance and a better maintenance of fat-free mass.

Another reported change with age in amino acid kinetics that has been suggested to influence protein utilization relates to the splanchnic sequestration of dietary amino acids (the first pass effect) during the absorption of a protein meal. While the first pass effect is generally only important in relation to the interpretation of tracer-kinetic studies of amino acid and protein turnover, because there are suggestions that this splanchnic uptake increases with age,[19] it is often included as one of the features of aging that could impair dietary protein supply to the peripheral tissues, especially muscle.

However, it is not clear whether this is a real effect of aging or a consequence of confounding in the original studies because the elderly subjects were mainly overweight and obese compared with the younger subjects. Thus, the main correlate of splanchnic extraction in this study was body mass index (BMI), which indicates that as the relative splanchnic mass increases with BMI, so does apparent splanchnic extraction. This is unlikely to be an age-related change in protein and amino-acid metabolism that is relevant to the issue of protein requirements.

Measurement of Protein Requirements of the Elderly

In the new WHO report,[3] the protein requirement for adults was derived from a meta-analysis of nitrogen balance studies in healthy adults,[20] which identified no significant age effects but included only one relevant study. It also accepted the view of a rigorous reassessment of other reports of reanalysis and aggregation of earlier nitrogen balance data and all other available data[21] that no convincing evidence exists for a change in the protein requirement with age. This conclusion was confirmed by the recent nitrogen balance studies of younger and older men and women,[8] in which linear regression of the three protein intakes on nitrogen balance indicated protein intakes for nitrogen equilibrium of 0.59 g/kg/d, with no significant age or sex effects, especially when calculated on the basis of fat-free mass. This requirement value is within the range of the average requirement proposed in the new report, i.e., 0.66 g/kg/d.

The new WHO report[3] makes the point that because individuals consume food rather than individual nutrients there has been increasing interest in deriving food-based dietary requirements. Furthermore, because food intakes are largely driven by the organism's needs for energy, the expression of nutrient requirements as nutrient-energy ratios becomes the important descriptor of the dietary requirements for that nutrient.[22] Thus, the protein:energy ratio (protein calories/total calories), or PE requirement, the ratio of requirements for protein and for energy, identifies the required protein density of the diet that when consumed to energy needs will satisfy the protein requirements of an individual or population group.

Protein requirements for all ages are currently defined in terms of a fixed function of body weight. In contrast, energy requirements vary according to lifestyle (i.e., physical activity) and basal metabolic rate, itself a variable in relation to age, size and gender. This means that the PE requirement varies markedly with age and size (increasing as the BMR/kg falls), with gender (being higher for women than men because of the lower BMR/kg in women), and especially with lifestyle (falling as physical activity increases).

Thus, in contrast to protein requirements per kg, which fall markedly from infants to adults, the relationships between energy requirements and age, size, gender and activity protein result in a somewhat counterintuitive set of values for the PE requirement, which are lowest for preschool children and highest for elderly, large, inactive women. In other words, the very high energy requirements (per kg) of the child are satisfied by consuming large amounts of food that need only contain relatively low concentrations of protein to supply the protein requirement. In contrast, the much lower energy requirement (per kg) of an elderly, inactive woman is satisfied by smaller amounts of food that must therefore contain a relatively higher protein concentration.

While there are considerable theoretical difficulties in identifying reference or safe PE requirement values,[22] as shown in Table 2 from the new WHO report,[3] the PE requirement values for sedentary elderly women at 70 kg are more than twice the value for 5-year-old girls, i.e., 0.085 compared to 0.039 for mean PE requirement values and 0.106 compared to 0.052 for the reference (safe) PE requirement values. Although this may be surprising, compared with the PE ratio of an average adult omnivore diet, (e.g., PE ratio = 0.15) breast milk, with a PE ratio of 0.06, is a low-protein food. In practice this means that for any diet thought to be low in protein, it is the inactive elderly who are most likely to be at risk of protein deficiency, although the dietary PE value would have to be less than 0.11 (11% protein calories).

Protein Intakes

For the elderly, like the general adult population, surveys of protein intakes show that, with few exceptions, most mixed diets consumed to appetite provide intakes well above the requirement. This is certainly the case in the United Kingdom elderly population as shown in Figure 1. A wide overall range of protein intakes, up to >2 g/kgld, mainly reflects relative intakes of meat and other animal source foods.[23] Most of the population (80%) have intakes between 0.83 and 1.44 with a mean of 1.14 g/kgld. Because almost everyone has an intake above the mean protein requirement, prevalence of deficiency (intakes< requirement) is very low; the actual value, calculated with the algorithm reported in the WHO/FAO protein report,[3]

Table 2. Changes With Age in the Mean and Safe Protein:Energy Ratio of the Requirements[3]

