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Nutritional interventions in sarcopenia: a critical review

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The aim of the present paper is to critically review the details of the published nutrition intervention trials, with and without exercise, targeting sarcopenia. Sarcopenia is the loss of muscle mass, strength and/or performance with age. Since amino acids and energy are required for muscle synthesis it is possible that nutritional intake influences sarcopenia. Nutritional studies are challenging to carry out because of the complexity of modulating dietary intake. It is very difficult to change one nutrient without influencing many others, which means that many of the published studies are problematic to interpret. The studies included evaluate whole protein, essential amino acids and β-hydroxyl β-methylbutyrate (HMB). Whole-protein supplementation failed to show a consistent effect on muscle mass, strength or function. This can be explained by the variations in study design, composition of the protein supplement and the failure to monitor voluntary food intake, adherence and baseline nutritional status. Essential amino-acid supplements showed an inconsistent effect but there are only two trials that have significant differences in methodology and the supplement used. The HMB studies are suggestive of a beneficial effect on older adults, but larger well-controlled studies are required that measure outcomes relevant to sarcopenia, ideally in sarcopenic populations. The issues of timing and distribution of protein intake, and increased splanchnic amino-acid sequestration are discussed, and recommendations for future trials are made.

Sarcopenia: Vitamin D: Protein: Essential amino acid: β-Hydroxyl β-methylbutyrate

Sarcopenia is the loss of muscle mass, strength and/or performance with age. There is as yet no universally agreed definition but a number of groups have published definitions which differ mainly around the exact cut-off points used for measures of mass, strength and performance[1–6].

Since a consistent supply of amino acids and energy are required for muscle protein synthesis, as well as a number of other nutrients, it is possible that nutritional intake contributes to the process of sarcopenia[7]. Consequently modifying nutritional intake may also provide an opportunity to treat sarcopenia.

The process of muscle synthesis and breakdown is complex and influenced by various internal and external factors. A comprehensive review can be found at[8]. In particular, from the nutritional perspective, it is accepted that amino acids have a stimulatory effect on muscle synthesis, especially leucine[9]. Thus, protein and amino acids have been among the first targets for interventions to combat sarcopenia, alone or in combination with exercise, and form the majority of nutritional intervention trials.

The aim of the present paper is to critically review the details of the published nutrition intervention trials, with and without exercise, targeting sarcopenia. Nutritional studies are challenging to carry out because of the complexity of modulating dietary intake. It is very difficult to change one nutrient without influencing many others, which means that many of the published studies are problematic to interpret. Studies were included if the

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outcome measures reported for the interventions included muscle mass and at least one measure of muscle strength or physical performance, even when the population studied was not defined as sarcopenic, since these outcome measures form part of the definition of sarcopenia.

Whole-protein studies

A recent systematic review[10] identified five protein supplement studies[11–15], which are summarised in Table 1; Cruz-Jentoft et al. concluded that there was no consistent effect on muscle mass, strength or function. A closer examination of the studies reveal why this may be so. Table 1 shows that the studies are highly variable in a number of different aspects, including the population recruited, the duration, whether the intervention was combined with exercise, supplementing protein and energy or protein alone, the formulation and precise content of the supplements, and whether placebo controlled or not. In addition, the methodological quality as assessed by Cruz-Jentoft et al. varied from a PEDro score[16] of 4–10 (10 representing the highest quality score possible), outcomes measured all varied and all sample sizes were relatively small ranging from fifty-seven to ninety-eight people[10].

The duration of the interventions ranged from 6 to 18 months, but the results show no advantage to the longer duration, but this may be explained by other factors. Tieland et al. suggest that the lack of effect of protein supplementation with exercise on physical performance and strength may be due to duration, postulating that longer than 6 months intervention would be required[14]. Unfortunately the two available longer term studies[11,12] had significant design weaknesses making it difficult to deduce whether this hypothesis can be supported. To further confuse the issue, in another Tieland et al. study protein supplementation for 6 months without exercise did improve physical performance but did not show gains in muscle mass or strength[15]. The populations were also inconsistent, including older adults (mean age range 68–83 years) who were free living[12–15] or in residential care[11]. Three studies specifically identified frail or pre-frail people[11,14,15]. This means that the results cannot be combined and they can only be applied to these specific groups.

The ability to identify the singular effect of protein with or without exercise is only possible with two studies, where a four arm design was used (exercise, protein, both or none)[11,12]. Two other studies looked at the additive effect of protein on exercise[13,14], and the final study examined protein supplementation excluding the effect of exercise[15]. Since there is widespread agreement that exercise can improve both muscle strength and physical performance, it would be useful to be able to examine how nutritional supplements enhance these effects or not. This requires four arm studies, until the optimally effective exercise intervention is described, when two arm studies could be justified looking at nutrition in addition to exercise. Nevertheless, two arm studies examining nutrition only still have value given the difficulties in motivating people to take up exercise. This lack of consistency in study design means that comparing or combining results is difficult or impossible and explains to a certain extent the variability of the results to date.

