dotFIT™ WheySmooth™

Goal

WheySmooth (WS) is designed to deliver nature's highest known biological value (BV) protein source, whey protein, with a BV of 104. Additionally, whey protein has a 100% Protein Digestibility Corrected Amino Acid Score (PDCAAS), which is a composite score indicator of protein quality used to determine the ability of protein to meet the body's amino acid requirements. PDCAAS considers the protein's essential amino acid (EAA) composition and digestibility. The protein extraction from the milk source used in WS is a concentrate, allowing greater overall health including immune support compared to other forms of whey protein because of the naturally occurring health and growth globulins contained in the whey portion of the dairy protein.

Proper use of WS helps conveniently increase dietary protein intake as needed, while simultaneously minimizing calories and eliminating unwanted food stuffs that often accompanies whole food protein sources for overall health, athletic training, and body composition goals. WS comes in a low-calorie powdered mix (chocolate, vanilla and unflavored) form which enables one to adjust the total protein and other nutrient content as desired, while remaining within their specific calorie needs based on fitness goals.

Because of whey protein's superior absorption and amino acid profile (specifically EAAs including leucine), the purpose of WS is to improve on the mechanisms of action related to muscle protein synthesis (MPS), diet and training outcomes when compared to other sources of protein. Therefore, gram for gram compared to other complete proteins, WS can: 1)improve lean body mass (LBM) gains or preservation and appetite control during fat/weight loss, leading to favorable body composition changes; 2)maximize MPS especially timely as needed (peri-workout), which may also optimize muscle hypertrophy and performance; 3) allow more protein (EAA) with fewer calories to assist in lifelong weight control, while also staving off inevitable age-related muscle loss; 4) deliver other potential health benefits, including immune system support; 5) in its native high protein, low calorie powdered form, including all-natural and unflavored versions, it can serve as the starting ingredients for the user to add as desired (e.g. fruits, vegetables, dairy, etc.) to complete a healthy meal/shake – i.e. serves as a tasty delivery system to include foods not consumed regularly. And finally, WheySmooth's accompanying ingredients allows for easy mixing and is ideal for baking.

Rationale

The constituents of whey protein including its comparatively higher leucine and other essential amino acid amounts per gram of protein along with the natural health contributing bio-actives contained in the concentrate, make whey protein sources reign premier in supporting muscle protein synthesis and body composition goals. Further, the digestibility, absorption, and amino acid retention (muscle deposition) score compared to other popular protein sources, validates whey's benefits and therefore, when protein supplementation is included to meet individual recommendations in maximizing exercise/performance outcomes or daily life recovery, whey protein is a first choice when diet restrictions do not impede the selection.

Background

Like vitamins and essential minerals (VM), dietary protein and its constituent amino acids (AA) are indispensable to the creation, development, and maintenance of life.[1] Dietary protein once ingested, is broken down to its constituent AA and can be reformed into the thousands of specific human bodily proteins, each of which is uniquely designed to accomplish specific tasks.[2] The AA also act as signaling molecules to regulate their myriad functions, including but not limited to, hormone and neurotransmitter formation, act as fuel and contribute to energy production when needed (e.g. supply intermediates in the Krebs Cycle and gluconeogenesis), and muscle protein synthesis and breakdown (MPB) modulation, with the MPS being the focus of this review paper.[3,4]

20 proteogenic L-amino acids are the building blocks for protein synthesis, all vital to life and health.[1,5] The AA are classified as (see Table 1): essential amino acids, because these nine AA must be supplied by exogenous sources (e.g.

diet, supplements) to begin and sustain life and health. The remaining 11 AA, although indispensable to life, are non-essential amino acids (NEAA) because they can be produced from other substances/AA in the body. Six of these NEAA are considered to be conditionally essential, meaning under times of stress (caloric restriction, illness, injury, etc.) the body cannot produce them in high enough quantities to properly support health and daily recovery.[1,6] Although there are often minor disagreements on one or two of the NEAA, such as tyrosine and serine that should be labeled "conditional", it is of little consequence since all can be manufactured within the body at some level as long as substrates are available (standard diet), and the more important EAA are clearly defined and agreed upon.[1,2,5,6]

Table 1 - Twenty Proteogenic Amino Acids Necessary For Protein Synthesis[1,2,5,6]

Essential Amino Acids[a]	Conditionally Essential[b]	Non-Essential[c]
Histidine	Arginine	Alanine
Isoleucine	Cysteine	Asparagine
Leucine	Gluamine	Aspartate
Lysine	Glicine	Glutamate
Methionine	Proline	Serine
Phenylalanine	Tyrosine	
Threonine		
Tryptophan		
Valine		

[a]Must be consumed because the body cannot manufacture them at all or in sufficient quantites to support life
[b]Required to be consumed to some degree during during growth and development, stress, caloric restriction or illness
[c]Can be synthisized in sufficient amounts provided that necessary building blocks and enzymes are availble

Among the many structural and functional tasks performed by protein's NEAA and EAA, they are responsible for skeletal muscle creation, development and maintenance with the EAA being the most important, not solely because mammals must acquire them from exogenous sources and their respective tissue distribution, but EAA are the primary signaling molecules that trigger MPS (and MPB) and maintain AA homeostasis.[3,4,7,8] Therefore, it's understood that protein sources, plant or animal, with the greater EAA count and composition that is closest to human needs per gram of protein, along with higher ratings of digestibility, absorption and retention, are scored as proteins of the greatest value.[9,10] As displayed in Table 2 and 3, whey protein scores highest, which currently makes whey protein the most popular supplemental protein for supporting MPS.[11,12]

Table 2 - Protein Quality Assesment Based on Human Needs

Source: Berrazaga, et al.[11]

Protein Type	Protein Digestibility (%)	Biological Value (%)	Net Protein Utilization (%)	PDCAAS	DIAAS
Animal source					
Red meat [1]		80	73	92	
Casein [1,3,6]	99	77	76–82	100	
Whey [1]		104	92	100	
Milk [1,4,6]	96	91	82	100	114
Egg [1,4,6]	98	100	94	100	113
Plant source					
Black bean [1,6,8]	70			75	
Cooked black bean [7,8]	83			65	59
Soy flour [5,8]	80			93	89(SAA)
Soy protein isolate[1,6]	98	74	61	100	
Green lentil [3,4]	84			63	65
Yellow split pea [4,6]	88			64	73
Cooked pea [7]	89			60	58
Pea protein concentrate [7]	99			89	82
Chickpea [3,4]	89			74	83
Peanuts [1]				52	
Roasted peanuts [7]	98			51	43
Peanut butter [3,4]	98			45	46
Whole grains [2]				45	
Wheat [3,5,6]	91	56–68	53–65	51	45(Lys)
Wheat gluten [1]		64	67	25	
White bread [4,6]	93			28	29
White rice [4,6]	93			56	57
Cooked rice [7]	87			62	60

[1] Hoffman and Falvo [52]; [2] van Vliet et al. [53]; [3] Sarwar et al. [54]; [4] Marinangeli and House [55]; [5] Mathai et al. [56]; [6] ANSES [57]; [7] Rutherfurd et al. [58]; [8] Sarwar [59]. Abbreviations: PDCAAS: protein digestibility-corrected amino acid score; DIAAS: digestible indispensable amino acid score; Lys: lysine; SAA: sulfur amino acids.

The Protein Digestibility Corrected Amino Acid Score (PDCAAS) is a composite score indicator of protein quality used to determine the ability of protein to meet the body's AA requirements.

It factors the protein's EAA composition and digestibility.

A given dietary protein cannot fully meet the body's EAA requirements when its PDCAAS is less than 100%.

Table 3 - Comparative Essential Amino Acid Scores of Plant and Animal Based Protein Sources

Sources: Berrazaga, et al.[11] and adapted from Laleg et al.[13] and Witard et al.[14]

	Plant-Based Proteins						Animal-Based Proteins			
	Wheat	Maize	Soybean	Pea	Faba Bean	Lentil	Whey	Casein	Milk	Beef
	Essential amino acid scores (%) [1]									
Histidine	140	187	173	167	231	176	127	180	180	240
Isoleucine	137	127	157	153	112	154	213	167	170	167
Leucine	115	219	136	125	121	132	168	151	161	144
Lysine	31	62	147	182	158	160	204	169	153	207
Methionine + Cysteine	120	127	91	73	79	91	130	125	134	157
Phenylalanine + Tyrosine	290	300	277	267	247	263	227	343	313	280
Threonine	109	161	174	191	156	165	291	187	174	209
Valine	108	128	126	131	95	135	162	162	159	133

[1]Scores are calculated based on recommendations for a healthy adult human[15]

Daily Protein Requirements

There are too many variables to secure a magic number for everyone, but more than enough data exists to construct a blanket recommendation of daily protein for mostly healthy persons who desire to develop, increase or maintain muscle or stave off the inevitable loss in aging to prolong health and independence. Collective modern research points to 1 gram per pound of lean body mass (LBM) distributed throughout daily meals as a safe and effective daily protein intake that can support MPS for all age groups to: optimize growth and development, maximize exercise induced muscle gains and performance, reduce LBM losses during calorie restriction, overcome age related anabolic resistance to extend the ability to maintain a positive MPS balance while staving off the inevitable aging loss of muscle to help remain active and independent throughout a lifetime. Meeting the current expert recommendations for different sub-populations often requires convenient supplementation with whey protein-based formulas being the common suggestion because whey has the highest protein scores and potential efficacy in meeting user's needs, especially as it relates to MPS.

Although the Recommended Dietary Allowance (RDA) of protein has stubbornly remained constant at 0.8 g/kg (.36 g/lb.) of body weight for many years (a range from 10-35% of total calorie intake is considered safe),[16] the recommendation is largely ignored by the general population and more often challenged as insufficient by scientists who study best practices in specialized areas such as sports, exercise, weight/fat loss and aging.[17,18] Further, while most of the US population (and other developed western societies) meets or slightly exceeds the protein RDA (or minimum requirement), protein intake, as a percentage of energy intake, remains well below the upper end of the Acceptable Macronutrient Distribution Range (AMDR) published in the Dietary Guidelines for Americans (DGA).[16,19] Therefore, dietary protein recommendations or suggestions for best health, fitness and sport related outcomes have diversified over the years into more specific recommendations for sub-populations such as athletes,[20,21] exercisers,[22,23,24] dieters,[25,26,27] and the expanding aging population.[12,28,29,30,31,32]
While the protein RDA may support nitrogen balance in a typical healthy but predominately sedentary youth through young adulthood, significantly higher levels have been found to be more effective for: exercise, injury recovery, overall dieting (calorie restriction) and weight control results, exercise-induced muscle hypertrophy and performance gains, and improving age-related muscle function while reducing losses from the natural ageing process.[7,11,12,17,18,20,21,22,23,24,25,26,27,28,29,30,31,32,33,34] The surge in attention around protein and the benefits of its constituent AA sparked the beginning of a paradigm shift in overall diet research, concepts and preliminary recommendations and appears to have accelerated following the Protein Summit 2.0 in Washington DC.[28,35] Additionally, higher protein intakes (2-4 times the RDA or 20-35% of total calories) in healthy individuals are for the most part no longer considered potentially harmful, and in fact using lean sources may be helpful in specific areas[24,33,36,37,38,39,40] as described above. AA consumed well above the protein RDA appears quite safe.[28,33,41,42] The Tolerable Upper Limit (ULs) for the few AA studied, are generally three to five fold greater than typical intakes in the United States, making it highly unlikely for people to surpass this daily amount.[43]

Whey Protein Supplementation

While all complete protein sources (e.g. meat, dairy, fish, egg, soy, etc.) may deliver the AA necessary for basic protein synthesis, each protein source has other unique bio-active contributions based on overall structure.[44,45,46,47,48,49] Therefore, they are commonly compared in studies to determine which protein source is best for specific health, sport and fitness outcomes[50,51,52,53,54,] – i.e. most bang for the buck, which often means the greatest results with the fewest calories in order to achieve and maintain a desired body composition during all stages of life and fitness goals.[52,53,54,55,56,57] As captured above, whey proteins, which are extracted from milk protein, are generally considered the superior human protein source especially as it relates to muscle protein synthesis[52,58,59] because of its rapid absorption rates[51,60,61,62] and constituents including its high leucine content[15] and other EAA.[52,53,54,55,56,57,63,64,65] Therefore, in summary, as supplementation is needed for meeting the current specific field expert recommendations including timing around activities as described throughout this review, whey protein concentrate because of its EAA content, absorption and muscle retention rates, and natural health contributing bio-actives, would be a preferred choice to improve recovery, health, fitness, and performance outcomes when compared with other sources.

Milk Proteins

The main constituents of milk are considered functional foods, with direct impact on human health.[63,66] Milk has two primary 'fractions' of proteins, casein and whey. These fractions are further sub-divided as: four caseins (CN), α_{S1}-, α_{S2}-, β- and κ-CN, and two primary whey proteins, α-lactalbumin (α-LA) and β-lactoglobulin (β-LG), that collectively account for approximately 90% of all milk protein fractions.[67,68] Whey is the liquid portion making up approximately 20% of the total protein content of bovine milk with casein being 80% (human milk is 60/40, respectively).[69,70] Bovine whey is composed of β-lactoglobulin (50%–60%), α-lactalbumin (15–25%) and minor contributions of bovine serum albumin (BSA, 6%), lactoferrin (<3%) and immunoglobulins (<10%).[70]

Processing to Produce Whey

Processing, such as ultra-filtration (UF) and microfiltration create different whey protein products. The most utilized whey proteins include concentrate (35-90% protein, with or without lactose), isolate (~90-95% of protein, normally without carbohydrates, cholesterol and other whey fractions), hydrolyzed (smaller peptide fractions that are considered less allergenic but costly), and non-denatured (native protein structures).[71,72] Whey protein concentrate (WPC) powders with protein contents as high as 85%, such as the source used in WheySmooth, is produced by direct ultrafiltration to remove components such as lactose and non-protein nitrogen, and diafiltration to wash out the final unneeded lower molecular components – i.e. virtually all lactose and unwanted minerals, which now pass through the membranes.[72,73] (Note: casein protein has its own unique properties but requires longer digestion than whey leading to a delayed and more prolonged absorption.[50,51,74])

Forms of Whey (What is the 'best" form of whey proteins?)

All three common forms of whey protein, WPC, isolate, and hydrolysates are all used in positive clinical trials, often specific to their properties but at a minimum they all contain the same EAA profile best for MPS.[52,75] WheySmooth uses WPC because WPC not only includes the same EAA necessary for MPS, it also contains other bio-active health, immune and growth factor components including the minerals calcium, sodium, phosphorus, and potassium; proteins including alpha-lactalbumin, beta-lactoglobulin, lactoferrin, serum albumin, lysozyme; immunoglobulins A, G, and M; and cysteine,[76] all which may have positive impacts on human health.[34,63,66,71,77] Further, WPC has been shown to be more effective in controlling or reducing fat mass when compared to whey isolate and hydrolysate.[47] While the process of creating whey isolates (and hydrolyzed whey) eliminates some of the bio-active components of whey named above, isolates or hydrolysates/peptides fully support MPS and have a clinical or allergen application.[78,79] The other substances naturally found in WPC may be contraindicated for unique dairy allergies, high cholesterol, or other health reasons. dotFIT LeanMR uses whey isolates for this purpose.