	Mean Protein:Energy Ratio[a]						Safe Protein:Energy Ratio[b]					
	Males			Females			Males			Females		
Physical Activity Level / Age (y)	1.55 Light	1.75 Moderate	2.2 Heavy	1.55 Light	1.75 Moderate	2.2 Heavy	1.55 Light	1.75 Moderate	2.2 Heavy	1.55 Light	1.75 Moderate	2.2 Heavy
0.5		0.056			0.056			0.078			0.076	
2.5		0.036			0.039			0.05			0.053	
5		0.036			0.039			0.05			0.052	
10	0.054	0.046	0.04	0.059	0.05	0.043	0.074	0.062	0.054	0.081	0.068	0.059
15	0.061	0.052	0.045	0.068	0.06	0.05	0.084	0.071	0.062	0.093	0.082	0.069
Adults at 70 kg Body Weight												
18–29	0.068	0.06	0.048	0.068	0.069	0.055	0.094	0.083	0.067	0.108	0.096	0.076
30–59	0.071	0.063	0.05	0.071	0.074	0.059	0.098	0.087	0.069	0.117	0.103	0.082
>60	0.085	0.075	0.06	0.085	0.082	0.065	0.117	0.104	0.083	0.128	0.113	0.09
Adults at 50 kg Body Weight												
18–29	0.059	0.052	0.041	0.059	0.061	0.049	0.081	0.072	0.057	0.096	0.085	0.068
30–59	0.059	0.052	0.041	0.059	0.06	0.048	0.081	0.072	0.057	0.094	0.083	0.066
>60	0.073	0.064	0.051	0.073	0.068	0.054	0.1	0.089	0.071	0.106	0.094	0.075

[a]Calculated from the values for protein and energy requirements as (protein (g/kg) × 16.7) /energy (kJ/kg).
[b]Safe protein:energy ratio for an individual calculated from the values for protein and energy requirements.[3] These values are generally similar to calculations assuming a reference protein requirement = mean +3SD for adults and mean +3-4SD for infants and children.

is 2.7%, an acceptable level. Even after adjusting for <100% digestibility for the plant protein component, prevalence of deficiency is unlikely to warrant concern[23] especially when judged in the context of the adaptive metabolic demand model for the protein requirement.[5]

Protein Intakes and Disease and Disability in the Elderly

Because there is no objective measure of protein status, "deficiency" as calculated above is defined as a statistical construct, i.e., a measure of prevalence of intakes<requirement. The unambiguous identification of disease risk in relation to protein intakes or an optimal protein requirement is exceedingly difficult,[1,4] and in the review of the subject within the recent protein requirements report,[3] few specific benefits of higher-than-average protein intakes were identified. In fact, populations with lower protein intakes associated with low meat diets exhibit generally similar or even lower rates of morbidity and mortality compared with meat eaters.[24,25]

The issue whether variation in protein intakes toward marginal intakes is detrimental in elderly people consuming self-selected diets was addressed some years ago[26] with measurements of dietary intakes, plasma protein and arm muscle area for 691 men and women aged 60 to 98 years old consuming on average 1.04 g protein/kg. Some 12 to 15% of subjects had protein intakes < 0.8 g/kg, but clear, overt protein deficiency was not observed as far as serum albumin, triceps skin-fold thicknesses and transferrin concentrations, with no evidence that lower intakes of protein in the group adversely influenced any of these variables. Indeed, both arm circumference and a "nutritional index" score calculated from albumin, triceps skin-fold thicknesses and transferrin concentrations were inversely correlated with protein intakes, implying at the very least no deleterious effect of consuming protein at the lower end of the observed range.

Another important study[27] reported actual N balances for elderly people (70 to 86 years old) who were either housebound, consuming low energy and consequent low-protein intakes (recorded values 0.67g/kg/d), or healthy (70 to 86 years old) with higher protein intakes (0.97 g/kg/d). While the housebound subjects were mostly in negative balance and the healthy were at zero balance, there was no indication that protein intake determined balance with no correlation between protein intake and balance in either group over a wide range of intakes: 24-79 g protein/d in the housebound and 35-92 g protein/d in the healthy group.

Furthermore, at the same intakes, housebound subjects tended to be in negative balance whereas the healthy subjects were in positive balance. The immobility and/or illness or the lower energy intake of the housebound subjects accounted for the negative nitrogen balance. Notwithstanding the limitations of the N balance measurements, these data do not support any effect of protein intake on N balance over a range of intakes as wide as that likely to be observed in a free-living population.

Thus, these two studies point to free-living elderly individuals being able to adapt to protein intakes over a wide range, with no benefit from higher protein intakes, at least in terms of either biochemical indicators or measured N-balance.