Most importantly the protein interventions tested varied considerably. In two trials the protein was delivered with additional energy and micronutrients[11,12], but these trials scored lowest in terms of quality on the PEDro scale. Bunout et al. do not describe the exact content of the supplement since it was apparently incorporated into either soup or porridge and it is not stated what additional nutrients these foods added. The protein content is relatively low at 13 g/d, but a range of vitamins and minerals are also included to provide a more complete supplement[12]. Bonnefoy et al. also aimed to provide a nutritionally complete supplement containing vitamins and minerals in addition to protein and energy. The quantities of energy and protein appear significant at 1686 kJ and 30 g protein/d, but this is difficult to assess without measuring usual food intake to enable an estimate of the overall energy and protein content of the diet[11]. The other three trials aimed to examine the effect of protein only, with no additional energy or micronutrients. Chale et al. specifically chose to use whey protein due to the high content of essential amino acids (EAA) in this protein[13]. Tieland et al. do not state the source of protein[14,15]. These three studies provide either 30 or 40 g protein/d, significantly increasing protein intake as shown by an estimation of dietary intake using 3 d dietary diaries.

The measurement of concurrent food intake is a key design issue in nutritional intervention trials, but is not always monitored. Since any nutritional supplement may have an impact on usual food intake it is important to measure macronutrient intakes. In the case of protein, an energy deficit will result in a reduction in protein synthesis even with apparent adequate supplies of amino acids[17]. Similarly, if protein intake from the diet is inadequate the supplement will simply improve intake rather than increasing intake above recommended levels. If positive benefits are found in the supplemented groups, then it is important to be able to identify the overall nutrient intake to enable accurate interpretation of the results. If food intake is not measured, then it is impossible to assess what has produced any positive effect, or explain why no effect is apparent. The two lower quality studies did not aim to measure usual food intake[11,12]; the other three studies used 3 d diet diaries at least at the start and end of the study[13–15].

There are a number of recent articles from expert groups reviewing the recommended protein levels for older adults. These suggest that protein intake may need to increase with age (specifically over 65 years) to 1·0–1·2 g/kg body weight per d[18,19]. These levels are not universally accepted and there remains significant debate around this issue[20]. Therefore, it is vital that trials testing protein supplementation need to enable subjects to at least meet these newly proposed levels, rather than just providing a fixed amount in the hope protein intake increases appropriately.

Table 1. Details of protein supplementation trials to treat sarcopenia

Reference (population)	Intervention (duration)	Total supplement/d	No./type of supplements	Volume/ supplements	Energy/ supplements	Protein /supplements	Extra nutrients?	Masking	Placebo	Adherence	Dietary intake	Main results
Bonnefoy et al.[11] (residents of retirement homes)	(a) Ex + Supp (b) Con + Supp (c) Ex + Placebo (d) Con + Placebo (9 months)	30 g protein 1686 kJ (400 kcal)	Two drinks 10·00 and 16·00	200 ml	843 kJ (200 kcal)	15 g (30 % of energy); g/kg bw per d not given	Yes – range of vits and mins providing 25 % RDA /supp	Attempted same opaque packaging but products likely to taste and look different	Yes No protein, energy, vits or mins. Presumably was artificially sweetened and flavoured water but not stated	54 % at 9 months counting unused units	No measure made but BMI increased in the supp group and supp designed to increase energy intake by approximately 20 %	Supp significantly increased MP at 3 months v. Con (P = 0·03), but not at 9 months. Supp did not improve MM or PP v. Con
Bunout et al. [12] (free-living elderly attending out-patient clinics)	(a) Ex + Supp (b) Supp (c) Ex (d) Con (18 months)	13 g protein 1746 kJ (418 kcal)	Two prepared as soup or porridge (probably powdered product but not stated)	50 g	873 kJ (209 kcal) (excluding soup or porridge contribution)	6·5 g; g/kg bw per d not given	Yes – range of vits and mins providing 25 % RDAs /supp	Not applicable	No Supp provision not randomised	44–49 % Self-reported	No assessment of dietary intake and no change in body weight or composition	Supp alone had no effect on MM, MS or PP. Supp did not show an additive effect over Ex outcome
Chale et al.[13] (free-living elderly)	(a) Supp + Ex (b) Placebo + Ex (6 months)	40 g whey protein 1582 kJ (378 kcal)	Two powder sachets am and pm. If doing Ex taken after the session	Not stated	791 kJ (189 kcal)	20 g; g/kg bw per d not given	No	Numerically coded packages	Yes Iso-energetic 45 g maltodextrin 1 g fat; 791 kJ (189 kcal) PABA for compliance checking	67 % Urinary PABA 72 % returned packages	3 d diet diary × 2 Protein intake greater in supp group and energy intake the same as baseline. Showed voluntary energy intake decreased but supp maintained it	Supp + Ex did not improve MM, MS or PP significantly v. Con + Ex
Tieland et al.[14] (Free living frail or pre-frail elderly)*	(a) Supp + Ex (b) Placebo + Ex (6 months)	30 g protein	Two drinks after breakfast and lunch	250 ml	Not stated	15 g; 1·3 g/kg bw per d	0·4 g Ca	All drinks flavoured vanilla opaque packaging	Yes No protein same lactose and Ca	98 % Ticked calendars and returned cartons	3 d diet diary × 3 No change in energy intake, protein increased in supp group	Supp + Ex significantly improved MM (P = 0·006), but not MS or PP v. Placebo + Ex
Tieland et al.[15] (free-living frail or prefrail elderly)*	(a) Supp (b) Placebo (6 months)	30 g protein	Two drinks after breakfast and lunch	250 ml	Not stated	15 g; 1·4 g/kg bw per d	0·4 g Ca	All drinks flavoured vanilla opaque packaging	Yes No protein same lactose and Ca	92 % Ticked calendars and returned cartons	3 d diet diary × 3 Energy intake decreased at 24 months, protein increased in supp group	PP improved significantly with supp (P = 0·02), but not MM or MS v. Placebo