Manufacturing Processes & Marketing Hype

As discussed, WPC and isolates go through a process to remove most of the carbohydrates, fat and lactose from regular unprocessed whey from whole milk. Both result in an almost pure protein with the isolate minus the other health contributors.71[,72,73] The protein in WheySmooth is 90% WPC (80% protein), 5% whey isolate (90% protein) and 5% casein (90% protein) and contains negligible lactose.[80]

Marketing Hype: the advertising of filtering processes in producing whey products such as WPC, isolates, etc. are marketing spins. The desired outcome (supply high content of EAA and/or growth factors for recovery, muscle building and health) will be the same when processing proceeds as properly outlined in manufacturing guidelines for dairy products.[72,73] Examples of marketing hyperbole include "fairy tales" around the terms cold filtered, ultra-filtration, ion-exchange, micro-filtration, cross-flow filtration, etc. Nutrition research scientists who are not under contract with specific supplement manufactures agree that currently these terms are all basically the same as described above.

Marketing talking points about filtration methods is hype that only confuses us all and makes little to no difference in the MPS value of the whey protein.[52,71,72,73]

Summary of Protein in WheySmooth

Based on efficacy and desired total contents, WS uses an UF, diafiltration, ion-exchange instantized protein blend containing 90% whey concentrate, 5% whey isolate and 5% casein for immediate and extended release and easy mixing. There is only trace (1.4 g) of lactose per serving and therefore unless you have been diagnosed with "severe lactose intolerance," which is rare,[81] this amount should have no adverse reaction.* There is a normal reduction in lactase (enzyme to breakdown lactose for absorption) in childhood ascribed to be an evolutionary trait necessary to facilitate weaning.[82] Most lactose mal-digesters (lactose malabsorption from natural lactase non-persistence) and individuals who consider themselves lactose intolerant, can consume 6-12 grams of lactose in one feeding without major symptoms. As a reference, one cup of milk or yogurt has 12 and 9 grams of lactose, respectively.[83]

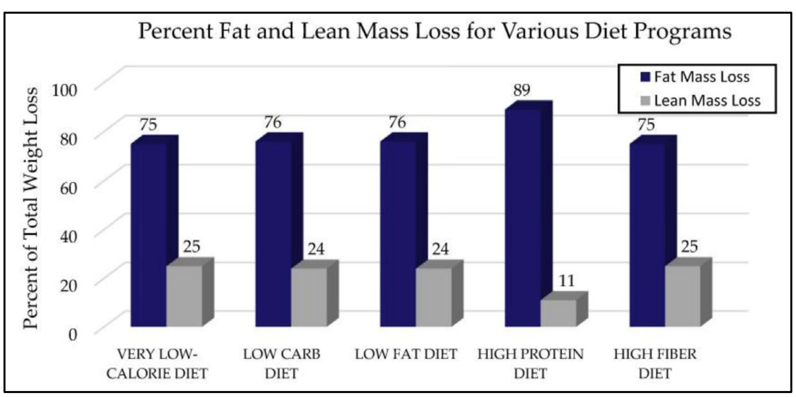
***Most individuals with lactose malabsorption tolerate a dose of at least 12 g of lactose (corresponding to 250 mL of milk) without problems. Larger doses may be tolerated if consumed with food or spread over a whole day.**[83]

Protein Intake in Weight/Body Fat Loss

Loss of lean body mass (LBM) is an undesirable and mostly unavoidable consequence of conventional weight loss practices. Not solely because of the misery incurred as the human body mounts its natural defenses (e.g., increase in appetite, decrease in energy and metabolism, etc.) to continuous losses of evolutionary driven perceived hard earned body mass, which most often eventually leads to surrender and weight regain, but diet induced loss of LBM is strongly associated with the weight regain phenomenon. Higher protein diets in a dose-dependent manner (25-50% of total calorie intake) have been shown to ameliorate the body's weight loss defense actions including supporting LBM and forcing greater losses of body fat during weight reduction attempts.

Higher protein diets (25-50% of total calories or significantly greater than the RDA) which include low/moderate fat and/or low carbohydrate are generally more successful for weight loss than lower protein diets, at least in the short term, especially in terms of protecting LBM (see Figure 1 below from Willoughby et al.[84]).[85,86,87,88,89,90] The basic mechanisms of action include greater satiety, increased daily energy expenditure (including thermic effect of food [TEF]), fat oxidation,[91,92,93,94,95,96,97,98] and preservation of lean body mass (LBM).[22,86,98,99,100,101,102,103] The latter arguably being protein's most important action since loss of LBM not only compromises the body's structure, functions, and total energy expenditure, but greater losses of fat free mass (e.g. muscle, bone, other organs[88,104,105,106,107,108]) are strongly associated with weight regain and appetite.[109] Of all these actions of protein, whey proteins compared to other sources appear to deliver superior outcomes when integrated into daily meal planning.[65,91,101,110,111,112,113,114]

Figure 1 – Fat Mass vs. Lean Mass Loss in Various Diets (Source: Willoughby et al.[84])



Whey Protein in Weight Loss

Whey protein appears to have greater influence on satiety,[56,91,110,111,112,113,115] MPS, LBM preservation[56,63,65,116,117,118] fat oxidation, body composition,[47,56,57,59,65,91,118,119] and health when compared to other protein sources.[114,120] Much of whey's added value may be due to its EAA structure including high leucine content and rapid amino acid absorption rate.[63,64,65,112] Whey protein compared to other protein sources such as soy, red meat/steak, chicken, etc., has a relative significant greater amount of leucine per gram or protein.[11,13,14,15,65] 25 grams of whey protein contains three (3) grams of leucine whereas soy has 1.4 grams, casein contains 2.3 grams and most meats contain even less.[52] Scientific data suggests that at least 2.5 grams of leucine may be the turning point for benefits when it comes to protein synthesis.[65,121,122,123] Xu ZR et al. found that leucine supplementation alone is useful to address the age-related decline in muscle mass in elderly individuals because it increases the muscle protein fractional synthetic rate,[124] but it appears that a leucine fortified whey protein is even more effective in supporting MPS, suggesting that leucine supplementation alone has a MPS trigger threshold based on the need for the supporting factors of the other AAs.[123,125,126,127] In other words, the available remaining complimentary EAAs and/or NEAAs would be the limiting factor in leucine's potent MPS actions.[125,126,127,128,129] For more details on whey protein mechanisms of actions in supporting weight loss, the practitioner is referred to the LeanMR document here.

WheySmooth in Weight Loss

In its native form (starting mix formula), WS is a low calorie, high anabolic protein source with cofactors to support protein's function and taste. The nutrition profile of the mix makes it flexible in matching it to the user's goal because the user can adjust the protein, fat, carbohydrate, and calories as necessary, including adding other foods and ingredients to meet fitness goal requirements while keeping calories under control. One serving (scoop) of WS contains 25 g of protein, 7 g of carbohydrate, 3 g of fat, 200 mg calcium, 224 mg potassium in only 160 calories; mindful that you can adjust the serving size to fit your needs – i.e., 1.5 scoops is ~40 g of protein in 240 calories. WS is commonly used to meet the higher protein within lower calorie requirements necessary for protecting LBM, supporting appetite etc., during weight/body fat reduction including being incorporated into a meal replacement/substitute weight control strategy.

WheySmooth and Meal Replacements in Weight Control

Proper use of meal replacements during calorie restriction is considered one of the most effective treatments for weight control for both initial weight loss and maintenance.[98,130,131,132,133,134,135,136,137,138,139,140] Based on the definition of a food product labeled as a "Meal Replacement,"[141] WS is purposely formulated to not meet that criteria by itself, allowing it to be a single product to satisfy multiple goals (see summary at end of document), including serving as the high protein, low calorie starting nutrition mix for the user to create an individualized meal replacement or substitute to support weight control. In other words, WS can be used as a complete meal replacement by mixing in other foods to reach desired macronutrient ratios and calories. The excerpt on meal replacements and weight loss below, "*Successful use of Meal Replacements within the Daily Meal Planning,*" is taken from the LeanMR section of the Practitioner Dietary Supplement Reference Guide with all related references.

Additionally, WS can simply serve as a daily whey protein source to enhance weight loss outcomes as described above. One serving adds 25 g protein and 3 g leucine with only 7 g of carbohydrate to help achieve desired levels of daily protein, including leucine, during weight loss. Suggested protein daily intake during normal weight loss (which includes performing some form of physical activity) to deliver described benefits should be approximately 0.8-1.0 g per pound of body weight (or 1 g/lb of LBM) spread evenly across four to five meals; [18,23,24,25,26,27,34,142,143,144] and greater if weight loss is aggressive – i.e. the larger the caloric deficit, the greater the need for protein to support LBM.[18,21,22,23,27,39,84,116,117,118,119, 145,146,] Further, as referenced above, whey protein has demonstrated better body composition outcomes than other protein supplementation and therefore, WS is structured to meet this criteria and serve as an ideal complement to the daily traditional food diet to reach this protein level within the calorie allotment for the desired rate of weight/bodyfat loss.

Successful use of Meal Replacements within the Daily Meal Planning

Overall Diet

Taken in totality for the goal of weight loss and subsequent maintenance, as referenced above, science favors a whey protein mix-type “meal replacement” (or substitute) to be integrated and complement a high protein calorie restricted diet containing 30-40% carbohydrates, 30-35% protein and the remainder dietary fat (always maintaining a minimum of 1 g of protein/lb./LBM/day). In other words, daily menus containing traditional food meals, with protein in each meal, and the inclusion of controlled calorie high protein-based mixes to complete the allowed caloric allotment, has been validated as a top successful strategy to support weight/fat loss while protecting LBM and reducing the likelihood of weight regain. Further, using the category “meal replacement” for a malleable protein mix in the context of inclusion (or addition) to traditional food meals versus an actual replacement, is often a misnomer depending on formulation and how it’s used within daily meal planning, since properly prepared meal replacements (MRs) are food products, thus actually a meal offering potentially more nutritious daily meals within the daily goal calories.

Meal Replacement Integration

Weight Loss Phase:

- Except in the early stage of diets when meal replacements may be used extensively in daily meal planning (often physician monitored and sole/predominant food source),[137,138,139,147] they are generally used to replace two meals a day and allow freedom of choice from traditional foods for the remaining allotted foods/calories.[132,133,134,135,136,137,138,140,148]
- Meal replacements may supply two small meals within any calorie restricted meal plan of four to five meals/snacks since it's been shown that frequent, smaller meals are generally better for weight loss than fewer larger ones, particularly as it relates to satiety, preservation of LBM and energy levels.[18,23,24,25,26,27,136,137,138,142,144,148,149,150,151]

Maintenance Phase

- Consume the required calories for maintenance spread between four to five meals/snacks daily which may include two meal replacements for convenience and to help ensure overall diet quality while reducing food costs.[144,148]

Protein in Hypertrophy

The 20 AAs contained in complete protein sources are required for muscular growth and development. 8 of the 9 EAAs are the signaling molecules necessary to trigger MPS while the remaining AAs are needed to complete and prolong the process. Animal proteins are superior to plant sources in supplying the AAs necessary for human muscle development with whey protein rated at the top, primarily because of whey protein’s digestibility and EAA content per gram of protein. 1 gram of protein per pound of LBM divided and timed properly throughout the day including exogenously supplied EAA exposure in close proximity to exercise, has been scientifically proposed to maximize muscle hypertrophy, while minimizing body fat stores. Further, the amount of protein per meal, regardless of the ambiguous “muscle full effect,” (how much skeletal muscle can use/incorporate from one feeding) to maximize MPS (net protein balance) should be primarily determined by body weight or LBM and suggested to be .18-.25 g/lb of LBM (.18-.25 g/lb of body mass). Whey protein, because if its digestibility rating and greater EAAs content per gram of protein/calories, is a preferred supplement to help accomplish the protein requirements for maximizing exercise-induced muscle hypertrophy unless an individual’s diet restrictions prohibit its use.

Exercise and Protein (Amino Acids)

Without exogenous protein and its constituent AAs, hypertrophy cannot take place at any stage in life. [18,24,45,152] Maximizing skeletal muscle hypertrophy requires regular unaccustomed exercise combined with proper overall nutrition[45,46,153] that includes frequent daily protein (amino acids) feedings[17,18,21,24,33,143,144,154] and daily totals of approximately 1g of protein per pound of LBM (or body weight) or more depending on energy balance.[7,11,12,17,18,20,21,22,23,24,25,26,27,28,29,30,31,32,33,34,22,25,145,146,155] Meeting protein needs, including timing and desired body composition calorie needs, has established protein supplementation as a common, safe, and effective practice for enhancing exercise induced muscle size and strength in healthy humans of all ages.[120] The Morton et al. systematic review and meta-analysis looked at 49 studies with 1,863 subjects and showed that protein supplementation compared to placebo significantly increased changes in: strength (one-rep maximum), fat free mass (FFM) and muscle size—muscle fiber cross-sectional area (CSA) and mid-femur CSA during periods of prolonged resistance training.[120]

Mechanisms of Action

Exercise and amino acids stimulate skeletal muscle protein turnover independently, thus when combined properly they have profound additive effects on recovery, performance and muscle size.[45,46,152,153] Exercise is an event that can trigger a desired result based on the body parts incorporated and activity type intensity/duration performed (e.g. contraction mode, power, endurance, planes of motion, etc.),[156,157] and although mechanical stresses from exercise activate their respective channels of MPS signaling, muscle protein balance remains negative without subsequent feeding of AAs, thus exercise alone would continually decrease muscle size and performance.[18,24,25,45,46,152,158,159] Exercise and amino acids stimulate skeletal muscle protein turnover by affecting the activity of intracellular signaling networks such as the mammalian target of rapamycin complex 1 (mTORC1) and the mitogen activated protein kinases (MAPK) cascades.[160] Exercise-induced skeletal muscle hypertrophy directly correlates with mTORC1 activation, with subsequent increases p70S6K (a mitogen-activated Ser/Thr protein kinase that is required for cell growth) and rpS6 phosphorylation.[161,162,163] Figure 2 from Condon et al.,[164] displays the complex interactions of the nutrition and hormonal signaling pathways in MPS. Figure 3, in a simplified view from Drummond et al,[165] shows how the diet derived AAs, together with exercise, potentiates an exaggerated MPS response, first initiated through mechanical loading (contractions) enhancing mTORC1 activation and other intracellular AA sensing mechanisms such as MAPK (e.g. extracellular signal regulated kinase 1 and 2 [ERK1/2], c-jun NH2-terminal kinase [JNK], etc.), and the human vacuolar protein sorting-34, (hVps34),[4,166,167,168,169,170,171] and also by increasing AA transporter expression.[172,173,174]

Figure 2 - Interactions of the nutrition and hormonal signaling pathways in MPS (Source: Condon et al. [164])