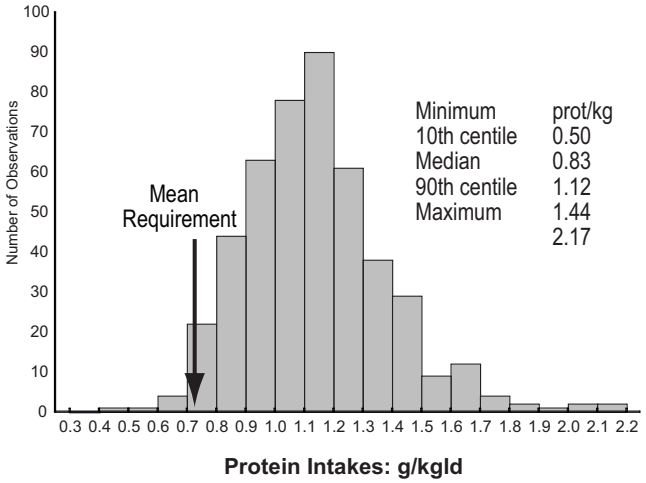
Protein Intakes and Sarcopenia

A major driver of the current advocacy of higher protein intakes for the elderly is the suggestion that higher protein intakes will protect against sarcopenia.[28] Sarcopenia, the decline in skeletal muscle strength and mass with advancing age, is a major determinant of impairment and disability. This is because skeletal muscle strength correlates with walking speed, balance, time to rise from a chair, ability to climb stairs, incidence of falls, and survival rates[29] and predicts physical dysfunction.[30]

Because it has been known for many years that strength training is quite effective in restoring age-related losses in muscle mass and strength function,[31,32] it is generally assumed that the main determinant of sarcopenia appears to be the decline in resistance-type physical activities.[33-37] It has been reported that a lifetime of intense aerobic physical activity has little impact on the decline in mass and strength,[38] and even in extremely fit elderly subjects or endurance-trained master athletes,[39] sarcopenia occurs. However, in the MINOS study,[17] low physical activity at work was identified as a risk factor for sarcopenia, and it has recently been demonstrated that muscle mass and strength in the elderly does respond to aerobic exercise.[40] This tends to suggest that sarcopenia is related to and can be reversed by all types of muscle activity.

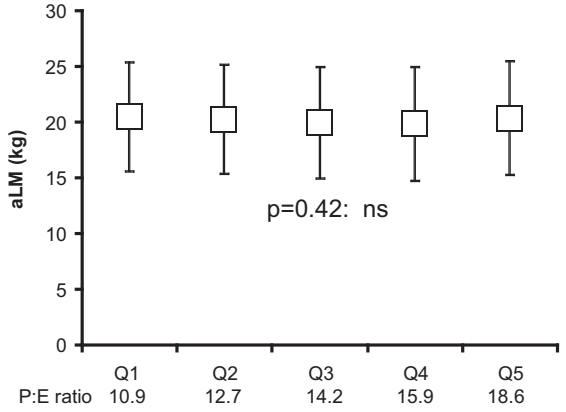
Nevertheless, it must be assumed that an adequate dietary protein intake is also a prerequisite for healthy aging, and it is clearly important to identify the extent of any nutritional influence on this decline in relation to both muscle strength and the metabolic implications of sarcopenia. There are two ways to establish the likely influence of protein intakes on sarcopenia, namely epidemiology and randomized controlled trials (RCTs). As far as epidemiology is concerned, the fact that sarcopenia occurs in the extremely fit implies that it is highly unlikely that a simple nutritional etiology, especially inadequate protein, would be identifiable. This is because high-intensity physical activity will promote increased energy demands and associated increased food intakes.

Figure 1: Distribution of Protein Intakes in the UK Elderly



Source: UK National Diet and Nutrition Survey[72] trimmed for under-reporters (i.e., energy intake <1.35 x estimated BMR).

Figure 2: Appendicular LM (kg) By Quintiles of Protein Intakes



Source: Health ABC study[44] at baseline: n=2066 mean age 74.5.

It can be assumed therefore that such populations will exhibit protein intakes at the upper end of the normal range. It is not surprising therefore to find that cross-sectional studies of sarcopenia and protein intakes have generally failed to show any association.[17,41-43] However, a recent large community-based longitudinal study of the loss of appendicular lean mass (aLM) conducted over three years (n=2066) in men and women aged 75,[44] showed that the aLM loss was greater in the lowest compared with the highest quintile of protein intake. This suggests that dietary protein may be a modifiable risk factor for sarcopenia in older adults and that higher intakes afford some protection.

In fact, some caution is needed in the interpretation of this study. First, there was no relationship between aLM and protein intake at baseline when the dietary data was collected (see Figure 2). This means that, as others have found, on a cross-sectional basis for the men and women studied in the Health ABC study, dietary protein intake was not identified as a causal factor for sarcopenia. Second, the positive protein intake-aLM relationship during the three-year longitudinal phase of the study was only observed for those who either lost or gained weight. The loss of aLM in the otherwise weight-stable subjects, half of the entire cohort, was not related to protein intake. This means that the relationship observed for the whole cohort is unlikely to be a simple consequence of

higher dietary protein intakes reducing sarcopenia.