Ex, exercise component; con, control; supp, supplement; bw, body weight; PABA, para-aminobenzoic acid; MM, muscle mass; MP, muscle power; MS, muscle strength; PP, physical performance; vits, vitamins; mins, minerals.
* Registered as two separate two arm studies.

Only two studies assessed this[14,15] and showed protein intake increased up to 1·3–1·4 g/kg body weight per d, but provided inconsistent results in terms of protein supplementation as mentioned previously.

Adherence is another critical factor in nutritional interventions. It is well known that nutritional supplements or changes to dietary intake are notoriously difficult for subjects to follow consistently[21]. All studies in this group reported adherence, but three had relatively low rates[11–13], suggesting that a lack of adherence could be the reason for a lack of effect.

Producing a placebo supplement that enables single or double blinding to limit bias is also extremely difficult, particularly if a liquid supplement drink is employed. Some studies have attempted this; one aiming for an energy and protein-free placebo[11] and another aiming for an iso-energetic but protein-free placebo[13]. Both these placebos are likely to taste and feel very different to the supplement, resulting in unsuccessful masking, at least to the individuals taking it. Other studies either did not attempt to provide a placebo[12] or provided limited information regarding energy content[14,15]. None of the researchers have attempted to test whether masking was successful, but have simply obscured the liquids from view to avoid comparison or visual assessment of the drink. Chale *et al.* do not provide details of how the supplement powder is consumed; whether added to solid food or mixed with liquid to produce a drink. In all cases there is a significant risk that the placebo drink or powder will be readily identifiable to those consuming it, thus allowing potential bias to influence results.

Baseline nutrient status is similarly critical to assess. People who are deficient in any particular nutrient may respond differently to the given supplement. Vitamin D status provides a good example of how a deficient status may influence the effect of a protein and energy supplement. This is discussed later in the β-hydroxyl β-methylbutyrate (HMB) section.

Finally, recruitment in all the published trials appears to have been difficult. Recruitment rates ranged from 4 to 9 % of the initially screened population. One author comments 'The greatest difficulty is to motivate elderly individuals to volunteer for such a study or to perform any effort to improve their health'[11]. This may introduce bias since it may be that only particularly health conscious individuals will participate in the studies and these people are not representative of older adult populations. It also illustrates the challenges faced by researchers in planning and funding such studies. They are likely to be resource intensive and expensive, and it may be necessary to conduct extensive preliminary work to establish interventions that are more attractive and acceptable to older adults.

In summary the trials testing whole-protein supplementation failed to show a consistent effect on muscle mass, strength or function. This can be explained by the variations in study design, composition of the protein supplement and the failure to monitor voluntary food intake, adherence and baseline nutritional status. Further large-scale clinical studies are warranted but will require significant funding and resources.

Essential amino-acid trials

The rationale for supplementing with EAA is that they are an anabolic stimulus for muscle synthesis[9], in particular leucine. However, there is no agreement on what mixture of EAA may provide the best stimulus. There are two studies which have used such a mixture (see Table 2), providing very limited evidence that there may be some effect on muscle mass and function through amino-acid supplementation[22,23]. The two studies used very different EAA mixtures with the main differences in threonine, phenylalanine and leucine, yet neither provided a clear rationale for the mixture in use. Furthermore, the method of delivery differed with one study providing capsules (20/d)[22] and other asking subjects to mix a powder with water or milk[23]. This is particularly problematic since using milk as an optional delivery vehicle for the EAA means that some subjects will also receive a supplement of energy, protein (including EAA) and other nutrients contained in the milk. Many amino acids have a bitter taste and it remains unclear how palatable this oral supplement was or whether other flavours or sweeteners were used within the powder to make its taste acceptable.