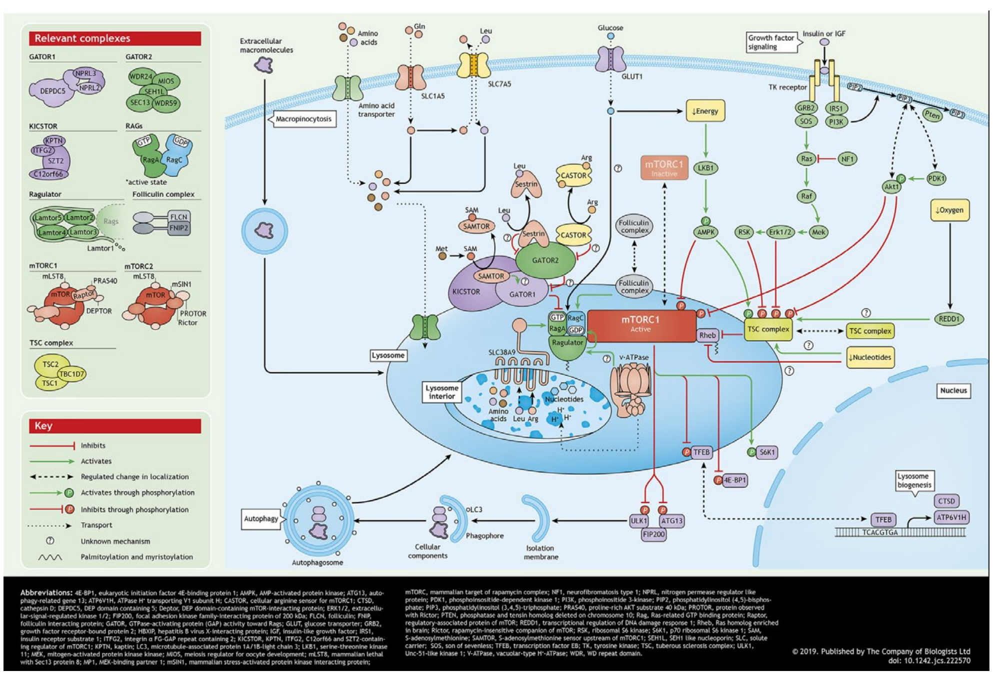
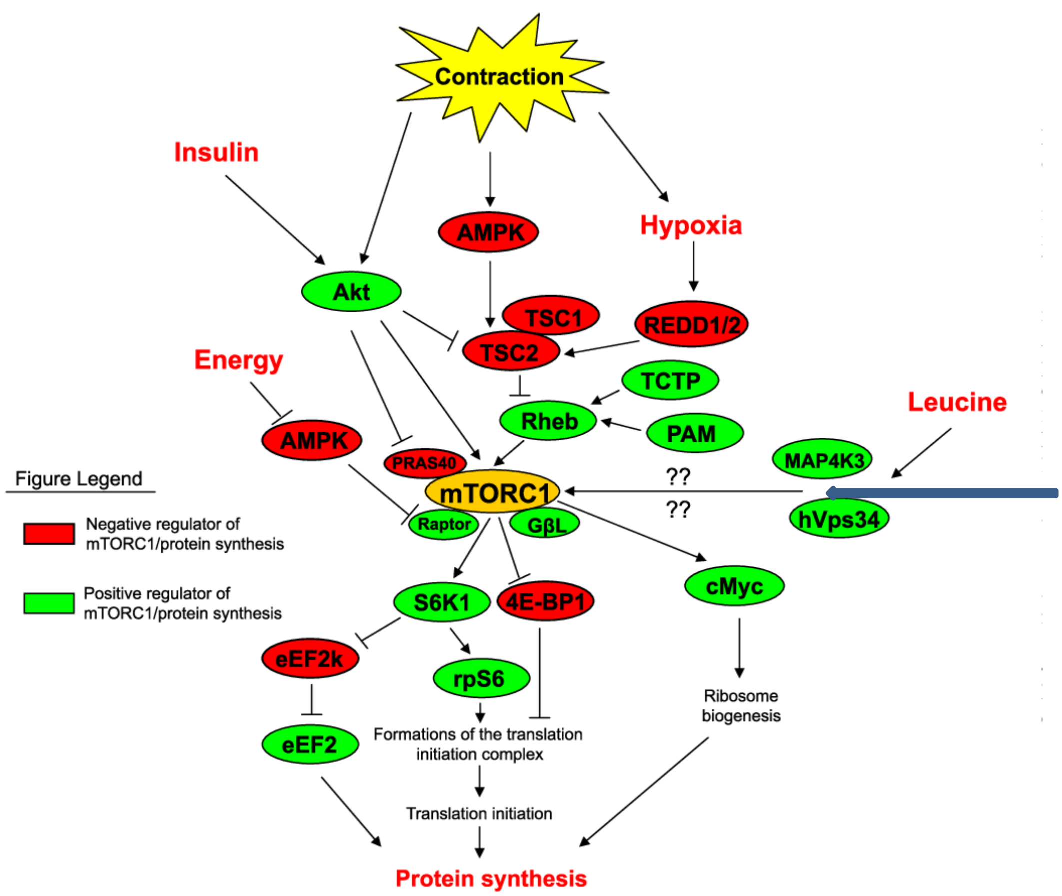


Figure 3 – The mTORC1 signaling pathway is driven by muscle contraction, insulin, essential amino acids (especially leucine) and energy supply and shows the positive and negative influencers of MPS when diet and exercise converge. Source: Drummond et al.[165]



8* of the 9 EAAs, primarily leucine, are the triggering molecules for MPS:

- Leucine
- Valine
- Isoleucine
- Phenylalanine
- Lysine
- Threonine
- Histidine
- Methionine

Abbreviations: AMPK, AMP-activated protein kinase; Akt, protein kinase B; TSC1, tuberous sclerosis complex 1; TSC2, tuberous sclerosis complex 2; REDD1/2, regulated in development and DNA damage responses; Rheb, Ras-homologue enriched in brain; TCTP, translationally controlled tumor protein; PAM, protein associated with Myc; Raptor, regulatory associated protein of mTOR; G L, G protein -subunit-like protein; MAP4K3, mitogen activated protein kinase-3; hVps34, human vacuolar protein sorting-34; S6K1, p70 ribosomal S6 kinase 1; 4E-BP1, 4E binding protein 1; eEF2k, eukaryotic elongation factor 2 kinase; eEF2, eukaryotic elongation factor 2; rpS6, ribosomal protein S6; PRAS40, proline-rich Akt substrate-40.

Figures 2 and 3 also show the AMP-activated protein kinase (AMPK) role as an energy sensor, as AMPK activation suppresses MPS. Skeletal muscle during exercise increases the use of ATP (turnover may increase >100 fold) resulting in the accumulation of adenosine monophosphate (AMP) thus affecting the cellular AMP/ATP ratio causing the activation of AMPK.[175] Therefore AMPK is a sensor of intracellular energy status and works to maintain stores for cell survival by regulating anabolic and catabolic pathways including MPS and muscle protein breakdown (MPB) as necessary. Further, exercise intensity and duration regulates different AMPK heterotrimer complexes leading to different functional responses.[176] In short, mTORC1 regulates skeletal muscle by controlling protein translation initiation through its two major downstream targets: p70 ribosomal S6K and the eukaryotic initiation factor 4E binding protein 1 (4E-BP1).[177] mTORC1 activity is regulated by energy sensing AMPK through phosphorylation of tuberous sclerosis complex 2 (TSC2) at Thr1227 or Ser1345, which improves the ability of TSC2 to inhibit mTOR activity as necessary.[4,176,178] The interplay between mTORC1 and AMPK is tightly regulated to support maintaining exercise

induced muscular energy, activity and development and highlights the need for adequate energy and protein (AA, especially EAA) to maximize hypertrophy and performance.[4,160,161,162,163,164,165,166,167,168,169,170,171,172,173,174,175,176,177,178]
In summary, mTORC1 is the primary complex that determines muscular growth via sensing the cellular contents before initiating the building processes. The presence of AA, specifically the EAA, regulates the activation of mTORC1. Further, exercise leads to MPB, which induces a heightened nutrient demand and their respective receptors sensitivity, allowing exogenous AA, when delivered in an energy rich environment and *timely in the right amounts* (specifically EAA including a relatively high leucine content), to maximize an individual's MPS potential, leading to enhanced size and/or performance training-induced results when all else is equal (e.g. training and overall diet).[128]

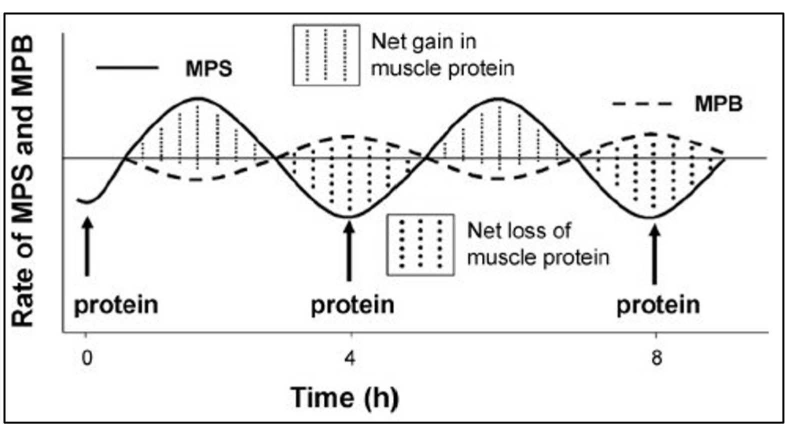
Essential Amino Acids

The stimulation of MPS from AA is dependent on the EAA content.[179] Although non-essential amino acids are necessary components of complete muscle tissue, they are not required to stimulate MPS.[44,180,181,182] Additionally, it has been shown that EAA feedings can stimulate protein synthesis independently or incrementally to intact protein alone – i.e. can have an additive effect on MPS.[46,128,183,184,185]

Protein (amino acid) Timing

Muscle protein dynamics (breakdown and synthesis) involves approximately four hour cycles (Figure 4) in which following digestion of a protein rich meal, synthesis is greater than breakdown but returns to baseline within four hours at which time breakdown begins to exceed synthesis until another protein meal is consumed and digested.[45,146,186,187,188] Therefore in exercisers/athletes, it has been long proposed that to maximize MPS, a person should consume protein in three to four hour intervals including before and after exercise when there becomes an exaggerated potential for MPS (see next section and Figure 5). Arguments often surface on the timing of protein ingestion's relationship in maximizing MPS, such as, albeit in the minority, "it doesn't matter when you eat your protein as long as you get enough daily protein."[18,189] For serious hard training athletes, the argument is unintuitive and frankly meaningless. Considering its well documented (see references above) for athletes to maximize MPS, they are recommended to eat ~1 g/lb/LBM/day of protein (many sports nutrition experts recommend protein at .73-1.0 g/lb of ***body weight/***day and higher amounts during body/weight loss), why wouldn't you spread it out throughout the day to match the protein balance cycles including before and after a workout?[190] Consuming a day's total recommendation of protein in two to three meals daily would be uncomfortable at best, and over time, based on being in a negative protein balance more hours than a positive one, logically the subject should have less exercise induced gains compared to a counterpart consuming protein when the body is ready to use it based on natural cycling – i.e. every three to four hrs.[20,21,22,24,154,191] Further, there would be no possibility of a MPS advantage, shown in many studies, in not consuming protein (fast acting, such as a shake) before and/or after training.[20,21,22,24,51,52,58,59,60,61,62,191,192]

Figure 4 - Natural Muscle Protein Balance in Non-exercising Young Adults. Source: Adapted from Phillips et al.[45]



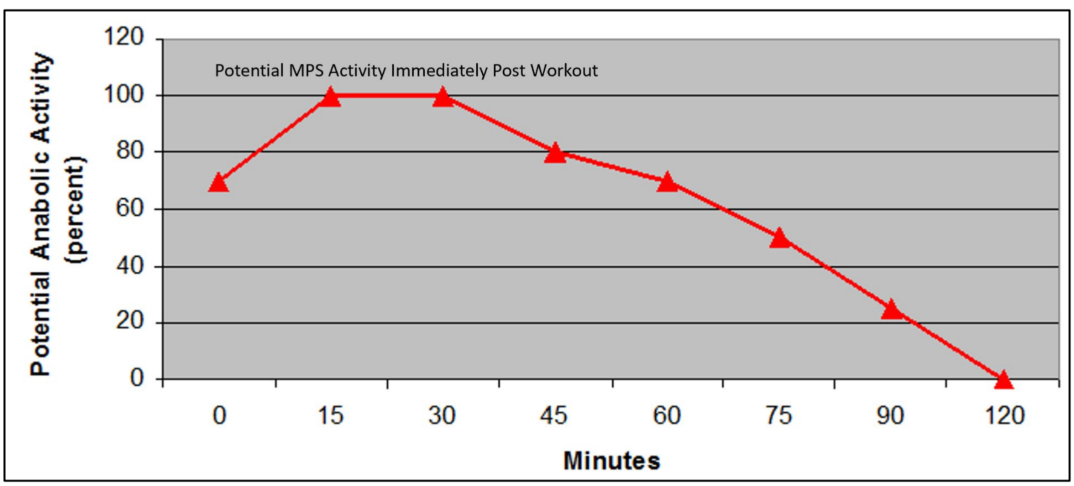
The processes of MPS and MPB in post pubertal healthy humans up to ~30 years in the normal (non-exercised) state. Protein synthesis fluctuates with protein intake and fasting across the diurnal cycle, and changes in the increases in muscle protein mass are equaled by losses.
Note: Cost of MPS & MPB (protein turnover): 1.04 Kcal/g and ~1-2% of all protein replaced daily[44,45,158,159]

Exercise Induced "Metabolic Window"

The so called "metabolic or anabolic window" is a period when there is an exaggerated anabolic potential created by exercise and realized by the simultaneous presence of exogenous amino acids within a specific timeframe. This convergence results in a period of enhanced MPS that would potentially contribute to improved daily recovery, and thus greater long-term gains, as opposed to no AA feeding during this opportune timeframe where nutrient sensitivity is heightened from exercise-induced muscular damage. As shown in Figure 5, the proposed timeframe would begin immediately following exercise at which point the anabolic potential would be at its highest and slowly wane until ending 90-120 minutes post-exercise. Thus the "window" is open widest upon exercise cessation, slowly closing to baseline during the next 90-120 minutes. Although theoretical, there is no downside to this practice. Yet as many studies have demonstrated, there may be a significant recovery/MPS incremental upside that may not be accounted for at another point in time without this regular pre/post exercise feeding (i.e., not a complete "catch up"), even when all things are equal (e.g., total daily protein intake, exercise protocol, etc.). Logically then, this practice may have slow accruing benefits to an athlete's competitive lifespan, and to exercisers over a lifetime of activity, possibly prolonging years of desired movement modalities and independence.[193]

Because exercise sensitizes muscles to hyper-aminoacidemia environment,[194] the long-held practice by strength and physique/bodybuilding athletes of ingesting a fast acting protein (with or without fast acting carbohydrates) via liquid delivery system (i.e. powder mixes) before and immediately after exercise is now mainstream and commonly recommended to serious/competitive athletes[21,22,24,191,195,196] and popularized by the everyday exerciser as a safe and effective means of maximizing and potentially prolonging exercise results.[51,52,58,59,60,61,120,128,190,192]
There is an exaggerated MPS response if and when exercise and AA converge -i.e., the so-called exercise-induced "Anabolic Window."[45,46,128,165,193,195,198,199]

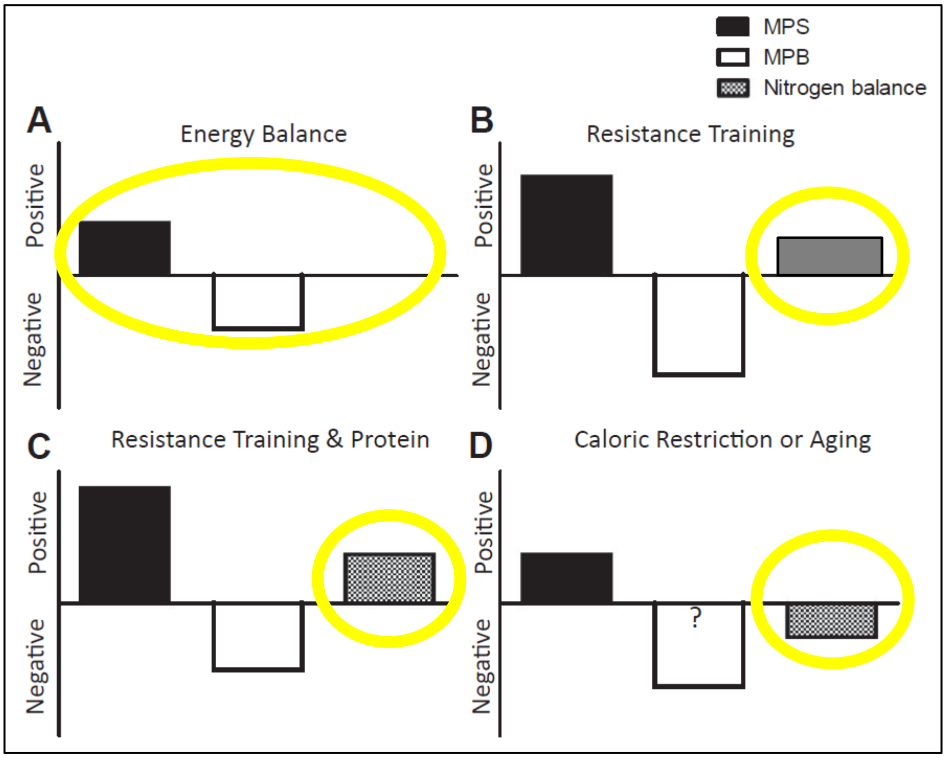
Figure 5 - Closing of the Proposed Exercise-Induced "Anabolic Window" (Source: Adapted from Ivy et al. [195])



MPS and glycogen synthesis potential (channel activation, nutrient sensitivity, etc.) reach their highest respective points almost immediately post exercise, returning to baseline within 2-3hours, leading athletes to attempt to capture the peak activity by supplying quick acting protein/EAA to improve MPS outcomes as opposed to no feeding during this timeframe of an exaggerated MPS response when exercise & AA converge -i.e., the so-called "Anabolic Window."