As for RCTs of protein intakes and sarcopenia, given the slow rate of its development over many years,[17] RCTs for the prevention of sarcopenia are impractical. However, there is extensive literature on the influence of dietary protein on the efficacy of strength training. In one early study it was shown that muscle hypertrophy and strength gains can be achieved in elderly subjects even on a protein intake at the RDA level (0.8 g/kgld), which represented a reduced protein intake for the subjects.[45] According to a recent review,[32] "most of the limited research suggests that resistance training induced improvements in body composition, muscle strength and size, and physical functioning are not enhanced when older people who habitually consume adequate protein (modestly above the RDA) increase their protein intake by either increasing the ingestion of higher-protein foods or consuming protein-enriched nutritional supplements."

On the basis of a review of interventions for sarcopenia and muscle weakness in older people,[47] a recent task force on sarcopenia[46] concluded "it is not clear if protein supplementation in the absence of malnutrition enhances muscle mass and muscle strength, as protein supplementation alone or in association with physical training has proved unsuccessful." It would appear therefore that dietary protein intakes have little influence on the development of sarcopenia or the effectiveness of its treatment with resistance exercise.

Protein Intakes and Bone Health

Bone health and osteoporosis is clearly an important issue for the quality of life in the elderly. In men, osteoporosis and sarcopenia appear to go together, although this doesn't occur in women because bone mineral content is to a considerable extent a function of fat mass and associated oestrogen status, which tends to overwhelm any relationship between muscle mass and bone health.[48]

Dietary protein has been associated with both positive and negative influences on bone. There is a requirement for amino-acid precursors from dietary protein to maintain bone structure, and, in addition, the anabolic drive of amino acids on the organism includes an influence on bone, mediated in part through the stimulation of growth factors such as insulin-like growth factor I (IGF-I).[49] IGF-1 has been suggested to increase bone mass by increasing osteoblast activity, and may also increase the mineralization of bone matrix[50] in part by increasing calcium absorption.[51] Therefore, an inadequate anabolic drive due to insufficient dietary protein[52] may decrease bone strength through adverse changes in bone microarchitecture.[53] This indicates a need for adequate protein intakes for both the elderly and the general population to help optimize bone health. However, the balance between beneficial and detrimental influences of dietary protein on bone health is a long-standing debate.[3,54-56]

Dietary protein is a major contributor to acid production[57] as a result of the oxidation of the sulphur amino acids, and declining pH values influence the balance between osteoblastic and osteoclastic activity[58] and increase urinary calcium excretion.[59] The key question is the balance between these effects. High animal protein intakes have been associated with high bone fracture rates in cross-cultural studies,[60] but within populations there is little evidence for this. In fact, our recent systematic review and meta-analysis of dietary protein and bone health found only benefit or no influence.[61] Thus, we found that in a pooled analysis of cross-sectional surveys of the relation between protein intake and bone mineral density (BMD) or bone mineral content at the main clinically relevant sites, there was a beneficial influence with protein intake explaining 1 to 2% of BMD. Similarly a meta-analysis of randomized placebo-controlled trials indicated a significant positive influence of protein supplementation on lumbar spine BMD. However, there is no evidence that higher protein intakes reduced the relative risk of hip fractures at least in terms of studies published to date.

Protein Intakes and Cardiovascular Disease

There is a complex relationship between protein intake and cardiovascular disease that has yet to be fully resolved.[3] On the one hand, animal studies point to animal protein intakes having hypercholesterolaemic and atherogenic influences, but no such influences are observed in humans. Indeed, human studies have suggested higher protein intakes to be beneficial for the heart although no consensus has been reached about the causality or mechanisms of such associations. One possibility is a protective influence of protein intake on hypertension with inverse relationships between protein intake and blood pressure identified in several cross-sectional studies, the most recent being the INTERMAP study.[62]

A meta-analysis of studies up to 2002[63] indicated a convincing cross-sectional inverse association between dietary protein intake and blood pressure although longitudinal studies of intakes or changes in protein intake in relation to change in blood pressure or incidence of hypertension have been inconclusive. Elliott[64] cautions about over-interpretation of these studies, many of which involve secondary analyses and could be subject to various sources of bias. Certainly to date, there is no convincing body of evidence from intervention studies and no clear underlying mechanisms. Thus, while it can safely be assumed that high protein intakes are not likely to be damaging for cardiovascular health, any benefit must remain uncertain.

Conclusions

After reviewing the literature on protein intakes and health, WHO[3] concluded on a cautious note: "Current knowledge of the relationship between protein intake and health is insufficient to enable clear recommendations about either optimal

intakes for long-term health or to define a safe upper limit." Since then, although the expanding evidence base is certainly weighted toward benefit of higher protein intakes rather than harm, it does not contain the clear evidence from properly conducted randomized controlled trials that would warrant any change in this conclusion.