As with the whole-protein trials, these studies also suffer from a lack of adherence information, no assessment of baseline nutritional status and no assessment of voluntary food intake. They are also very different designs; two arm compared with four arm study, and with and without placebo.

Both these studies found an effect of the supplements; one on muscle mass only[22] and the other on mass, function and strength, but only in combination with exercise[23]. The EAA supplement alone did not result in significant change[23]. Because of the lack of rationale and different EAA supplements used, and the various design problems in both studies it is difficult to interpret whether further studies into the use of EAA are warranted. It may be that using HMB, a downstream metabolite of leucine, is a better approach, and there are five studies exploring this option.

β-Hydroxy β-methylbutyrate trials

HMB has been shown to reduce protein degradation, up-regulate protein synthesis and increase muscle cell cholesterol production, leading to more stable cell membranes[24,25]. It is a downstream metabolite of leucine, with approximately 5–10 % ingested leucine being converted to HMB[26]. This means that in order to meet the dose of HMB generally given in trials (3 g/d)[27,28], 60 g leucine/d would have to be consumed. To obtain this quantity of leucine through the diet would require an individual to eat impractically large amounts (>600 g) of high-quality protein sources (eggs, dairy and meat) daily[29]. The theory is that if only 10 % leucine is converted to HMB and HMB is what makes leucine a stimulator of muscle synthesis, a far larger effect could be elicited if the rate-limiting conversion is bypassed. A number of human studies have tested the theory of

Table 2. Details of amino-acid trials to treat sarcopenia

Reference (population)	Intervention (duration)	Presentation	Essential amino acid content/d g (%)									Extra nutrients?	Placebo	Adherence	Dietary intake	Main results for treated group
			His	Ile	Leu	Lys	Met	Phe	Thr	Trp	Val					
Dillon et al.[22] (Healthy women)	(a) 15 g EAA (b) Placebo (3 months)	Twenty capsules twice daily (forty in total)	1·64 (10·9)	1·56 (10·4)	2·78 (18·6)	2·34 (15·5)	0·46 (3·1)	2·34 (15·5)	2·2 (14·7)	0 (0)	1·72 (11·5)	No	Yes Content not stated	Checked through empty capsules but not reported	Not assessed	Improved muscle mass and synthesis rate
Kim et al.[23] (Sarcopenic women)	(a) 6 g EAA + Ex (b) Ex (c) 6 g EAA (d) Con (3 months)	3 g powder twice daily	Not stated	0·63 (10·5)	2·52 (42)	0·84 (14)	Not stated	0·42 (7)	0·63 (10·5)	Not stated	0·63 (10·5)	Added to water or milk	No	Checked through record sheets but not reported	Not assessed but BMI maintained	Improved muscle mass and strength with Ex

EAA, essential amino acids; Ex, exercise component; Con, control (health education sessions); His, histidine; Ile, isoleucine; Leu, leucine; Lys, lysine; Met, methionine; Phe, phenylalanine; Thr, threonine; Trp, tryptophan; Val, valine.

supplementing HMB rather than leucine,[29,30] but only five examine outcomes relevant to sarcopenia[31–35] (see Table 3).

There is some interesting work in ageing rats which supports the hypothesis that HMB modulates negative age-related changes in body composition and function[36]. This group supplemented young (44 weeks) and old (86 weeks) rats with Ca-HMB for 16 weeks to produce middle-aged (60 weeks) and very old (102 weeks) rats, which were compared with baseline, middle-aged control and very old control data. The results showed a leaner, stronger body phenotype for those rats supplemented with HMB at both middle and very old age. Animal data are particularly useful in modelling ageing since longitudinal human studies are so expensive, difficult and time-consuming. Nevertheless, human intervention trials to demonstrate efficacy of HMB supplementation are required.

The results of the five studies examining sarcopenia relevant outcomes are more consistent than either EAA or whole protein, with three studies showing an effect on muscle mass[31,33,35] and the other two studies showing an effect for strength[32,34]. The details of the studies also show a more consistent approach. All included free-living older adults with exclusions for major illness, although two studies recruited from assisted living and care facilities[32,35]. All provided 2–3 g HMB/d, although two studies included additional amino acids, arginine and lysine, and vitamin C[32,35]. The rationale for this addition was that lysine is one of the three indispensable amino acids[37] and its requirement is potentially higher than previously thought; thus it was added to avoid an inadequate supply[38]. Both lysine and arginine, such as HMB, are also thought to be stimulants of muscle protein synthesis[9,39]. The format of the supplement however varied between studies; either as capsules or a powder, and the powder either mixed with water or other liquid. All studies used a placebo, which appeared adequately masked, although testing this was not done. Only one study used an iso-energetic and iso-nitrogenous placebo[35]; the others all using what appear to be iso-energetic placebos, although this is not clearly stated in the papers. Adherence was generally checked through measurement of urinary HMB, with expected increases in urinary excretion to demonstrate adherence, and in some studies this was backed up with logs or returned unused capsules. Only one study rigorously controlled voluntary dietary intake, including the protein intake to 0·8 g/kg body weight per d[33], but this study looked specifically at prevention of muscle loss during bed rest, rather than muscle gain in free-living populations. A second study monitored dietary intake with 3 d diet diaries[34].