Earlier studies, based on AA MPS mechanisms of actions in the face of exercise described above, suggested that dosing protein pre and post exercise would help establish the EAA concentrations at necessary levels in the affected muscles, to not only deliver their molecular signals to trigger MPS at this opportune time when the body is most responsive, but also to reduce muscle protein breakdown (MPB).[45,46,197,198] Supplementation of complete fast acting protein before and after exercise has demonstrated an incremental MPS benefit when everything else (diet, exercise and total protein) was equal.[65,195,199,200,201] See Figure 6. Subsequent pre- and post-exercise protein or EAA supplementation (with or without carbohydrates) studies have duplicated these earlier results demonstrating improved MPS and recovery compared to no feeding in this "window," hence this practice has become part of the total daily protein ingestion timing protocol recommendation.[21,22,24,51,52,58,59,60,61,120,128,190,191,192,195,196]

Figure 6 - Incremental Overall Nitrogen Retention (skeletal muscle) with Pre/Post Exercise Feedings
Source: Adapted from Devries,[40] Philips,[45] and Pasiakos[46]



A: No net increase (nitrogen balance) in skeletal muscle (SM) as in young healthy (~20-30 yrs.) non-exercising adults, MPS=MPB.
B: Addition of exercise and normal diet with adequate protein but without immediate pre/post AA feeding, MPS≥MPB up to point.
C: Pre/post AA feedings (anabolic windows) in addition to normal diet and exercise may produce greater daily MPS signaling and activity (including through less MPB), which may be incremental to normal feedings
D: Caloric restriction and/or ageing lead to MPB>MPS, in which increased protein intakes and exercise can minimize or reverse up to a point based on deficit, age and/or training experience

The two major opportunities that present themselves in this exercise induced "anabolic window" are 1) reducing excessive exercise induced muscle damage, and although MPB is necessary to stimulate exercise increases in MPS, too much MPB may be counterproductive since exercise protocols that induce hypertrophy show an eventual decrease in muscle damage, compared to the initial phase of exercise, while hypertrophy becomes measurable and continues to manifest,[202] (to be sure, androgens/testosterone have anti-catabolic actions via inhibition of the actions of the catabolic hormone, cortisol, that leads to increases in MPS[203,204]). Therefore, during intense training, reducing MPB by a slight protein-induced simulation of insulin (insulin may primarily regulate muscle anabolism through its known inhibitory effects on MPB[205]) and presenting EAA to the affected tissues before and during activity,[46,128,206,207] may more quickly and continuously support enhanced remodeling.[128,187,208,209,210] Further, Gieske et al. demonstrated that protein before exercise can increase rates of energy expenditure and fat oxidation compared to placebo or fasting

before exercise, which may also contribute to the fitness end goals.[192] 2) MPS (and glycogen synthesis) potential is at its highest point (see Figure 5) immediately post exercise but this sensitivity also wanes quickly,[46,211,212,213] thus there is no harm and may be a benefit (as noted above), to an almost immediate delivery of a fast releasing protein/EAAs (carbohydrate as necessary[214]) to potentially maximize the activated MPS machinery by creating a hyper aminoacidemia environment.[128,195,199,200,201,212,215] Again, you have to consume a known amount of daily protein split throughout the day anyway, so it might as well include a pre and post exercise portion.[216]
Post exercise ingestion would take place independently of whole foods in order to minimize normal EAA clearance by the splanchnic bed and perhaps more importantly, to avoid slower gastric emptying by accompanying foods since the goal is rapid hyper-aminoacidemia during this timeframe.[51,128,217,218] Timed ingestion of whey protein both pre- and post-workout[219,220,221,222,223] facilitates a more rapid absorption of amino acids into the bloodstream and their subsequent delivery to the target tissues with less splanchnic extraction, when compared to other sources of proteins.[11,12,15,51,52,53,54,55,56,57,58,59,60,61,62,63,64,65,224,225,226]

Protein During Sleep

Ideal MPS protocol may also include a final dose before bedtime since sleep time is generally the longest lapse in which there would be a reduced extracellular EAA presence, and protein ingestion before sleep has demonstrated increases in MPS rates during overnight recovery from exercise bouts.[227] In an update on pre-sleep protein supplementation studies, Snijders et al. found protein ingestion prior to sleep can be applied in combination with resistance type exercise training to further augment the gains in muscle mass and strength when compared to no protein supplementation and that 30-40 grams may be most effective due to length of sleep time.[228]

In summary, athletes must consume 1 g/lb/LBM/day regardless of daily timing (most consume more). Further, sports nutrition experts recommend protein at .73-1.0 g/lb of ***body weight***/day (higher amounts during body/weight loss). Therefore, doing so timely as described here and by other experts, puts the subject in position to take advantage of all possible events that drive and potentially maximize MPS/recovery to maintain a positive (or even nitrogen balance) muscle protein balance as often as possible, which is the goal of all athletes and should be the goal for all humans to help stave off the inevitable in support of lifelong independence. As Arent et al. in their thorough review of nutrient timing titled, *"Nutrient Timing: A Garage Door of Opportunity?"* "Current evidence shows feeding consistently throughout the day, particularly in the peri-exercise period is the most optimal strategy for maximizing performance. On the question of an 'anabolic window,' based on our (the authors) current understanding of protein metabolism and resistance training, if anything, it would simply appear that this window is much longer than originally proposed and may in fact be more like a garage door. Unfortunately, this has been used to argue that post-exercise refeeding is not essential. However, it may be optimal and represents an opportunity to improve adaptation and recovery and especially if continued over time."[193]

Whey Protein in Muscle Protein Synthesis/Hypertrophy

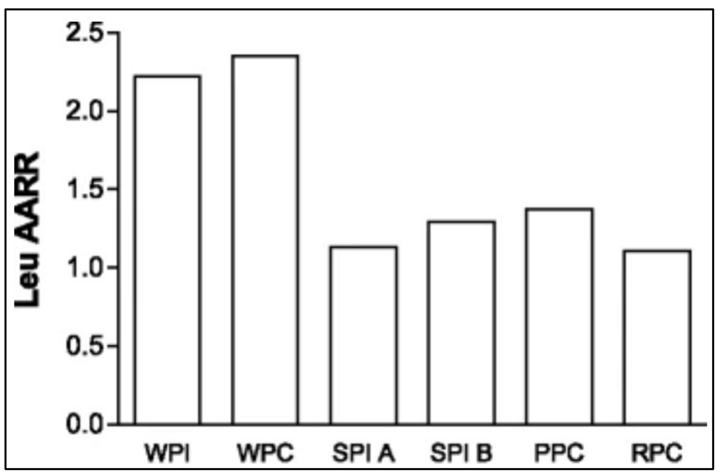
Depending on total diet, the quality of the ingested protein determines the degree of the MPS response. The quality of individual proteins is established by their amino acid content, bioavailability and digestibility (see opening section).[9,10,11,12,13,14,15,229] As during weight loss, whey protein appears to be superior to other complete protein sources, including soy and casein, in stimulating MPS and muscle hypertrophy.[11,12,15,51,52,53,54,55,56,57,58,59,60,61,62,63,64,65,230,231] As presented in Table 4, per gram of protein, whey protein contains more EAAs, including leucine and the other branched chain amino acids (BCAAs),[63,64,65] which are the primary AAs necessary to trigger MPS, and therefore this factor alone establishes whey protein's higher potential for muscle hypertrophy.[46,51,52,53,54,55,56,57,58,59,60,61,62,63,64,65,232,233] Figure 7 from Phillips et al. shows the leucine reference ratio for different protein concentrates and protein isolates showing WPC having the superior ratio.

Table 4 - Comparison of Protein Sources

	Whey	Casein	Soy
Complete protein?	Yes	Yes	Yes
Digestibility	Fast	Slow	Fast
Amino acid content (g/25 g protein)			
Leucine	3.0	2.3	1.5
Σ EAA	12.4	11.0	9.0
Σ BCAA	5.6	4.9	3.4
Splanchnic AA extraction	Low	Low	High
PDCAAS	1.0	1.0	1.0

Source: Devries and Phillips.[52] Whey and soy are labeled "fast proteins." Tang et al. demonstrated the rapid rise in plasma AA concentrations with soy and whey compared with a slower rise with casein consumption.[65] Additionally, the amount of protein subject to splanchnic extraction is based on its amino acid content. The BCAAs undergo less splanchnic catabolic activity and therefore proteins with higher BCAA content have more AA available to support muscle protein synthesis (MPS) [234,235]

Figure 7 – The Leucine Amino Acid Reference Ratio (AARR)



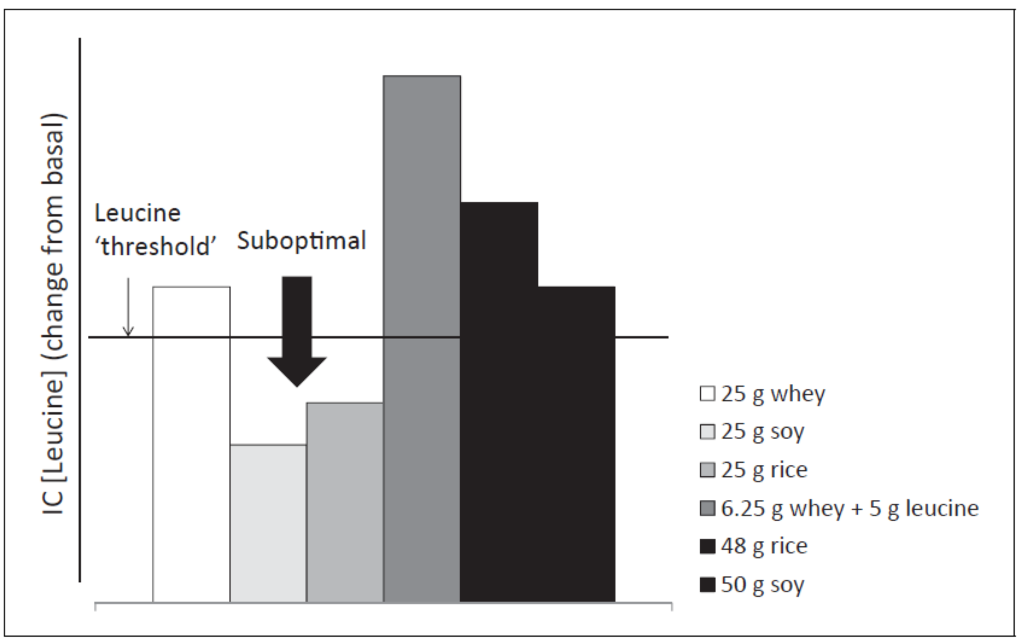
Protein Source

Source: Phillips et.al. [236]The leucine amino acid reference ratio (AARR) is the content of leucine in the protein measured compared to a hypothetical best protein to provide the EAA needed and shown here for several popular protein concentrates and isolates. Values are from reference.[237] WPI (Whey Protein Isolate); WPC (Whey Protein Concentrate) from the Fonterra Co-operative Group; soy PI A (Supro 670) and soy PI B (Supro XF) were from Solae; pea PC (Nutralys S85) from Roquette; and rice PC (Oryzatein 90) was from Axiom Foods.

As previously discussed, the importance of leucine in stimulating MPS is well established,[238,239,240,241,242] and therefore researchers conceptualize a leucine threshold for maximizing MPS as shown in Figure 8. The leucine threshold ("trigger") proposes that for maximum MPS to take place following protein ingestion, the muscular intracellular leucine concentration needs to reach a given level – i.e. "the leucine threshold."[45,75,121,122,174] In order to maximize protein synthesis, this leucine threshold, depending on age and activity, may be in amounts greater than 2.5 g per

protein dose.[45,236,65,121,122,124] (Readers should be reminded regardless of the excess in leucine, the remaining EAA with the lowest concentration relative to demand, will be the limiting factor in the anabolic response – i.e. all EAAs required for MPS need to be supplied proportionately to the demand, with leucine as the primer leading the way[122,127,128]). There has also been some data that shows whey protein may uniquely trigger and prolong MPS in part by enhancing the phosphorylation of select proteins within the mammalian target of rapamycin (p70S6K, eEF2) and by activating proteins within the mitogen-activated protein kinase (ERK1/2, p90RSK) signaling.[223, 243, 244, 245]
Additionally, whey protein's more rapid digestion and absorption properties, including being acid soluble,[51,60,61,62] along with less EAA splanchnic extraction compared to other proteins allows the intramuscular amino acid levels to rise quickly to create the hyper-aminoacidemia environment associated with enhanced exercise-induced MPS.[52,65,212,224,225,226, 246] To be sure, whey and soy proteins are commonly referred to as "fast" digesting proteins while casein is considered "slow" because it tends to clot due to the acid pH of the stomach thereby entering slowly into the small intestine.[224,225,247] Although soy protein leaves the stomach quickly, besides having a lower leucine content and AARR (as shown in Tables 5 and Figure 7 above), the bioavailability of the AA from soy protein to support MPS is also inferior to that of whey and casein gram per gram.[51,234] Because of soy's AA profile/structure, greater amounts are directed toward splanchnic catabolic activity, urea synthesis[234,235] and oxidation.[248] The higher BCAAs, particularly leucine, found in milk proteins as compared with soy, offers greater EAA availability to peripheral tissues in support of MPS.[51,249] All considered, because whey protein's characteristics are superior in digestibility, AA content, and AA bioavailability, whey protein gram for gram affords a greater, faster rise in blood leucine and other EAA concentrations (desired intramuscular hyper-aminoacidemia) following consumption compared with other commonly ingested protein supplements, which may be beneficial overtime as it relates to MPS and MPB daily cycles including timing around exercise and calorie allotments for desired body composition.[11,12,15,51,52,53,54,55,56,57,58,59,60,61,62,63,64,65,188,208,224,225,226,243,250,251,252]

Figure 8 - Leucine Intracellular Concentration from Various Protein Sources



Source: Devries and Phillips.[52] Intracellular (IC) leucine concentration following the consumption of varied doses of protein in relation to the proposed "leucine threshold." This data is gathered from young, resistance-trained subjects therefore this "leucine threshold" would increase with age and physical inactivity.[45] The leucine threshold proposes that for maximum MPS to take place following protein ingestion, the muscle intracellular leucine concentration needs to reach a given level and the amount of leucine should be >2.5 g – i.e. "the leucine threshold.[65,121,122,124]

Whey Protein Supplementation (WPS) is Effective and Superior in MPS (studies and comparisons)

In general, based on the above-described characteristics of whey protein, WPS is found superior to other complete protein (or carbohydrate) supplementation protocols when the measured endpoints are related to MPS.