The only report of a successful protein-related intervention in the elderly that improved lean tissue mass is a double-blinded control trial in 78 elderly men and women (76 years old) given for one year daily an amino-acid cocktail containing arginine, lysine and the leucine metabolite β-hydroxy-β-methylbutyrate (BHMB).[65] This cocktail had been shown previously to improve functionality, strength, fat-free mass, and protein synthesis after 12 weeks,[66] and in the yearlong study it increased lean tissue mass. However, somewhat disappointingly, this time no change in strength was observed raising questions about the physiological significance of the increases in lean tissue mass.

In terms of protein intake related to resistance exercise and sarcopenia, a recent review concluded "research has not identified a synergistic effect of protein supplementation and resistance exercise in aging populations,"[67] and this was echoed in a more recent review.[68] A subsequent report reinforced that modestly increasing protein intake (from 0.9 to 1.2 g/kgld) predominantly from eggs had no influence on the gain in muscle induced by resistance training in older people.[69] In terms of optimal intakes for bone health, while the new analysis of the protein intake bone health literature points toward a benefit of protein, as yet fracture prevention has not been identified.

What has emerged in relation to maintaining muscle and bone health in the elderly is a clear benefit from adequate vitamin D. Not only has the importance for bone health of much higher levels of plasma 25-hydroxy vitamin D than are currently observed been unequivocally demonstrated in most populations with limited sunshine exposure, but an important role in maintaining muscle strength in older adults is now clear, resulting in less falls and thereby contributing to less fractures.[70,71] Muscle weakness is a prominent feature of the clinical syndrome of vitamin D deficiency, and vitamin D and its receptor are important for normal skeletal muscle development and in optimizing muscle strength and performance, with many supplementation trials of vitamin D in older adults showing a reduction in the risk of falls and improvements in tests of muscle performance.[70]

The most recent meta-analysis of double blind RCTs with supplemental vitamin D confirms a dose-dependent benefit on fall prevention in women, although not in men.[71] This is not to underestimate the importance of an adequate protein intake for the elderly, especially for sedentary populations with only modest energy needs. For these in particular, diets with adequate protein are necessary to enable current requirements to be met. For those who are active with higher energy needs and consuming higher food intakes, most balanced diets will provide more than enough protein.

The recent task force on sarcopenia[46] summarized its view on nutrition as follows: "A well-balanced diet, with adequate amounts of essential minerals, fatty acids and amino acids, together with an active and healthy lifestyle with regular periods of aerobic and resistance training, would be a correct life-course approach toward reducing the prevalence of sarcopenia and other chronic diseases in future elderly generations." As recently discussed,[73] my own highly speculative view is that sarcopenia results from reduced tension on muscle as bones slightly shorten with age. Thus, the key to health and active longevity may be sufficient appropriate exercise and healthy eating to ensure adequate intakes of protein and most other key nutrients to maintain muscle and bone strength and mobility. The demand for animal protein will no doubt continue to grow in the emerging economies, because meat is a preferred food in most societies. How much protein is needed will certainly continue to be debated, but whether global demands can be met is another story.[23]

References

1. Millward DJ. Optimal intakes of dietary protein. *Proceedings of the Nutrition Society*. 1999;58:403-413.

2. Millward DJ, Fereday A, Gibson NR, Pacy PJ. Aging, protein requirements and protein turnover. *Am J Clin Nutr*. 1997;66: 774-786.

3. World Health Organization/Food and Agriculture Organization/United Nations University. Protein and Amino Acid Requirements in Human Nutrition. *Report of a Joint WHO/FAO/UNU Expert Consultation*. WHO Technical Report Series No. 935. 2007;Geneva:WHO.

4. Rodriguez NR, Garlick PJ. Introduction to Protein Summit 2007: Exploring the impact of high quality protein on optimal health. *Am J Clin Nutr*. 2008;87(suppl):1551S–1553S.

5. Millward DJ. An adaptive metabolic demand model for protein and amino acid requirements. *British Journal of Nutrition*. 2003;90:249-260.

6. Fereday A, Gibson NR, Cox M, et al. Protein requirements and ageing: metabolic demand and efficiency of utilization. *British Journal of Nutrition*. 1997;77:685-702.

7. Short KR, Vittone JL, Bigelow ML, et al. Age and aerobic exercise training effects on whole body and muscle protein metabolism. *Am J Physiol Endocrinol Metab*. 2004;286:E92–E101.

8. Campbell WW, Johnson CA, McCabe GP, Carnell NS. Dietary protein requirements of younger and older adults. *Am J Clin Nutr*. 2008;88:322–329.