The importance of assessing baseline nutritional status is effectively illustrated in a re-analysis of the Baier et al. study[35]. The symptoms of vitamin D deficiency are known to include muscle pain and weakness[40]. There is some evidence that there is a vitamin D receptor on muscle[41], and observational studies indicate a positive association between vitamin D and muscle function[42]. A recent systematic review confirmed that vitamin D supplementation can improve muscle

Table 3. Details of β-hydroxyl β-methylbutyrate trials to treat sarcopenia

Reference (population)	Intervention (duration)	Dose	HMB supplement	Placebo	Adherence	Dietary intake	Main result
Vukovich et al.[31] (free-living older adults)	(a) HMB and Ex (b) Placebo + Ex (2 months)	Twelve capsules/d (four caps × 3/d)	3 g/d One capsule: 250 mg Ca-HMB 50 mg potassium phosphate	Yes Rice flour	Returned caps every 10 d Elevated plasma HMB during study period 1·69 v. 67·73 umol/l	Not controlled but weight stable	Muscle mass improved for HMB group
Flakoll et al.[32] (free- and assisted-living older women)	(a) HMB, Arg + Lys (b) Placebo (No Ex) (3 months)	One 8 oz drink taken at breakfast	2 g/d Orange drink containing: 2 g Ca-HMB; 5 g Arg; 1·5 g Lys HCl; 0·5 g ascorbic acid	Yes Same vol, same flavour, iso-energetic. Maltodextrin; 0·5 g ascorbic acid ± 1·8 g non-EAA protein	None reported	Not monitored but weight stable	Performance and muscle strength improved for HMB group, but not muscle mass
Baier et al.[35] (free- and assisted-living older women)	(a) HMB, Arg + Lys (b) Placebo (No Ex) (12 months)	One sachet/d orange flavoured mixed with water	2 or 3 g/d depending on body weight >or <68 kg 2/3 g Ca-HMB; 5/7·5 g L-Arg; 1·5/2·25 g Lys HCl; 0·1 g ascorbic acid	Yes Iso-nitrogenous/energetic. 11·8 g non-essential amino acids (alanine 5·6 g, glutamic acid 0·9 g, glycine 3·1 g, serine 2·2 g) 0·1 g ascorbic acid	Monthly questionnaires and daily supplementation records Approximately 95 % over 12-month. Urinary HMB increased in HMB group	Not monitored but weight stable or increased	Muscle mass improved for HMB group but no changes for strength or function
Deutz et al.[33] (healthy older adults)	(a) HMB (b) Placebo (No Ex) (10 d)	Two sachets/d mixed with water	3 g/d One sachet contained: 1·5 g Ca-HMB; 4 g maltodextrin; 200 mg Ca; additional sweetener flavouring agents	Yes One sachet contained: 4 g maltodextrin; 200 mg Ca; additional sweetener flavouring agents	Urinary HMB increased in HMB group	Metabolically controlled 0·8 g/kg per d protein. Harris–Benedict for energy with different activity factors for bed rest	Preserved muscle mass during bed rest for HMB group
Stout et al.[34] (free-living older adults)	(a) Placebo (b) HMB (c) Placebo + Ex (d) HMB + Ex (6 months)	Two sachets/d mixed with water, milk or juice	3 g/d one sachet contained: 1·5 g Ca-HMB; 4 g carbohydrate	Yes 200 mg Ca; 4 g carbohydrate	Urinary HMB increased during study, Plus logs	Dietary recall x 3 d × 3	Improved muscle strength for HMB group

Ex, exercise component to the intervention; EAA, essential amino acid; HMB, β-hydroxyl β-methylbutyrate; Arg, arginine; Lys, lysine.

strength, in particular in those most deficient and those whose levels increase most[40]. Thus, it is possible that vitamin D status will influence the response to tests of muscle strength and performance. Fuller et al. re-analysed data to account for baseline vitamin D status[43]. They found that subjects who had a low vitamin D status (<30 ng 25OH-vitaminD₃/ml serum) did not respond to the HMB/Arginine/Lysine supplement to the same extent as the higher status subjects (≥30 ng/ml). Lean mass was increased in both vitamin D groups but muscle strength was only increased in the high vitamin D group, suggesting a synergistic effect of vitamin D and the supplement. Clearly, in future studies it will be critical to assess vitamin D status at baseline and supplement those who are insufficient.

There is one other study in bed-ridden older adults receiving artificial tube feeding for 6 months or more[44], which also shows an effect for HMB on nitrogen excretion and body weight and body circumference. The results are suggestive of a positive effect of HMB on muscle mass but this was not measured, neither were measures of strength or function.