- Farup et al. investigated WPS in 24 young healthy subjects versus isocaloric carbohydrate (CHO) following eccentric exercise on fiber type-specific skeletal muscle satellite cells (SCs), which are essential for muscle remodeling/growth processes following muscle breakdown. They found in type II fiber-associated SCs, the whey group increased SCs/fiber and mononuclei significantly more than the placebo (CHO only) and concluded: "whey protein supplementation may accelerate satellite cell proliferation as part of the regeneration or remodeling process after high-intensity eccentric exercise."[253] The same group found 19 g of whey combined with concentric exercise accentuated type II fiber hypertrophy compared to CHO only. [254] Farup et al. also demonstrated that whey protein (with high leucine) increased muscle and tendon hypertrophy compared to placebo.[255]
- Smiles WJ et al. found that 30 g of whey protein, compared to CHO placebo consumed immediately post-exercise favorably altered autophagic responses (normal physiological process in the body that deals with destruction of cells to help maintain homeostasis) during energy deficit and resistance training.[256]
- Snijders et al. demonstrated that exercising subjects using 27.5 g of whey protein (PRO) before sleep, increased muscle strength to a significantly greater degree than in the placebo (PLA) supplemented group. Additionally, quadriceps muscle cross-sectional area had a greater increase in the whey group than in the PLA group (+8.4 cm vs. +4.8 cm, respectively). Both type I and II muscle fiber size increased with a greater increase in type II muscle fiber size in the PRO group (+2319 μm) than in the PLA group (+1017 μm).[227]
- In a 12-week study, Tahavorgar et al. found that WPC preloads 30 minutes prior to the ad libitum main meal exerted stronger beneficial effects than did soy protein isolate preloads on appetite, caloric intake, anthropometry, and body composition of free-living overweight and obese men.[56]
- Probably the most telling study was done by Tang et al.[65] which has since been duplicated by others.[52,63,91,121,122,221,222,224,225,226,248,249,250,251] They found that MPS after consumption of whey was approximately 93% and 18% greater than casein and soy, respectively. A similar finding was shown after exercise. MPS following whey consumption was ~122% greater than casein and 31% greater than soy. They concluded: "Feeding-induced simulation of MPS in young men is greater after whey or soy protein consumption than casein, both at rest and after resistance exercise." Also, despite both being fast proteins, whey stimulated MPS to a greater degree than soy following exercise. The differences may be related to how quickly the proteins are digested and EAA splanchnic extractions and probably differences in leucine content of each protein as described throughout this section.[52]
- Noteworthy, Reidy et al. found a greater and prolonged increase in MPS post-exercise with ingestion of a protein blend of whey, casein and soy as opposed to whey protein alone, thus the blend promoted greater total muscle protein synthesis measured by the protein fractional synthetic rate (FSR).[257] They also found that ingesting the protein blend, or whey protein alone, enhanced the rate of amino acid transport into muscle, increased select amino acid transporter mRNA expression, and increased post-exercise myofibrillar protein synthesis. The results provide support for consuming a protein blend to increase and prolong post-exercise muscle protein anabolism. Presumably, the greater results from the blend may be based on the varied digestibility of each source[51,234] and because soy protein may help spare whey from splanchnic catabolic activity, urea synthesis and oxidation.[52,65,234,235,245,245,247,248,249]
- In the same vein as the Reidy et al. study, Mitchell et al. found that 30 g of whey or soy protein resulted in similar p70S6 kinase phosphorylation (important step in the initiation of protein translation factors) at two hours post-exercise but soy failed to promote prolonged phosphorylation of p70S6K up to four hours as whey protein did.[258]
- Oikawa et al. found supplementation (30 g twice daily) with whey protein (WP) produced greater increases in both acute and longer-term MPS than collagen peptide protein supplementation in older women, suggesting better aging muscle retention with WP.[54] An earlier study by the same group and same supplemental protein dose, found

WP supplementation but not collagen, augmented lean leg mass and muscle protein synthesis during recovery from inactivity and a hypo-energetic state.[259] Other similar comparisons generally find the same results whey/collagen protein comparison result.[57]

Whey Protein in Health and Aging

The loss of skeletal muscle and performance with aging at some point is inevitable but can be slowed with proper nutrition and activity. In the context of this paper, specifically resistance exercise and proper protein/amino acid intake throughout life are two well-known treatments for maintaining muscle performance, thus long-term health and functional independence. Although it is now clear that older humans, primarily based on the natural decline in hormone levels, require more protein per pound of body weight than most (non-athletic) younger counter parts to support exercise recovery and prevent or slow net muscle protein losses, the recommendation given throughout this document neatly fits this group as well as all others: 1 g/lb/LBM/day split throughout 3-6 meals including a dose before and after exercise. And for all the same reasons listed above, especially calorie per calorie and EAA content per gram of protein, supplementation as needed would make whey protein a top alternative.

Background

From a whole-body perspective, the efficiency of an organism's proteostasis control systems, which work to maintain and recycle the proteome (entire set of proteins that is, or can be, expressed by a genome, cell, tissue, or organism at a certain time), diminishes in aging eventually leading to age related dysfunction. To be sure, all proteomes deteriorate at some point with age partially caused by deregulation of nutrient signaling and accumulation of resulting damage leading to a decrease in protein synthesis. It has been suggested that this protein synthesis decrease may serve as an adaptation of the organism to age-related changes and may be advantageous to longevity, because a downregulation of protein synthesis and an increase in proteome stability have been associated with increased lifespan.[260]

Notwithstanding the above, muscular performance is one of the most important determining factors of long-term health and functional independence whether you are a young competitive athlete or a lifelong sedentary aging human or everyone in between. Therefore, preserving skeletal muscle, which is central to structure and functional mobility thus health, as long as possible, would be everyone's goal.

Athletic Analogy – Sports Nutrition Supports Healthy Aging

Competitive athletes and exercisers constantly seek physical improvement to remain competitive in their respective sport by attempting to make continuous progression in strength and performance gains, or as with avid exercisers, simply enhance exercise sessions over time. It should be no exception that no matter who you are or what you do, daily recovery to maintain a desired muscle protein balance and related functions would be the goal of every human seeking to maintain preferred activities and physical independence throughout a lifespan.

As detailed above, in the presence of amino acids (protein), exercise stimulates natural human skeletal muscle synthesis and muscle performance throughout life when compared to a non-exercise state.[24,45,155,197] Various forms of mechanical loading (exercise design) initiate muscle protein's related anabolic signaling and the mode, intensity and volume of exercise differentially affect signaling, thus acute and long-term adaptations/outcomes.[176,261,262,263,264,265]

The functional demands of specific contractile activities lead to adaptations in muscle fiber type distribution, size, endurance capacity, contractile velocity, etc., demonstrating the plasticity of skeletal muscle (SM) and leads to changes in protein activity and abundance,[264,265,266] making protein intake and exercise key components in support of aging muscle regardless of a person's vocation, thus "sports nutrition" can indeed be considered a part of healthy aging.

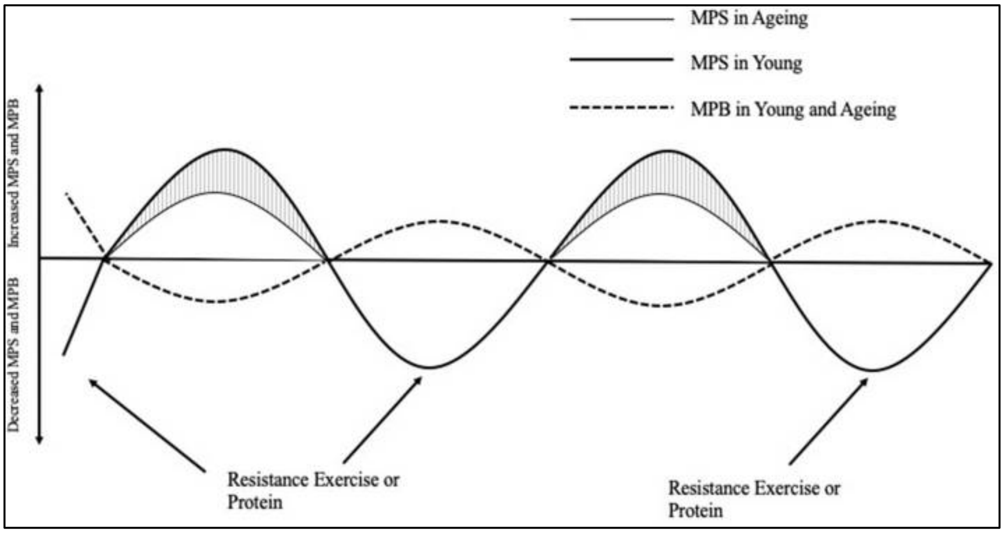
The general goal of most athletes is to maximize the body's natural muscle protein synthesis (MPS) processes, which include applying peak strength during exercise and recovering adequately from each training bout to constantly increase performance and if desired, thru proper/specific training protocol, increase SM size. Thus, athletes/exercisers attempt to progressively improve physically by making each training session build on the previous, leading to continuous athletic and physical progress since unaccustomed exercise regularly sets the stage for the desired muscle remodeling (anabolism) that would potentially improve performance or size.[45,46,267,268,269] However, despite exercise's constant MPS initiation or stimulus, positive training progress slows significantly with age and experience (the younger and/or less experienced the more gains[270,271,272]),[273] and training plateaus become common occurrences,[45,159,274,275] leading researchers and athletes to believe that something may be missing (nutritionally) in the pre or post exercise period that would otherwise continue progression from proper unaccustomed training.[20,276,277,278] In other words, although at some point aging clearly blunts the human response to exercise and nutrition, and eventually there will always be an inevitable decline in performance,[279,280,281] unaccustomed exercise is a successful continual trigger event for the desired result, leaving nutritional/bio-ingredient modulations to deliver the progressive or responsive outcome including maintaining muscular health.[45,158,159,267,282] These conditions set the stage for dietary supplementation (e. g. intact protein, amino acids, etc.) when all else is equal and training and diet protocols are optimized for the desired progression, including in attempts to stave off the eventual age-related final size and/or performance plateau and decline.

The same rationale applies to staving off age-related losses of muscle size/performance for everyone, meaning performing nutritionally what can be done to slow age-related declining net muscle balance (predominately average negative balance), which is why 1 g/lb/LBM/day works for all age groups. Aging, thus long term health, may increase protein needs because aging naturally increases the body's resistance to the anabolic effects of exercise, amino acids, insulin/hormones and other related protein synthesis mechanisms.[12,29,31,32,33,34,35,36,124,125,203,275,283,284,285] In fact Yang et al. found that in contrast to younger adults in whom it's been proposed that post-exercise rates of MPS are saturated with 25-30 g of protein per meal,[143,146,286] exercised muscles of older adults respond to higher protein doses of up to 40 g.[29] This data has led modern researchers to discover that the RDA for protein is insufficient in slowing the natural age-related anabolic resistance. Therefore, newer expert recommendations ranging from 0.5 to 0.8 g/lb of ***body weight***, for older adults (exercisers or not), demonstrate greater maintenance of net protein balance and are sufficiently covered with the same amount (1gm/LB/LBM/D) that works for younger active humans.[287,288,289,290,291,292,293,294,295]

Aging and Anabolic Resistance - Basis for Protein Recommendation

The 1 g/lb/LBM/day recommendation supports the growth (including natural and exercise-induced net protein gains), activity and performance increases in the younger population with naturally higher anabolic/androgenic hormone (testosterone, growth hormone [GH], insulin-like-growth factor-1 [IGF-1], etc.) activity,[203,296] and helps overcome the older adult's natural declining anabolic/androgenic hormone activity when net protein balance becomes predominantly negative.[12,297,298] In other words, evolution has designed younger humans to naturally utilize available nutrition more efficiently than older counterparts, because GH, IGF-1 and the hypothalamic GH-releasing hormone (GHRH) make up the somatotropic axis (growth axis), which influences the regulation of puberty, gonadal function, and resulting structural and functional maintenance of tissues and growth.[297,299,300] Aspects of this youth efficiency (nutrients and the GH/IGF-1 axis[301]) begins to wane following puberty with a significant decline by the late third or early fourth decade of life (14% per decade).[300,302] Further, in line with the declining MPS in response to exercise as depicted in Figure 7, loss of muscle mass with aging is largely due to the progressive loss of motoneurons and subsequent reduction in muscle fiber number and size causing muscle function to decline, and this evolutionary-programmed loss may be slowed but cannot be stopped with the remaining motoneurons/muscle fibers.[303] Therefore, humans use exercise/activity and adequate nutrition (e.g. vitamins, minerals, essential fatty acids, protein, etc.) that may be altered by aging, to help stave off the inevitable including countering sarcopenia.[12,304,305,306,307,308,309,310,311] Additionally, adequate protein intake higher than the RDA has turned out to be an important factor in fighting off the physical detriments of skeletal muscle loss.[312,313,314,315,316,317] Further, studies including the Framingham Heart Study Offspring cohort study, protein intake has been inversely associated with changes in the inflammation and oxidative stress score suggesting that overall inflammation and oxidative stress increased less in those with the highest intake compared to lowest.[318,319,320]

Figure 9 - Exercise*or Feeding-induced Age-related Decline in MPS



Source: Adapted from Breen and Phillips[321] The ***response*** of MPS and MPB on net protein balance after acute resistance exercise or protein ingestion in young and aging populations*.[321] Between meals or overnight fast, MPB exceeds MPS leaving net protein balance negative. Following resistance exercise or the ingestion of protein, younger humans have a greater myofibrillar protein synthesis response compared to older people, thus exercise and protein adjusted protein intake play a major factor in attenuating age-related decreasing net protein balance leading to skeletal muscle protein loss over time.[322]
****Exercise alone stimulates the MPS response, but the body cannot increase net protein balance without exogenous intake of protein (EAA)***

Whey Supplementation in Health and Aging

Given the role of EAAs, especially leucine and the other BCAAs in MPS,[44,65,179,180,181,182,238,239,240,241,242,243] and whey having the highest content of these MPS activators per gram of protein (EAA density), whey protein is a top choice when supplementation is necessary.[9,10,11,12,13,14,15,52,55,56,63,64,65,314,320,323,324]

To be sure, there are protein, including WPS, interventions in aging populations that have delivered null results but upon thorough review, none to our knowledge produced negative results.[325,326,327,328,329,330] Like most nutrition interventions related to treatment versus prevention (mindful prevention of skeletal muscle decline including maximizing MPS is the basis for the lifelong human dietary protein recommendation described in this paper) that yield null results, generally suggests: wrong population (e.g. exercise history [trained/untrained], physiological state, lifestyle, etc.), total baseline nutrient levels/intake (including protein supporting nutrition) or history of dietary intake not accounted for, improper/low dosages, intervention was too late (condition taken irreversible hold) or too short to make up for lifelong nutrition shortages including low daily protein intake (≤RDAs). The preponderance of evidence supports WPS for aging humans with or without exercise to support healthier musculoskeletal aging and overall health.