9. Rennie MJ, Edwards RHT, Halliday D, et al. Muscle protein synthesis measured by stable isotope techniques in man: the effects of feeding and fasting. *Clin Sci*. 1982;63:519-523.

10. Bennet WM, Connacher AA, Scrimgeour CM, et al. Increase in anterior tibialis muscle protein synthesis in healthy man during mixed amino acid infusion: studies of incorporation of [1–13C]leucine. *Clin Sci*. 1989;76:447-454.

11. Bennet WM, Connacher AA, Scrimgeour CM, Rennie MJ. The effect of amino acid infusion on leg protein turnover assessed by L-[15N]phenylalanine and L-[13C]leucine exchange. *Eur J Clin Invest*. 1990;20:37-46.

12. Bohe J, Low JF, Wolfe RR, Rennie MJ. Latency and duration of stimulation of human muscle protein synthesis during continuous infusion of amino acids *J Physiol*. 2001;532:575.

13. Bohe J, Low A, Wolfe RR, Rennie MJ. Human muscle protein synthesis is modulated by extracellular, not intramuscular amino acid availability: a dose–response study. *J Physiol*. 2003;552:315.

14. Wilkes EA, Selby A, Atherton P, et al. Blunting of insulin inhibition of proteolysis in legs of older subjects may contribute to age-related sarcopenia. *Am J Clin Nutr*. 2009;90:1343-1350.

15. Cuthbertson D, Smith K, Babraj J, et al. Anabolic signaling deficits underlie amino acid resistance of wasting, aging muscle. *FASEB J*. 2005;19:422-424.

16. Glover EI, Phillips SM, Oates BR, et al. Immobilization induces anabolic resistance in human myofibrillar protein synthesis with low and high dose amino acid infusion. *J Physiol*. 2008;586:6049-6061.

17. Szulc P, Duboeuf F, Marchand F, Delmas PD. Hormonal and lifestyle determinants of appendicular skeletal muscle mass in men: the MINOS study. *Am J Clin Nutr*. 2004;80:496-503.

18. Arnal M-A, Mosoni L, Boirie Y, et al. Protein pulse feeding improves protein retention in elderly women. *Am J Clin Nutr*. 1999;69:1202-1208.

19. Boirie Y, Gachon P, Beaufrere B. Splanchnic and whole body leucine kinetics in young and elderly men. *Am J Clin Nutr*. 1997;65:489-495.

20. Rand WM, Pellett PL, Young VR. Meta-analysis of nitrogen balance studies for estimating protein requirements in healthy adults. *Am J Clin Nutr*. 2003;77:109-127.

21. Millward DJ, Roberts SB. Protein requirements of older individuals. *Nutr Res Rev*. 1996;9:67-87.

22. Millward DJ, Jackson A. Protein:energy ratios of current diets in developed and developing countries compared with a safe protein:energy ratio: implications for recommended protein and amino acid intakes. *Public Health Nutrition*. 2004; 7(3):387-405.

23. Millward DJ, Garnett T. Food and the planet: nutritional dilemmas of greenhouse gas emission reductions through reduced intakes of meat and dairy foods. *Proceedings of the Nutrition Society*. 2010;69:103-118.

24. Key TJ, Appleby PN, Spencer EA, et al. Mortality in British vegetarians: results from the European Prospective Investigation into Cancer and Nutrition (EPIC-Oxford). *Am J Clin Nutr*. 2009;89:(suppl):1613S–1619S.

25. Key TJ, Fraser GE, Thorogood M, et al. Mortality in vegetarians and nonvegetarians: detailed findings from a collaborative analysis of 5 prospective studies. *Am J Clin Nutr*. 1999; 70(suppl):516S-524S.

26. Munro HN, McGandy RB, Hartz SC, et al. Protein nutriture of a group of free-living elderly. *Am J Clin Nutr*. 1987;46: 586-592.

27. Bunker VW, Lawson MS, Stanfield ME, Clayton BE. Nitrogen balance studies in apparently healthy elderly people and those who are housebound. *Br J Nutr*. 1987;47:211-221.

28. Paddon-Jones D, Rasmussen BB. Dietary protein recommendations and the prevention of sarcopenia. *Curr Opin Clin Nutr Metab Care*. 2009;12:86-90.

29. Rantanen T, Guralnik JM, Sakari-Rantala R, et al. Disability, physical activity, and muscle strength in older women: the women's health and aging study. *Arch Phys Med Rehabil*. 1999; 80:130-135.

30. Brill PA, Macera CA, Davis DR, et al. Muscular strength and physical function. *Med Sci Sports Exerc*. 2000;32:412-416.

31. Frontera WR, Bigard X. The benefits of strength training in the elderly. *Science & Sports*. 2002;17:109-116.

32. Campbell WW, Leidy HJ. Dietary Protein and Resistance Training Effects on Muscle and Body Composition in Older Persons. *Journal of the American College of Nutrition*. 2007; 26(6):696S-703S.