Overall, these data are suggestive of a beneficial effect of HMB on older adults, but larger well-controlled studies are required that measure outcomes relevant to sarcopenia, ideally in sarcopenic populations.

Discussion of the evidence

The available evidence for protein does not allow us to make anything more than quite tentative suppositions. The most promising avenue does appear to be HMB supplementation but in the context of an otherwise adequate diet. However, the use of EAA is far from well-investigated and further work may offer other treatment options. It may also be that subjects with clearly defined sarcopenia respond differently to a generally healthy older population.

One other related issue is the timing and distribution of the dietary protein. Some researchers have suggested that there is a ceiling effect on the amount of protein used for synthesis at a given meal, and suggested an optimum protein intake of 20–30 g per meal[45,46]. Others have shown that feeding protein spread evenly throughout meals gives greater fractional synthesis rate compared with a skewed distribution[47], and that in free-living older subjects protein intake can be skewed towards a higher lunch intake, with less at breakfast and other meals[48]. However, other investigators refute the view that there is a maximal effect of protein at a meal, proposing a large bolus of protein should be as useful as providing the same amount spread over the day[49]. Others have provided evidence that pulse feeding protein (72–80 % provided at lunch) increases whole-body protein retention, lean muscle index (lean soft-tissue mass/height m^2) and appendicular skeletal muscle mass index (sum of lean soft-tissue in the four limbs/height m^2) compared with evenly spread protein[50,51]. Further research is evidently needed to better understand how protein distribution can affect optimum health in older people.

There is also the issue of increased splanchnic amino acid sequestration. The splanchnic bed includes the liver, stomach, intestines, pancreas and spleen (or portal-drained viscera). Because the portal-drained viscera receive nutrients from dietary digestion first, their requirements are satisfied first, potentially limiting the availability of nutrients, including amino acids, to peripheral tissues[52]. There is some evidence that splanchnic amino-acid sequestration is increased with age[53], and this could potentially complicate the supplementation of EAA to stimulate muscle protein synthesis and protein distribution throughout the day.

Protein may be the most obvious target for a treatment to prevent muscle loss but there are several other dietary strategies that have been explored, but require further investigation, such as vitamin D, alcohol, dietary acid–base load, fatty acids and antioxidants. These have all been recently reviewed by Welch[7].

Recommendations for the design of future trials

Cruz-Jentoft *et al.* make a series of recommendations in their review paper for the design of future nutritional studies. These include: clear well-defined populations, four arm studies nutrition with or without exercise, standardised outcome measures, attention to the timing of the nutritional supplement particularly in respect of exercise, and consideration of the baseline nutritional status and frailty of the study population[10].

In addition to these recommendations I would like to add that adherence to the nutritional intervention must be assessed as well as its effect on voluntary food intake, and if protein or amino acids are supplemented they are considered in the context of the subject's or group's present intake, with efforts made to improve poor intakes, as well as including an additional supplement. This of course is far more complex and is not as appealing as a simple daily drink, bar or capsule, yet if we are to successfully address the problem of sarcopenia and improve the health of the older population, it may be that a whole diet approach is required.

Conflicts of Interest

The author has previously received payment from Abbott Nutrition for acting as Chief Investigator for study: NCT01191125.

Authorship

The author was solely responsible for all aspects of preparation of this paper.

References

1. Chen L-K, Liu L-K, Woo J *et al.* (2014) Sarcopenia in Asia: consensus report of the Asian Working Group for Sarcopenia. *J Am Med Dir Assoc* **15**, 95–101.
2. Cruz-Jentoft AJ, Baeyens JP, Bauer JM *et al.* (2010) Sarcopenia: European consensus on definition and diagnosis: report of the European Working Group on Sarcopenia in older People. *Age Ageing* **39**, 412–423.
3. Fielding RA, Vellas B, Evans WJ *et al.* (2011) Sarcopenia: an undiagnosed condition in older adults. Current consensus definition: prevalence, etiology, and consequences. International working group on sarcopenia. *J Am Med Dir Assoc* **12**, 249–256.
4. Muscaritoli M, Anker SD, Argilés J *et al.* (2010) Consensus definition of sarcopenia, cachexia and pre-cachexia: joint document elaborated by Special Interest Groups "cachexia-anorexia in chronic wasting diseases" and "nutrition in geriatrics". *Clin Nutr* **29**, 154–159.
5. Dam T-T, Peters KW, Fragala M *et al.* (2014) An evidence-based comparison of operational criteria for the presence of sarcopenia. *J Gerontol A Biol Sci Med Sci* **69**, 584–590.

6. Morley JE, Abbatecola AM, Argiles JM *et al.* (2011) Sarcopenia with limited mobility: an international consensus. *J Am Med Dir Assoc* **12**, 403–409.