Readers interested in whey protein supplementation (WPS) studies in support of aging/sarcopenia, are referred to the Gilmartin et al. titled "Whey for Sarcopenia; Can Whey Peptides, Hydrolysates or Proteins Play a Beneficial Role?"[324] This review examined the evidence that whey peptides, hydrolysates, proteins (concentrates) or products can delay or reduce symptoms of sarcopenia or alter biomarkers of sarcopenia in the older human and animal adults and muscle cells lines. Clinical trials using WPS under many conditions or settings, including with or without exercise in older humans and animals, muscle cells in vitro, and older individuals with sarcopenia, are highlighted in Tables 1-6. The conclusion of the review was that daily WPS containing "35 g of whey is likely to improve sarcopenic biomarkers in frail or sarcopenia individuals. Whey supplementation, consumed by older, healthy adults certainly improves muscle mTOR signaling, but exercise appears to have the greatest benefit to older muscle".[324]

The EAA density of whey protein may be especially important when appetite is compromised as in aging and frailty,[331,332] or if calories or nitrogen content were to be limited based on the overall health and body composition goal.[12,30,323,324,333,334,335,336,337] To that point, Bauer et al. showed that 13 weeks of a Vitamin D and leucine enriched whey protein supplement resulted in improvements in muscle mass and lower extremity function among sarcopenic adults compared to the control group.[338] Similar formula and results were found by Lin et al.[337] Liberman et al. using the same nutritional intervention, found 13 weeks of nutritional supplementation with Vit-D and leucine-enriched whey protein to attenuate the progression of chronic low-grade inflammatory profile (CLIP) in older sarcopenic persons with mobility limitations.[123] Hashemilar et al. found similar reductions in markers of inflammation with 20 g/day of whey supplementation in subjects recovering from compromised heart conditions.[339] Another example of the potential advantage of EAA density was Niitsu et al. using 32.2 g of whey protein supplementation pre and post rehabilitation during a two week postoperative period. They found that the combination of whey protein intake and rehabilitation for two weeks in the early postoperative period has a beneficial effect on knee extension strength in *both* lower limbs and Barthel Index (transfer, walking and toilet use) scores in patients with hip fracture.[340] See Gilmartin et al, for more data on whey supplementation in specific elderly health outcomes.[324] Important safety concerns related to bone, renal function, etc. of higher protein intakes (above the RDA) have all but left the radar.[28,30,34,38,107,341] Kerstetter et al. used 45 g/day of whey protein supplementation so that total daily protein was well above the recommended dietary allowance (0.8 g/kg of body weight). Compared to placebo, they found the whey supplemented group preserved fat-free mass without adversely affecting skeletal health or renal function in healthy older adults.[342] Recent studies including meta-analysis have concurred with these previous safe and effective reports of higher protein intakes including use of supplements to support bone and overall health without negatively effecting other health parameters.[343,344]

Whey Protein in Cardiovascular Health and Blood Sugar

The use of whey protein as a dietary strategy is widespread in medical fields to assist in meeting nutrient requirements and offer potential unique bio-active components (as found in whey concentrates named above) that may contribute to healing or supporting pharmaceutical therapies.[339,345,346,347,348,349,350] The use of whey protein in clinical settings or as a specific treatment outside of skeletal muscle structure and function preservation, is not considered or supported by this document. And although studies clearly have demonstrated the efficacy of WPS in support of specific treatment outcomes, related studies cited here are only to validate safety, which may include adjunct health contributions during regular use. This paper is solely related to the use of WPS in support of meeting updated protein recommendations to help users remain active, maximize MPS sports and/or exercise goals as desired, and stave of the inevitable age-related loss of muscle to help persons remain active and independent throughout a lifespan. In other words, the use WPS to support growth, development, body composition and athletic goals, and healthy aging, safely and effectively.

- Bolh et al. found that a 60 g/day of a whey protein supplement decreased the postprandial chylomicron response compared with casein in persons with abdominal obesity, indicating a beneficial impact on CVD risk.[351]
- Jakubowicz et al. showed that over an entire 180-minute post-meal period, glucose levels were reduced by 28% after a 50 g whey pre-load with a uniform reduction during both early and late phases. Insulin and C-peptide responses were both significantly higher (by 105% and 43%, respectively) with the whey pre-load. The early insulin response was 96% higher after whey.[352]
- Winder et al. found that compared with control, whey and soy protein drinks reduced postprandial area under the curve (iAUC) by 56.5% and 44.4%, respectively. Whey protein was the only protein capable of avoiding large fluctuations and a peak in postprandial glycemia.[50]
- Arciero et al. found that exercise and timed ingestion of whey protein added to the diets of free-living overweight/obese adults, reduced visceral adiposity, improved body composition, total and regional body fat distribution, insulin resistance, and adipokines, independent of caloric restriction.[353]

- Ling-Mei Zhou et al. did a meta-analysis on whey protein's effect on circulating C-reactive protein (CRP), a marker of inflammation, and found that in people with high CRP (baseline ≥3 mg/liter) levels, using a daily dose of whey protein greater than 20 g significantly lowered CRP by 0.72 mg/liter.[354]
- Note: a meta-analysis revealed that a 20-gram increment of protein per day was associated with a 26% decrease in negative cardiovascular system health outcomes.[355]
- Fekete et al. found whey (and casein) protein supplementation of 56 g/d for 8 weeks to improve vascular reactivity, biomarkers of endothelial function, lipid risk factors, and lowered blood pressure.[356]
- The Badely et al. systematic review and meta-analysis of randomized controlled trials was conducted to systematically evaluate the effect of whey protein on the components of metabolic syndrome in overweight and obesity patients (2,344 individuals reviewed in this systematic review of 37 published articles).[357] They concluded that WPS significantly reduced the systolic and diastolic blood pressure (SBP and DBP), high density lipoprotein (HDL), waist circumference, triglycerides (TG) and fasting blood sugar (FBS) in intervention groups in comparing to placebo or control subjects.[357]

Whey Protein and the Immune System

Like most body systems, adequate protein is necessary to properly support the human immune system throughout life but, especially important in aging as the immune response weakens. Peripheral blood mononuclear cells (PBMCs) include monocytes, lymphocytes and natural killer (NK) cells all play important roles in the immune system and protein intake affects their expression.[358,359] Once again, whey protein supplementation as needed including whey protein concentrate (WPC), in support of immune function, has demonstrated success and comparatively superior actions.[350,360,361,362]

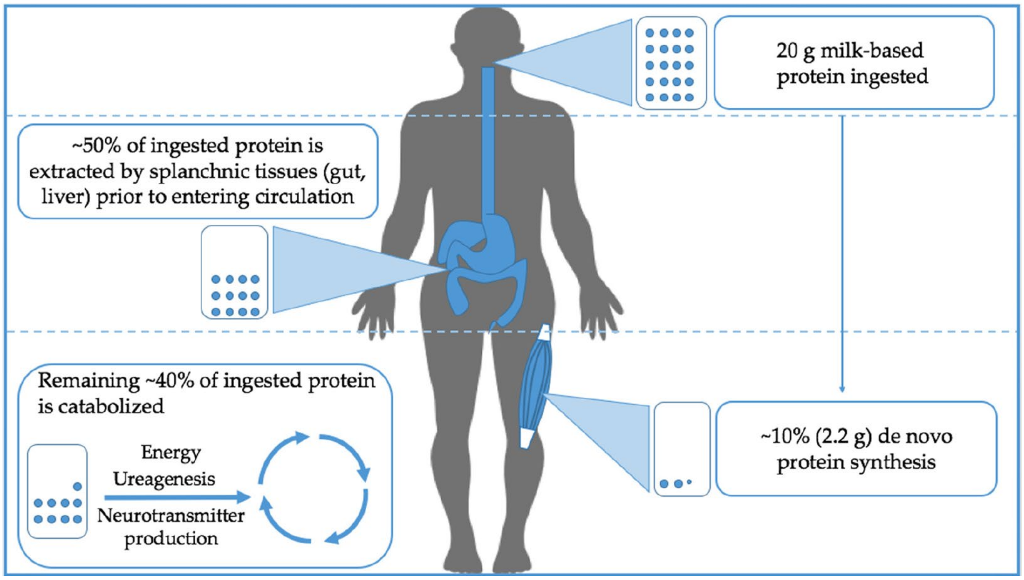
The unique components of WPC as described above (e.g. beta-lactoglobulin, alpha-lactalbumin, bovine serum albumin, lactoferrin, immunoglobulins, vitamin D, and minerals, etc.)[71] have been shown to positively affect the expression of PBMCs.[363] Additionally, whey's functional properties show anti-microbial activity and protection against viral and bacterial organisms.[364] Josse et al. also demonstrated anti-inflammatory effects,[365] which may be due to its amino acid content and profile (26% BCAA, plus L-arginine, L-lysine, L-glutamine and sulfur containing AA such as cysteine and taurine).[364] Whey protein supplementation might also function as an immune modulator through other mechanisms, such as L-glutamine, which is critical for the L-glutamine-GSH axis.[63] Therefore, high protein diets including whey protein, through its unique component profile, may impart its impact on immune function through redox regulations pathways,[71,366] which may be important to intense and endurance training athletes.[24,39,63,367,368]

Upper Limit of Protein's Anabolic Efficacy

The question about how much protein the body can use from one meal to build muscle or recover properly, is another one of those, "who cares" answers since: 1) as referenced throughout, the experts' established daily total protein to maximize MPS is ~1 g/lb of LBM (or 1.6-2.2 g/kg of body weight) and more during an energy deficit (1.36 g/lb); 2) as Figure 10 from Stokes et al. depicts, consumed protein/AA utilization is basically unlimited, meaning almost all ingested protein/AA will be used for something including local metabolism needs of the gut/splanchnic tissues, energy, suppressing muscle breakdown and other body protein/AA needs (whole proteome);[369,370] 3) regardless of the constituents of metabolized protein's final destination, it contributes to satiety helping control calorie intake;[56,91,92,93,95,110,111,112,113,115] and 4), it would be uncomfortable to consume the required daily amount in one or two meals so you might as well space it according to protein synthesis and breakdown daily rhythms as defined above including before and after exercise.[45,51,52,53,54,55,56,57,58,59,60,61,62,63,65,146,186,187,188,190,208,224,225,226,243,250,251,252]

As defined above, although size, age, health, genetics, energy balance and activity will determine a person's protein requirement for maximizing MPS throughout all stages of life, the exact amount per individual is academic, not necessarily practical. The simple formula as detailed above: consuming protein at ~1 g/lb of LBM daily divided between meals every 3-5 hours,[7,11,12,17,18,20,21,22,23,24,25,26,27,28,29,30,31,32,33,34,46,143,144,154,155,191] including before and after exercise, [21,22,24,51,52,58,59,60,61,65,120,128, ,190,191,192,193,195,196,199,200,201,208,209,210,211,212,216,217,218,219,220,221,222,223] would cover most anyone, exercisers or not, of all ages and goals, anabolic requirement in maximizing MPS. To support LBM maintenance and satiety, during prolonged calorie restriction as required by physique competitors, wrestlers or other athletes who must reach a specific weight or body fat level, additional protein may be appropriate.[20,21,22,23,24,25,26,27,145,146,371]

Figure 10 - Protein Utilization



Source: Stokes et al.[376] Overview of whole- body ingested protein utilization at rest. ~50% is extracted by splanchnic tissues before entering peripheral circulation. Only ~10% of the ingested protein is utilized for skeletal muscle protein synthesis while the rest is catabolized for other functions, thus basically unlimited utilization.

Is There an Anabolic Upper Limit?

Notwithstanding the aforementioned, the amount of protein per meal in non-calorically restricted athletes, regardless of the ambiguous "muscle full effect," to maximize MPS (positive net *muscle protein* balance) should be primarily determined by total muscles worked,[372, 373,374] body weight or LBM and therefore, suggested to be 0.18-.25 g/lb of LBM (0.4-.55/g/kg body weight).[375,376]

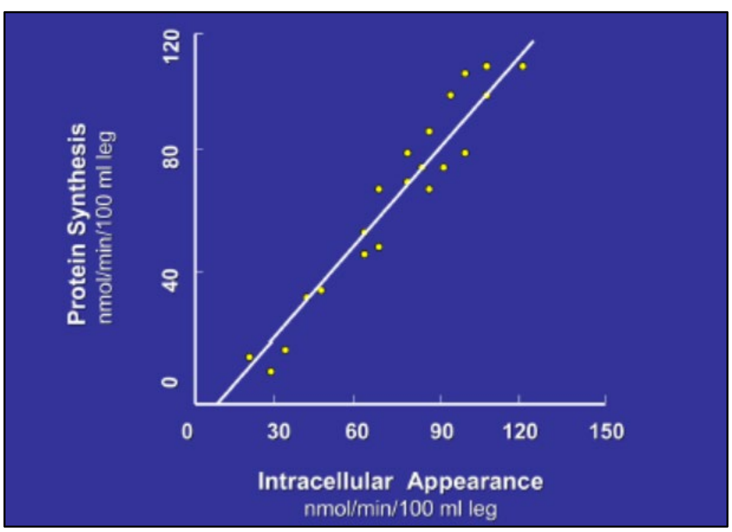
As mentioned, a protein anabolic response upper limit for acute skeletal muscle synthesis or "muscle full effect," is academic and an interesting topic but hardly applicable in the real world since athletes consuming their daily protein requirements generally consume more than the proposed ~0.2 g/lb of body weight at each meal (including a before and after exercise supplement) in order to meet total daily needs.[377,378,379,380,381,382] Based on recommendations, a non-dieting 200 lb. athlete at 10% body fat would require in the range of 180-200 g of daily protein. Dividing this amount into five meals including a before and after exercise supplement would be ~30-35 g per meal, which would only be about 6 ounces of a lean complete protein or a 35 g scoop of a common whey supplement, mindful, that there are protein/AA contributions from the remaining meal foods consumed. In meeting the total daily protein recommendation, if the athlete consumed four meals to reach the proper daily protein, each meal would need to contain ~50 g. In both cases in meeting the total protein daily recommendation, the athlete has exceeded the individual protein feeding requirements related to optimal skeletal muscle protein synthesis determined by fractional synthesis rates (FSR indicates the fraction of the protein pool that is synthesized over a given time – i.e. rate of AA incorporation into muscle[383]). Therefore, dividing up their recommended daily protein into recommended eating patterns around MPS/MPB cycles and exercise as described throughout this paper, automatically meets, or surpasses the proposed protein amounts/meal exercise induced skeletal muscle potential for maximizing a net muscle gain.

Whole Body Rates of Protein Synthesis, Breakdown and Net Balance (Anabolic Response)

Keeping muscles saturated with exogenous AAs has a limit in stimulating MPS or there would be no limit to skeletal muscle growth (or possibly other unnecessary/unwanted protein tissue growth), exercise or not.[211,213,384] These facts give rise to the need (or rationale) for muscle protein breakdown (MPB) and MPS cycles, and created the "muscle full" concept, or protein's SM anabolic threshold, but also spawned research on protein's overall anabolic (whole body) contribution that might push its proposed SM anabolic boundary for all age groups (see protein in aging section[29,385]), even if it is an almost unmeasurable amount in the immediate and short exercise term, but become relevant in the long-term.