33. Frontera WR, Meredith CN, O'Reilly KP, et al. Strength conditioning in older men: skeletal muscle hypertrophy and improved function. *J Appl Physiol*. 1988;64:1038-1044.

34. Klitgaard H, Mantoni M, Schiaflino S, et al. Function, morphology and protein expression of ageing skeletal muscle: a cross-sectional study of elderly men with different training backgrounds. *Acta Physiol Scand*. 1990;140:41-54.

35. Frontera WR, Meredith CN, O'Reilly KP, Evans WJ. Strength training and determinants of VO2 max in older men. *J AppI Physiol*. 1990;68:329-333.

36. Fiatarone MA, Marks FC, Ryan ND, et al. High-intensity strength training in nonagenarians. Effects on skeletal muscle. *JAMA*. 1990;263:3029-3034.

37. Fielding RA. Effects of exercise training in the elderly: impact of progressive-resistance training on skeletal muscle and whole-body protein metabolism. *Proc Nutr Soc*. 1995;54:665-675.

38. Harridge S, Magnusson G, Saltin B. Life-long endurance-trained elderly men have high aerobic power, but have similar muscle strength to non-active elderly men. *Aging* (Milano). 1997;9(1-2):80-87.

39. Louis J, Hausswirth C, Bieuzen F, Brisswalter J. Muscle strength and metabolism in master athletes. *Int J Sports Med*. 2009;30(10):754-759.

40. Harber MP, Konopka AR, Douglass MD, et al. Aerobic exercise training improves whole muscle and single myofiber size and function in older women. *Am J Physiol Regul Integr Comp Physiol*. 2009;297:R1452-R1459.

41. Baumgartner RN, Koehler KM, Gallagher D, et al. Epidemiology of sarcopenia among the elderly in New Mexico. *Am J Epidemiol*. 11998;47:755-763.

42. Starling RD, Ades PA, Poehlman ET. Physical activity, protein intake, and appendicular skeletal muscle mass in older men. *Am J Clin Nutr*. 1999;70:91-96.

43. Mitchell D, Haan MN, Steinberg FM, et al. Body composition in the elderly: the influence of nutritional factors and physical activity. *J Nutr Health Aging*. 2003;7:130-139.

44. Houston DK, Nicklas BJ, 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. *Am J Clin Nutr*. 2008;87:150.

45. Campbell, Wolfe, Evans. Adequacy of the RNI to support resistive training in older humans. *J Physiol.* 2002;542:631-642.

46. Abellan Van Kan G, André E, Bischoff-Ferrari HA, et al. Carla task force on sarcopenia: propositions for clinical trials. *The Journal of Nutrition, Health & Aging.* 2009;13(8);700-707.

47. Burst SE. Interventions for sarcopenia and muscle weakness in older people. *Age Ageing*. 2004;33(6):548-555.

48. Baumgartner RN, Stauber PM, Koehier KM, et al. Associations of fat and muscle masses with bone mineral in elderly men and women. *Am J Clin Nutr*. 1996;63:365-372.

49. Millward DJ, Rivers JPW. The need for indispensible amino acids: the concept of the anabolic drive. *Diabetes-Metabolism Reviews*. 1989;5:191-212.

50. Rizzoli R, Bonjour JP, Chevalley T. Dietary protein intakes and bone growth. *Bone, International Congress Series*. 2007;50–59.

51. Rizzoli R, Ammann P, Chevalley T, Bonjour JP. Effects of dietary protein insufficiency on the skeleton. In New SA, Bonjour JP (eds): *Nutritional aspects of bone health*. Royal Society of Chemistry, Cambridge, United Kingdom. 203;194-212.

52. Heaney RP, Layman DK. Amount and type of protein influences bone health. *Am J Clin Nutr*. 2008;87(suppl):1567S-1570S.

53. Bonjour JP. Dietary protein: an essential nutrient for bone health. *J Am Coll Nutr*. 2005;24:526S-5236S.

54. Roughead ZK. Is the interaction between dietary protein and calcium destructive or constructive for bone? *J Nutr*. 2003;133:866S-869S.

55. Ginty F. Dietary protein and bone health. Proc *Nutr Soc*. 2003;62:867-876.

56. Rizzoli R, Bonjour JP. Dietary protein and bone health. *J Bone Miner Res*. 2004;19:527-531.

57. Remer T, Manz F. Potential renal acid load of foods and its influence on urine pH. *J Am Diet Assoc*. 1995;95:791-797.

58. Arnett TR, Dempster DW. Effect of pH on bone resorption by rat osteoclasts in vitro. *Endocrinology*. 1986;119:119-124.

59. Lemann J, Litzow JR, Lennon EJ. Studies of the mechanism by which chronic metabolic acidosis augments urinary calcium excretion in man. *J Clin Invest*. 1967;46:1318-1328.