7. Welch A (2014) Nutritional influences on age-related skeletal muscle loss. *Proc Nutr Soc* **73**, 16–33.

8. Schiaffino S, Dyar KA, Ciciliot S *et al.* (2013) Mechanisms regulating skeletal muscle growth and atrophy. *FEBS J* **280**, 4294–4314.

9. Volpi E, Kobayashi H, Sheffield-moore M *et al.* (2003) Essential amino acids are primarily responsible for the amino acid stimulation of muscle protein anabolism in healthy elderly adults. *Am J Clin Nutr* **78**, 250–258.

10. Cruz-Jentoft AJ, Landi F, Schneider SM *et al.* (2014) Prevalence of and interventions for sarcopenia in ageing adults: a systematic review. Report of the Int. Sarcopenia Initiative (EWGSOP and IWGS). *Age Ageing* **43**, 748–759.

11. Bonnefoy M, Cornu C, Normand S *et al.* (2003) The effects of exercise and protein-energy supplements on body composition and muscle function in frail elderly individuals: a long-term controlled randomised study. *Br J Nutr* **89**, 731–739.

12. Bunout D, Barrera G, de la Maza P *et al.* (2001) The impact of nutritional supplementation and resistance training on the health functioning of free-living Chilean Elders: results of 18 months of follow-up. *J Nutr* **131** Suppl., 2441–2446.

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

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

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

16. Maher CG, Sherrington C, Herbert RD *et al.* (2003) Research report reliability of the PEDro scale for rating quality of randomized. *Phys Ther* **83**, 713–721.

17. Galloway DH (1975) Nitrogen balance of men with marginal intakes of protein and energy. *J Nutr* **105**, 914–923.

18. Bauer J, Biolo G, Cederholm T *et al.* (2013) Evidence-based recommendations for optimal dietary protein intake in older people: a position paper from the PROT-AGE Study Group. *J Am Med Dir Assoc* **14**, 542–559.

19. Deutz NEP, Bauer JM, Barazzoni R *et al.* (2014) Protein intake and exercise for optimal muscle function with aging: recommendations from the ESPEN Expert Group. *Clin Nutr* **33**, 929–936.

20. Fukagawa NK (2014) Protein requirements: methodologic controversy amid a call for change. *Am J Clin Nutr* **99**, 761–762.

21. Hubbard GP, Elia M, Holdoway A *et al.* (2012) A systematic review of compliance to oral nutritional supplements. *Clin Nutr* **31**, 293–312.

22. Dillon EL, Sheffield-Moore M, Paddon-Jones D *et al.* (2009) Amino acid supplementation increases lean body mass, basal muscle protein synthesis, and insulin-like growth factor-I expression in older women. *J Clin Endocrinol Metab* **94**, 1630–1637.

23. Kim HK, Suzuki T, Saito K *et al.* (2012) Effects of exercise and amino acid supplementation on body composition and physical function in community-dwelling elderly Japanese sarcopenic women: a randomized controlled trial. *J Am Geriatr Soc* **60**, 16–23.

24. Eley HL, Russell ST, Tisdale MJ (2008) Attenuation of depression of muscle protein synthesis induced by lipopolysaccharide, tumor necrosis factor, and angiotensin II by beta-hydroxy-beta-methylbutyrate. *Am J Physiol Endocrinol Metab* **295**, E1409–E1416.

25. Eley HL, Russell ST, Baxter JH *et al.* (2007) Signaling pathways initiated by β-hydroxy-β-methylbutyrate to attenuate the depression of protein synthesis in skeletal muscle in response to cachectic stimuli. *Am J Physiol* **293**, 923–931.

26. Stancliffe RA & Zemel MB (2012) Role of β-hydroxy-β-methylbutyrate (HMB) in leucine stimulation of muscle mitochondrial biogenesis. *FASEB J* **26**, 251–256.

27. Nissen S, Sharp R, Ray M *et al.* (1996) Effect of leucine metabolite β-hydroxy-β-methylbutyrate on muscle metabolism during resistance-exercise training Effect of leucine metabolite β-hydroxy-β-methylbutyrate on muscle metabolism during resistance-exercise training. *J Appl Physiol* 2095–2104.

28. Gallagher PM, Carrithers JA, Godard MP *et al.* (2000) Beta-hydroxy-beta-methylbutyrate ingestion, Part I: effects on strength and fat free mass. *Med Sci Sports Exerc* **32**, 2109–2115.

29. Fitschen PJ, Wilson GJ, Wilson JM *et al.* (2013) Efficacy of β-hydroxy-β-methylbutyrate supplementation in elderly and clinical populations. *Nutrition* **29**, 29–36.

30. Wilson GJ, Wilson JM, Manninen AH (2008) Effects of beta-hydroxy-beta-methylbutyrate (HMB) on exercise performance and body composition across varying levels of age, sex, and training experience: a review. *Nutr Metab* **5**, 1.