The original commonly held belief that the maximum anabolic response to protein intake is reached at ~20-30 g in a meal (or high-quality protein scaled to 0.14 g/lb of body mass) – i.e. amino acid "muscle full effect"[373,386] was challenged by Nicolaas Deutz and Robert Wolfe.[387] In their publication, they argue that there is no practical upper limit to the anabolic response to protein or amino acid intake in the context of a meal because the anabolic response to protein intake can only be determined when rates of whole-body synthesis and breakdown are measured simultaneously, rather than simply measuring the muscle fractional synthesis rate (FSR). The total anabolic response from a nutritional standpoint is determined by the combined net gain of whole-body protein or net protein balance (NPB). Muscle FSR measured in skeletal muscle only identifies the synthesis rate of SM protein and generally studied in a non-realistic setting – i.e., a single muscle group. All proteins in the body are in a continual state of turnover and therefore there is a constant process of both synthesis and breakdown. The net gain in muscle protein over time, or the anabolic response, would have to be calculated as the difference between the rate of synthesis and rate of protein breakdown, the later also being affected by meal insulin initiation.[205,387,388] The authors found a direct linear relationship between the total rate of appearance of EAA into the intracellular pool and the rate of muscle protein synthesis (Figure 11).[387,389] From this data, they conclude that the measurement of protein synthesis can only be decided, in the context of the anabolic response, if matched against simultaneous changes in the rate of breakdown, and the intra-cellular total rate of appearance of amino acids.[390,391,392] In other words, just because the maximum rate of protein incorporated into tissues has been reached, it does not mean the maximal AA anabolic response has halted. The response may continue as protein intake increases, thus increasing intracellular AA concentrations, providing a signal to limit the rate of protein breakdown.[128,198,387]

Figure 11 - Relationship Between Protein Synthesis and the Intracellular Appearance of Amino Acids

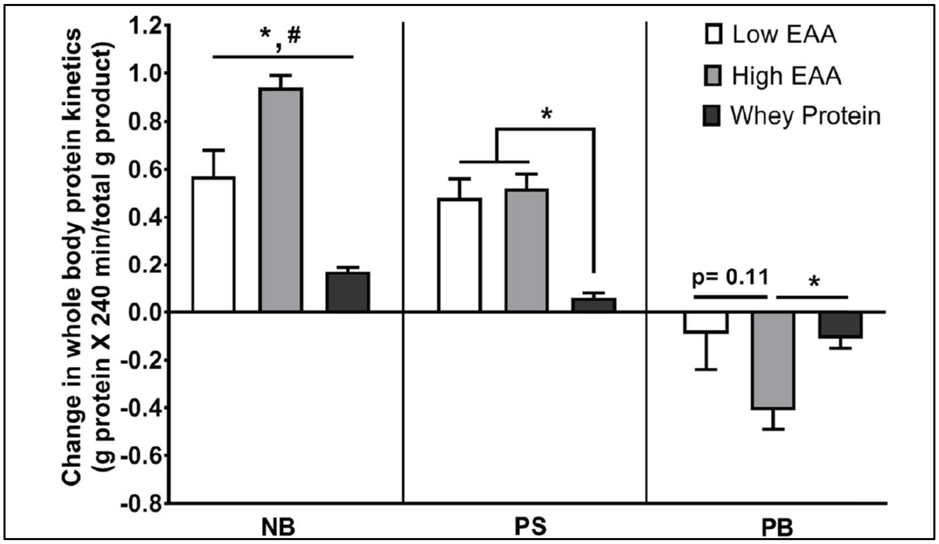


The relationship between muscle protein synthesis (protein synthesis minus breakdown) and the rate of intracellular appearance of amino acids. Rates were determined in human subjects using a three-pool model of leg protein metabolism. Source: Nicolaas and Wolfe. [387]

Further, Kim et al., supported this extended protein anabolic theory by comparing the anabolic response of 40 g vs. 70 g of protein following exhaustive resistance exercise on NPB and found that NPB was more positive in the higher protein group due to a greater suppression of whole-body protein breakdown, with a significant increase in whole-body protein synthesis.[210] On the other hand, Stokes et al. argued that suppressing the normal exercise-induced rise in MPB (especially important to initiating global remodeling, at least in early phases of resistance training) is not necessary and probably yields no physiological benefit making the concept of increasing protein (or insulin) to reduce MPB to potentially increase longer-term SM accretion (or NPB), a moot point as it relates to SM hypertrophy.[376] In fact, in the novice exerciser (unaccustomed activities), resistance training induced MPB necessarily potentiates a global remodeling of all muscle fractions, which robustly stimulates MPS.[197,202] As the novice exerciser transitions to experienced, MPB decreases and overall MPS is attenuated but directed more towards myofibrillar adaptations than earlier global remodeling.[202,393,394] This argues for no need to suppress MPB with nutritional interventions such as insulin stimulation or AAs beyond that necessary for SM MPS.[376] However, most recently, Park et al., compared the whole-body anabolic response in three treatments: intact whey protein supplementation (WPS) alone, and WPS combined with 6 g and 12 g of EAAs, measuring whole-body protein kinetics.[128] They found a dose-dependent greater anabolic response with the addition of EAAs. The increase in net balance between whole-body protein synthesis and breakdown was greatest in the high-dose EAA/WPS subjects. The greater anabolic response was due to greater increases in whole-body protein synthesis (three-fold anabolic response increase in the 6 g group and 6-fold in 12 g subjects) and a markedly greater suppression of whole-body protein breakdown (see Figure 12). Further, as shown in Figure 13, in the high dose group, the authors showed the muscle protein FSR reflected the changes in whole-body protein synthesis, also documenting a significant increase in the muscle FSR in a dose dependent manner.[128] Their conclusion appears to support protein's anabolic limits extend beyond the common "muscle full" FSR measurements and may include its components (EAA) contribution to suppressing MPB since the high dose did deliver a measurable increase in both NPB and FSR, thus may have been partially due to the greater suppression of MPB (mindful that it would take ~25 g of intact whey protein to supply the complement 12 g of EAA used in this treatment – i.e., added to the intact WPS). To be sure, the fact that the well-known accelerated muscle building effects of anabolic steroids are partially due to their anti-catabolic/nitrogen sparing effects, thus significantly suppressing MPB during strenuous exercise, gives credence to the use of nutritional interventions, albeit through different mechanisms, to also reduce MPB to under similar circumstances to further increase exercise-induced net SM protein gains.[395,396,397]

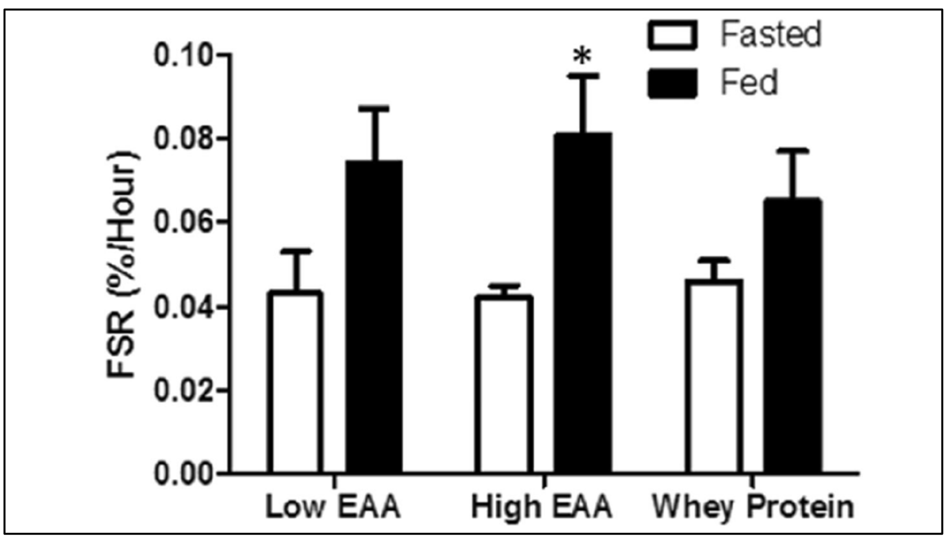
The practicality in real life to the question: "what is protein's anabolic threshold?" probably only matters to those athletes that may control their protein intake per meal based on the old school 20-30 g meal limit because they may believe that more is wasted or something bad might happen if you eat more. Unless you are consuming complete protein foods to the extent where you are replacing other healthy foods within your caloric allotment (or sport requirement needs based on macronutrient recommendations by sport), the athlete won't hinder their health or performance consuming slightly or moderately more than old-school or current "muscle full" guidelines.[16,24,28,30,33,36,37,38,39,40] Additionally, depending on individuality, slightly higher intakes per meal as shown here, may offer the potential to improve desired gains long-term. Mindful that SM makes up ~25% of whole-body proteins,[398] the relevance to hypertrophy in the short-term from consuming more than the old school muscle full 20-30 g, may be unmeasurable but indeed, if this whole body NPB improvement discussed here, contributes to better overall recovery, there may be a positive accumulating effect over time, thus extending and improving performance progression or helping prevent injury.

Figure 12 - Net Protein Gain from EAA Combined with WPS and WPS Alone



Source: Park et al.[128] Changes from baseline of whole-body net protein balance (NB), protein synthesis (PS) and protein breakdown (PB) following ingestion of the free EAAs/WPS composition (6.3 g and 12.6 g) and the whey protein product (17 g). Values are normalized for the amount of product consumed. *Statistically different from High EAA; #Statistically different between Low EAA and whey protein.

Figure 13 - Changes in Fractional Synthesis Rate (FSR) from EAA Combined with WPS and WPS Alone



Source: Park et al.[128] Muscle protein fractional synthesis rate (FSR) following consumption one of two doses of the free EAAs/protein composition (6.3 g and 12.6 g) and the whey protein product (17.9 g). *Statistically significant from fasted within treatment.

Summary

Like most nutrition recommendations, protein requirements are individual including being partially related to preference, and researchers are still in discovery of protein's anabolic threshold per meal (3 to 4-hour intervals), based on defining the anabolic response as whole-body protein turnover or net protein balance. MPB is unequivocally part of the MPS process for all humans, and whether using nutritional interventions (high protein or EAA intake) to reduce

MPB from exercise induced damage to yield a better MPS/recovery response, remains unknown but appears likely and may be important to the competitive lifespan of athletes. Nevertheless, in the properly fed athlete, meeting daily protein requirements (1 g/lb/LBM or body weight) divided between four to six meals will cover any differences in scientific opinions. Moreover, no matter the final destinations, almost all protein/AAs consumed will be utilized somewhere with higher than recommended amounts for supporting MPS deemed safe. Lastly, going beyond protein's anabolic contribution may have other personalized benefits such as satiety and lifetime weight control.
In summary, the current scientific consensus is as follows: the amount of protein per feeding (~3-to-4-hour intervals) that maximizes recovery including the desired exercise-induced musculoskeletal/ cardiovascular adaptations (e.g., hypertrophy, BMD, muscular endurance, etc.) is proposed to be **0.18 to .25 g/lb of LBM (0.4 to .55 g/kg body weight).**

Protein Safety and Upper Limit

Throughout each section of this document, protein intake safety and efficacy are discussed and referenced at or above the new proposed recommendations (1-1.2 g/lb/LBM/day) for each circumstance including age groups, with no known adverse effects. Additionally there is no established Tolerable Upper Limit (UL) for protein and a wide range of daily intake is now within the DGAs.[16,399] The Institute of Medicine's (IOM) review of studies examining adverse effects of high-protein diets was unable to identify a level of daily protein that increased the risk of health problems including renal, osteoporosis, kidney stones, obesity, etc.[399] One common question has always been the amount of protein that negatively effects renal function. Currently, there is no evidence that increased urea formation or changes in glomerular filtration rate from protein intake beyond nitrogen balance or within DGA guidelines (10-35% of total calorie intake) causes kidney damage in healthy persons.[400,401,402] In fact, clearance becomes more efficient with higher protein intakes.[33,403] Bone health was another common concern with high protein intakes. A systematic review by Darling et al. on the subject determined there was insufficient evidence that high protein intakes effect bone health either positively or negatively,[404] and recently possibly positive,[405] and especially with adequate dietary calcium.[406,407] The Groenendijk et al. systematic review supports that a protein intake above the current RDA may reduce hip fracture risk and may play a beneficial role in bone mineral density (BMD) maintenance and loss in older adults.[343] The bottom line is that chronic protein intake two to four times the RDA and up to 35% of daily energy intake is shown to be safe and effective for healthy individuals as long as protein is not replacing other necessary nutritious foods.[16,24,28,30,33,36,37,38,39,40,107,341,342,399,403]

Carbohydrate –Maltodextrins

WheySmooth is primarily a fast-acting protein supplement. The carbohydrate content in WheySmooth is strategically designed to not only allow the whey protein to maintain its natural quick absorption/utilization rate, but also: 1) for minimal calorie contribution allowing adjustments (added foods/fluids into mix) as desired; 2) rapid gastric emptying thus oxidation during pre/post workout periods to help maximize MPS within the proposed "anabolic windows"; 3) flavor and easy mixing properties.

Maltodextrin

Maltodextrin is a polysaccharide. It is a lightly hydrolyzed starch used as an ingredient in many food products as a thickener and carbohydrate source.[408] Maltodextrin is easily digestible, being absorbed as rapidly as glucose but moderately sweet or sometimes bland making it desirable in food manufacturing.[408] Carbohydrates in sports are generally placed in two categories. Those that can be oxidized (used for energy) rapidly (up to ~60 g/hr) and those oxidized slower (up to ~40 g/hr.).[409] Maltodextrins like glucose, maltose and sucrose fall in the rapid category. These carbohydrates are digested and absorbed at rapid rates making them readily available to working muscle and, when in small amounts as in WheySmooth, do not slow down amino acid absorption from protein.[51] These qualities, including maltodextrin's food mixture compatibility, make them ideal in a product like WheySmooth that is designed as a low calorie (but flexible) high protein meal supplement and pre/post workout protein supplement.[408,409,410]

Co-factors Including Sweeteners

Co-factors in a protein powder are combined to deliver better taste with low calories, texture, mixing ability, uniform nutrient distribution, ingredient flow and stability, including during cooking or baking, and a practical product shelf life.

Sweeteners Background

Health outcomes or adverse reactions to natural and/or added caloric sweeteners (CS), such as refined sugars, honey, syrups, fruit sugars, sucrose and their constituent molecules, include, but are not limited to obesity, blood sugar spikes, tooth decay and allergic responses from the residues from their sources of origin, has spawned the need and growing use of non-nutritive sweeteners (NNS).[411,412,413,414,415,416,417] Further, these issues related to CS have led to prevention policies such as sugar-sweetened beverage taxes and front-of-package labels, may also be incentivizing companies to utilize NNS as a way of reducing CS.[418,419,420,421] (New FDA label format can be viewed here.) Recognizing there are industry and cult biases towards the use of NNS (none have been validated or accepted),[422,423,424,425,426] the major scientific bodies around the world have firmly established their safety including for use with children,[427,428,429,430,431,432,433] thus require no label warnings as added CS and other natural ingredients (e.g. peanuts, shell fish, wheat, dairy, etc.)[416,418,419,420]
In summary, FDA approved NNS advantages over CS include:

- Higher nutrition per calorie/sweetness to support desired body composition, especially when used to replace CS[434,435]
    - Sucralose tested best of NNS and sucrose in weight management[435]
- Supports blood sugar (approved for diabetics)[423,428,434]
- Lower risk of adverse reactions common with "natural" sweeteners (honey, stevia, lactose, fruit sugar residues, etc.)[415,416]
- Supports weight control versus being a contributing factor in weight gain[429,434,436,437,438,439,440]
- Approved for children[429,430,431]

Finally, the Martyn et al. review titled "Low-/No-Calorie Sweeteners: A Review of Global Intakes" concluded that "Overall, the studies conducted since 2008 raised no concerns with respect to exceedance of individual sweetener acceptable daily intake (ADIs) among the general population globally. Additionally, the data identified do not suggest a shift in exposure over time, with several studies indicating a reduction in intake."[441]

Sweeteners in WheySmooth

Sweeteners used in WheySmooth appear at the end of the ingredient list as they are in minute amounts per serving and inert in human metabolism thus no effects within the body other than taste.[442] For frequently asked questions (FAQs) on non-nutritive sweeteners click here. Non-nutritive sweeteners (NNS) are those that sweeten with minimal or no carbohydrate or energy. NNS are regulated by the Food and Drug Administration (FDA) as food additives.[427] The FDA approval process includes determination of probable intake, cumulative effect from all uses and toxicology studies.[427,428] Eight NNS (aspartame, acesulfame potassium, luo han guo [monk] fruit extract, neotame, saccharin, stevia, sucralose and advantame) are approved for use in the United States (click here for a list, uses and metabolism) with acesulfame K and sucralose being among the most popular[414] largely because of their unique functional properties in enhancing food products including taste.[414] As with any fitness supporting food product, the better the taste and versatility, the greater chance of sustained use to support health and fitness goals.