60. Abelow BJ, Holford TR, Insogna KL. Crosscultural association between dietary animal protein and hip fracture: a hypothesis. *Calcif Tissue Int*. 1992;50:14-18.

61. Darling AL, Millward DJ, Torgerson DJ, et al. Dietary protein and bone health: a systematic review and meta-analysis. *Am J Clin Nutr*. 2009;90:1674-1692.

62. Elliott P, Stamler J, Dyer AR, et al. Association between protein intake and blood pressure: The INTERMAP Study. *Arch Intern Med*. 2006;166,79-87.

63. Liu L, et al. Epidemiological evidence of the association between dietary protein intake and blood pressure: a meta-analysis of published data. *Hypertension Research – Clinical and Experimental*. 2002;25:689-695.

64. Elliott P. Protein intakes and blood pressure in cardiovascular disease. *Proceedings of the Nutrition Society*. 2003;62:495-504.

65. Baier S, Johannsen D, Abumrad N, et al. Year-long Changes in Protein Metabolism in Elderly Men and Women Supplemented With a Nutrition Cocktail of β-Hydroxy-β-methylbutyrate (HMB), L-Arginine, and L-Lysine. *J Parenter Enteral Nutr*. 2009; 33:71.

66. Flakoll P, Sharp R, Baier S, et al. Effect of beta-hydroxy-beta-methylbutyrate, L-arginine, and L-lysine supplementation on strength, functionality, body composition, and protein metabolism in elderly women. *Nutrition*. 2004;20:445-451.

67. Paddon-Jones D, Short KR, Campbell WW, et al. Role of dietary protein in the sarcopenia of aging. *Am J Clin Nutr*. 2008;87(suppl):1562S-1566S.

68. Paddon-Jones D, Rasmussen BB. Dietary protein recommendations and the prevention of sarcopenia. *Current Opinion in Clinical Nutrition and Metabolic Care*. 2009;12:86-90.

69. Iglay HB, Apozan JW, Gerrard DE, et al. Moderately increased protein intake predominantly from egg sources does not influence whole body, regional, or muscle composition responses to resistance training in older people. *J Nutr, Health Aging*. 2009;13:108-114.

70. Ceglia L. Vitamin D and its role in skeletal muscle. *Current Opinion in Clinical Nutrition and Metabolic Care*. 2009;12:628-633.

71. Bischoff-Ferrari HA, Dawson-Hughes B, Staehelin HB, et al. Fall prevention with supplemental and alpha-hydroxylated vitamin D: a meta-analysis of randomized controlled trials. *BMJ*. 2009;339:b3692 doi:10.1136/bmj.b3692.

72. Finch S, Doyle W, Lowe C, et al. National Diet and Nutrition Survey: People Aged 65 Years and Over. *Report of the Diet and Nutrition Survey*. The Stationery Office, London. 1998, Volume 1.

73. Millward DJ. Sufficient protein for our elders? *Am J Clin Nutr*. 2008;88(editorial):1187-1188.

Q&A Discussion

Q: Dr. Dottie Laflamme, Nestlé Purina PetCare: So much of the data on protein requirements are based on nitrogen balance evidence. And there are some individuals who would say that nitrogen balance is probably not an appropriate measure of optimum protein requirements because you can have a loss of lean body mass and you can have a reduction in protein turnover and still maintain nitrogen balance. So I'd like to invite you to give your opinion on if you think that nitrogen balance truly is a sufficient measure, or if in the long term, we should be looking at something else?

A: Dr. Millward: It would be very nice to be looking at something else because nitrogen balance is not a measure of optimal protein intakes. What nitrogen balance does, in theory, is what any nutrient-balance study does. It tells you whether the organism is gaining or losing that nutrient. There are problems with nitrogen balance. Nitrogen balance measures the minimum protein requirements, and that has to be very clear. That is the minimum intake to maintain people in nitrogen equilibrium. There are many secondary problems with nitrogen balance in terms of whether it actually does that. There can be problems with methodology and with unmeasured nitrogen loss. But nitrogen balance should never be claimed to be a measure of optimum status. And there isn't a single measurement that can give you optimum status, because if it's bone health, if it's sarcopenia, if it's whatever it

is, then there has to be an evidence base that builds up sufficient quantitative data between intake and outcome that enables you to make sensible evidence-based conclusions about what intake would be appropriate. And I don't think there is an evidence base that allows us to do that.

Q: Dr. Bob Backus, University of Missouri: Is there any data on whether people eating more protein live longer?

A: Dr. Millward: I know of none. I think that whole area is problematic. I know we've got this huge animal database and primate database for energy and longevity and there is supposed to be a little bit of human data that supports that, which I'm quite skeptical of. Increased physical activity is supposed to give us better prolonged health, which, of course, would involve increased intakes. But as far as protein intake, it's actually not a question that we can answer now, I don't think.