31. Vukovich MD, Stubbs NB, Bohlken RM (2001) Body composition in 70-year-old adults responds to dietary beta-hydroxy-beta-methylbutyrate similarly to that of young adults. *J Nutr* **131**, 2049–2052.

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

33. Deutz NEP, Pereira SL, Hays NP *et al.* (2013) Effect of β-hydroxy-β-methylbutyrate (HMB) on lean body mass during 10 days of bed rest in older adults. *Clin Nutr* **32**, 704–712.

34. Stout JR, Smith-Ryan AE, Fukuda DH *et al.* (2013) Effect of calcium β-hydroxy-β-methylbutyrate (CaHMB) with and without resistance training in men and women 65 +yrs: a randomized, double-blind pilot trial. *Exp Gerontol* **48**, 1303–1310.

35. Baier S, Johannsen D, Abumrad N *et al.* (2009) Year-long changes in protein metabolism in elderly men and women supplemented β-hydroxy-β-methylbutyrate (HMB), L-arginine, and L-lysine. *Nutrition* **33**, 71–82.

36. Wilson JM, Grant SC, Lee S-R *et al.* (2012) Beta-hydroxy-beta-methyl-butyrate blunts negative age-related changes in body composition, functionality and myofiber dimensions in rats. *J Int Soc Sports Nutr* **9**, 18.

37. Reeds PJ (2000) Criteria and significance of dietary protein sources in humans dispensable and indispensable amino acids for humans. *J Nutr* 1835–1840.

38. Rand WM & Young VR (1999) Statistical analysis of nitrogen balance data with reference to the lysine requirement in adults. *J Nutr* **129**, 1920–1926.

39. Beaumier L, Castillo L, Yu YM *et al.* (1996) Arginine: new and exciting developments for an "old" amino acid. *Biomed Environ Sci* **9**, 296–315.

40. Beaudart C, Buckinx F, Rabenda V *et al.* (2014) The effects of vitamin D on skeletal muscle strength, muscle mass and muscle power: a systematic review and meta-analysis of randomized controlled trials. *J Clin Endocrinol Metab* **99**, 4336–4345.

41. Simpson RU, Thomas GA, Arnold AJ (1985) Identification of 1, 25-dihydroxyvitamin D3 receptors and activities in muscle. *J Biol Chem* **260**, 8882–8891.

42. Visser M, Deeg DJH, Lips P (2003) Low vitamin D and high parathyroid hormone levels as determinants of loss of muscle strength and muscle mass (Sarcopenia): the Longitudinal Aging Study Amsterdam. *J Clin Endocrinol Metab* **88**, 5766–5772.

43. Fuller JC, Baier S, Flakoll P *et al.* (2011) Vitamin D status affects strength gains in older adults supplemented with a combination of β-hydroxy-β-methylbutyrate, arginine, and lysine: a cohort study. *JPEN* **35**, 757–762.

44. Hsieh LC, Chow CJ, Chang WC *et al.* (2010) Effect of beta-hydroxy-beta-methylbutyrate on protein metabolism in bed-ridden elderly receiving tube feeding. *Asia Pac J Clin Nutr* **19**, 200–208.

45. Walrand S, Short KR, Bigelow ML *et al.* (2008) Functional impact of high protein intake on healthy elderly people. *Am J Physiol Endocrinol Metab* **295**, E921–E928.

46. Symons TB, Schutzler SE, Cocke TL *et al.* (2007) Aging does not impair the anabolic response to a protein-rich meal. *Am J Clin Nutr* **86**, 451–456.

47. Mamerow MM, Mettler J, English KL *et al.* (2014) Dietary protein distribution positively influences 24-h muscle protein synthesis in healthy adults. *J Nutr* 876–880.

48. Bollwein J, Diekmann R, Kaiser MJ *et al.* (2013) Distribution but not amount of protein intake is associated with frailty: a cross-sectional investigation in the region of Nürnberg. *Nutr J* **12**, 109.

49. Deutz NEP & Wolfe RR (2013) Is there a maximal anabolic response to protein intake with a meal? *Clin Nutr* **32**, 309–313.

50. Arnal MA, Mosoni L, Boirie Y *et al.* (1999) Protein pulse feeding improves protein retention in elderly women. *Am J Clin Nutr* **69**, 1202–1208.

51. Bouillanne O, Curis E, Hamon-Vilcot B *et al.* (2013) Impact of protein pulse feeding on lean mass in malnourished and at-risk hospitalized elderly patients: a randomized controlled trial. *Clin Nutr* **32**, 186–192.

52. Stoll B, Burrin DG (2006) Measuring splanchnic amino acid metabolism *in vivo* using stable isotopic tracers. *J Anim Sci* **84**, 60–72.

53. Boirie Y, Gachon P, Beaufrere B (1997) Splanchnic and whole-body elderly men13 leucine kinetics in young and elderly men. *Am J Clin Nutr* **65**, 489–495.