Acesulfame Potassium (Ace-K)

Acesulfame potassium (chemical formula C4H4KNO4S; CAS registry number 55589-62-3) is approximately 200 times sweeter than sugar and is often combined with other sweeteners as an additional flavor enhancer in foods because it is heat stable during baking and environmentally friendly.[443,444] Ace-K is typically used in frozen desserts, candies, beverages, and baked goods. More than 90 studies support its safety and is used in WheySmooth to support baking capacity and sweetness.[445] For a complete current review on Ace-K, readers are referred to Belton et al.'s "A Review of the Environmental Fate and Effects of Acesulfame-Potassium."[444]

Sucralose

Sucralose is also a NNS, and is made from sucrose by a process that substitutes three chloride atoms for three hydroxyl groups on the sucrose molecule.[446,447] Sucralose is a very versatile NNS that is 450–650 times sweeter than sucrose, possesses a pleasant sweet taste and a quality and time intensity profile that is close to that of sucrose making a popular NNS.[414,448] Sucralose has been extensively studied with more than 110 safety studies reviewed by the FDA in approving the use of sucralose as a general purpose sweetener for food.[427,428,446,449] A primary advantage of sucralose for consumers is its exceptional stability. It retains its sweetness over a wide range of temperature and storage conditions and in solutions over time. This stability allows manufactures to create greater tasting foods and beverages and maintain the fresh flavor. Like Ace-K, sucralose is heat stable, meaning that it stays sweet even when used at high temperatures during baking, making it a common sugar substitute in baked goods.[448,450]
The FDA established an acceptable daily intake (ADI) for sucralose of 5 milligrams per kilogram (Europe's is 7 mg/kg and Canada's is 11 mg/kg) of body weight (mg/kg) per day.*[427] The amount of sucralose per serving in WheySmooth is ~34 mg.

Out of safety precautions to protect all sub-groups of people,* ***the ADI represents an amount 100 times less than the quantity of sucralose found to be safe in research studies**.[427] For a person weighing 150 pounds (68 kg), the US ADI equates to 340 mg of sucralose—the amount found in nine cans of diet soda or more than 28 individual packets of sucralose—consumed, on average, every day over a lifetime.*

Carboxymethyl Cellulose

Carboxymethyl cellulose (CMC) or cellulose gum is a popular non-toxic cellulose (fiber) derivative, an FDA approved food additive and on the generally recognized as safe list (GRAS).[451] CMC is used in food as a viscosity modifier or thickener, and to stabilize emulsions (emulsifier) in food products.[452,453] CMC is used extensively in gluten-free and reduced fat food products such as WheySmooth.[454] Use of CMC also ensures smooth dispersion in flavor oils, and improves texture and overall quality.[453,454]

Xanthan Gum (XG)

Xanthan gum is a water soluble, high molecular weight natural polysaccharide produced by a fermentation process.[455] Due to its high molecular weight (2.0×10^6–2.0×10^7 Da) and unique chemistry, xanthan gum shows excellent pseudoplasticity, thickening, and rheological properties, and is highly stable to heat, acid, and alkali making it ubiquitous in food products.[456,457,458] Because of its safety profile the United States FDA approved xanthan gum as a food additive in 1969, and European countries followed suit where it is primarily used as a molding agent, stabilizer, viscosifier, and thickener.[459,460,461] Additionally, small amounts of XG can enhance taste and prevent insoluble ingredients in juice-type beverages from precipitating. Due to its soft texture and ability to function as a stabilizer it is used for many different formulations with applications in pharmaceuticals, dietary supplements, and food products such as WheySmooth.[462]

WheySmooth Summary

Compared to other proteins, gram per gram, whey protein has been shown to be superior in delivering muscle protein synthesis (MPS), health and weight control outcomes based on its structure and subsequent unique functional properties such as: 1) higher EAA content (12.4 g/25 g); 2) higher BCAA (5.6 g/25 g); 3) higher leucine (3 g/25 g); 4) faster digestion/absorption to timely amplify MPS around exercise; 5) less splanchnic AA extraction so more AA are directly available for MPS; 6) whey concentrate (WC), along with the AAs, also contains whey's natural unique growth and health/immune supporting molecules along with 200 mg of calcium and 224 mg of potassium per serving. WheySmooth (WS) uses an ion-exchange instantized protein blend containing 90% whey concentrate, 5% whey isolate (cold filtered) and 5% casein for immediate and extended release and easy mixing along with co-

factors that give the product its desirable taste, texture, uniformity, and stability. The WS ingredients and macronutrient profile of low fat, carbohydrate and calorie per protein amount (25 g protein, 7 g carbohydrate, 2 g fat) make it an ideal protein source for weight/fat conscious exercisers/athletes to use as a pre/post workout supplement and/or integrated as described above into a weight control daily meal plan.
WS in its native high protein, low calorie powdered form, including chocolate, vanilla, unflavored and all-natural versions, can serve as the starting ingredients for the user to add as desired (e.g., fruits, vegetables, dairy, etc.) to complete a healthy meal/shake – i.e., serves as a tasty delivery system to include important foods not always regularly consumed. Further, WS's accompanying ingredients allow for easy mixing and ideal for baking.
Finally, since WS is a convenient, flexible and superior low calorie protein source for the stimulation of MPS and supplementing the diet, regular use could act as a positive influence on the regulation of muscle mass, overall health and weight control across the lifespan.

Typical Use
WheySmooth™ (WS) is ideal for persons, including athletes or exercisers, seeking a protein source with the highest biological value rating (104) because of its gram per gram ability to meet the body's AA requirements, especially the EAAs including leucine, contained in a very low-calorie mix that can be adjusted to maximize training induced size, performance, strength and body composition outcomes.
- Low calorie, high protein source to support any goal because it can be adjusted as desired including adding other nutrition components.
- Anyone pursuing weight/fat loss as an ideal high protein, low calorie protein source.
- Anyone throughout life who is not meeting protein requirements for specific goals including aging.
- As a pre/post workout supplement for anyone especially physique competitors or other weight/body-fat conscious athletes during the final weeks of competition dieting, to help meet expanded protein requirements with fewer calories.
- Anyone wanting a great tasting, flexible (add desired nutrition to the high protein, low calorie mix) convenient (portable), high-quality protein source including all-natural versions.
- Fortifying foods such as in baking/cooking (e.g., muffins, pancakes, breads, cookies, brownies, etc.) to increase protein content per serving.
- As a protein supplement to help ensure meeting new recommended daily protein needs, WS can reduce monthly food bills while delivering higher nutrition in fewer calories that many popular food protein sources.

Precautions (see Protein Safety and Upper Limit Section)
Older data suggested an increase in calcium loss with high protein intakes may negatively affect bone health.[463] However, newer studies have found the link between high protein intake (above the RDA) and bone health to be positive[343,405,406,407,464,465] or no effect.[404,406] The Institute of Medicine's and other related studies have concluded that levels of dietary protein are not associated with a decrease in renal function with age.[28,33,399,400,401,402,403,466,467,468,469]

Contraindications
There is negligible lactose in WheySmooth (removed during production), therefore it would only be contraindicated in people unable to consume milk proteins.[72,73]

Adverse Reactions
There should be no adverse effects in healthy users at the recommended doses unless allergic to milk proteins.

Upper Limit/Toxicity

Currently there is no upper limit established for protein. Further, chronic protein intake two to four times the RDA and up to 35% of daily energy intake is shown to be safe and effective for healthy individuals as long as protein is not replacing other necessary nutritious foods.[16,24,28,30,33,36,37,38,39,40,107,341,342,399]

Summary

Purpose

- Because of whey protein's constituent essential amino acid content, including BCAAs (especially leucine) and other functional components and rapid skeletal muscle bioavailability, gram for gram it is superior to other protein sources in potentiating a greater muscle protein synthesis (MPS) and health response per calories ingested.
- Fast acting, low calorie and highly anabolic pre- and post-workout supplement for athletes to maximize MPS during restricted calorie intake to help maintain and timely amplify MPS during fat/weight loss (e.g., physique competitors, fighters/wrestlers, weightlifters, etc.).
- Especially important for older athletes seeking physical improvements as the body becomes more resistant to anabolic effects of food and exercise.
- Used as a primary protein source during a meal replacement integrated diet and weight loss program to establish the best possible outcome (e.g., appetite and calorie control, preservation of LBM, greater fat oxidation, etc.).
- Regular use throughout the aging process to support newer expert protein requirements necessary to support lean body mass (LBM) body mass in older adults.
- As a protein supplement to help ensure meeting new recommended daily protein needs, WS can reduce monthly food bills while delivering higher nutrition in less calories that many popular food protein sources.

Unique Features

- 25 grams of the highest biological value protein, 7 g of carbohydrate, 2.5 g of healthy fat in only 160 calories.
- Co-factors ensure nutrient uniformity and stability with great taste, easy mixing and baking qualities.
- No gas or bloating as is common with other protein powders.
- Contains only two grams of sugar.
- Aspartame free.
- NSF Certified for Sport (NSFCS), which is an additional product guarantee for drug tested athletes. Click here for the dotFIT NSFCS section.
- Formulated and manufactured for great taste and pleasing texture in a regularly inspected NSF certified facility, in compliance with Good Manufacturing Practices (GMPs) exclusively for dotFIT, LLC.

Supplement Facts Panel

Nutrition Facts

28 servings per container
Serving size 1 Scoop (41g)

Amount per serving
Calories 160

	% Daily Value*
Total Fat 3 g	**4%**
Saturated Fat 1g	**5%**
Trans Fat 0g	**
Cholesterol 60 mg	**20%**
Total Carbohydrate 7 g	**3%**
Dietary Fiber 1 g	**4%**
Total Sugars 2 g	
Protein 25 g	
Sodium 180 mg	**8%**
Vitamin D 0 mcg	0%
Calcium 200 mg	15%
Iron 0 mg	0%
Potassium 224 mg	5%

**Daily Value not established.

References

[1] Lopez MJ, Mohiuddin SS. Biochemistry, Essential Amino Acids. [Updated 2020 Apr 26]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2020 Jan-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK557845/
[2] Nix, Staci. Williams' Basic Nutrition and Diet Therapy, 15th Edition. 2017 Elsevier Inc. Part 1, Chapters 4 Proteins, pgs. 41-55. ISBN: 978-0-323-37731-7
[3] Ben-Tal, Nir, Kessel, Amit. Introduction to Proteins: Structure, Function, and Motion, Second Edition (Chapman & Hall/CRC Computational Biology Series). Amino Acids (2009) 37:1-17
[4] Bröer, Stefan, and Angelika Bröer. "Amino acid homeostasis and signaling in mammalian cells and organisms." *The Biochemical journal* vol. 474,12 1935-1963. 25 May. 2017, doi:10.1042/BCJ20160822
[5] Hou Y, Yin Y, Wu G. Dietary essentiality of "nutritionally non-essential amino acids" for animals and humans. Exp. Biol. Med. (Maywood). 2015 Aug;240(8):997-1007. [PMC free article: PMC4935284] [PubMed: 26041391]
[6] de Koning TJ. Amino acid synthesis deficiencies. Handb Clin Neurol. 2013;113:1775-83. [PubMed: 23622400]
[7] Baum JI, Kim IY, Wolfe RR. Protein Consumption and the Elderly: What Is the Optimal Level of Intake? Nutrients. 2016 Jun 8;8(6):359. doi: 10.3390/nu8060359. PMID: 27338461; PMCID: PMC4924200.
8 Kim IY, Park S, Smeets ETHC, Schutzler S, Azhar G, Wei JY, Ferrando AA, Wolfe RR. Consumption of a specially formulated mixture of essential amino acids promotes gain in whole-body protein to a greater extent than a complete meal replacement in older women with heart failure. Nutrients. 2019;11(6):1360.
[9] FAO. Dietary Protein Evaluation in Human Nutrition: Report of an FAO Expert Consultation 2011; FAO Food and Nutrition Paper 92; FAO: Rome, Italy, 2013
[10] Wolfe RR, Baum JI, Starck C, Moughan PJ. Factors contributing to the selection of dietary protein food sources. Clin Nutr. 2018 Feb;37(1):130-138. doi: 10.1016/j.clnu.2017.11.017. Epub 2017 Dec 6. PMID: 29233589
[11] Berrazaga, Insaf et al. "The Role of the Anabolic Properties of Plant- versus Animal-Based Protein Sources in Supporting Muscle Mass Maintenance: A Critical Review." *Nutrients* vol. 11,8 1825. 7 Aug. 2019, doi:10.3390/nu11081825
[12] Liao et al. Review. Prospective Views for Whey Protein and/or Resistance Training Against Age-related Sarcopenia. Aging and Disease. Volume 10, Number 1; 158-174. February 2019
[13] Laleg K., Cassan D., Barron C., Prabhasankar P., Micard V. Structural, culinary, nutritional and anti-nutritional properties of high protein, gluten free, 100% legume pasta. PLoS ONE. 2016;11:e0160721. doi: 10.1371/journal.pone.0160721
[14] Gorissen S.H.M., Witard O.C. Characterizing the muscle anabolic potential of dairy, meat and plant-based protein sources in older adults. Proc. Nutr. Soc. 2018;77:20–31. doi: 10.1017/S002966511700194X. [PubMed]
[15] WHO/FAO/UNU . Protein and Amino Acid Requirements in Human Nutrition. Report of the Joint FAO/WHO/UNU Expert Consultation. WHO; Geneva, Switzerland: 2007. (World Health Organization Technical Report Series 935).
[16] U.S. Department of Health and Human Services and U.S. Department of Agriculture. 2015–2020 Dietary Guidelines for Americans. 8th Edition. December 2015. Available at http://health.gov/dietaryguidelines/2015/guidelines/. Appendix 7- https://health.gov/our-work/food-nutrition/2015-2020-dietary-guidelines/guidelines/appendix-7/
[17] Optimizing Protein Intake in Adults: Interpretation and Application of the Recommended Dietary Allowance Compared with the Acceptable Macronutrient Distribution Range. *Adv Nutr*. 2017;8(2):266-275. Published 2017 Mar 15. doi:10.3945/an.116.013821
[18] Wirth J, Hillesheim E, Brennan L. The Role of Protein Intake and its Timing on Body Composition and Muscle Function in Healthy Adults: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. J Nutr. 2020 Jun 1;150(6):1443-1460. doi: 10.1093/jn/nxaa049. PMID: 32232404
[19] Berryman CE, Lieberman HR, Fulgoni VL 3rd, Pasiakos SM. Protein intake trends and conformity with the Dietary Reference Intakes in the United States: analysis of the National Health and Nutrition Examination Survey, 2001-2014. *Am J Clin Nutr*. 2018;108(2):405-413. doi:10.1093/ajcn/nqy088
[20] Kerksick, Chad M et al. "ISSN exercise & sports nutrition review update: research & recommendations." *Journal of the International Society of Sports Nutrition* vol. 15,1 38. 1 Aug. 2018, doi:10.1186/s12970-018-0242-y
[21] Antonio J. High-protein diets in trained individuals. Res Sports Med. 2019 Apr-Jun;27(2):195-203. doi: 10.1080/15438627.2018.1523167. Epub 2018 Sep 22. PMID: 30244609.
[22] Davies RW, Carson BP, Jakeman PM. The Effect of Whey Protein Supplementation on the Temporal Recovery of Muscle Function Following Resistance Training: A Systematic Review and Meta-Analysis. Nutrients. 2018 Feb 16;10(2):221. doi: 10.3390/nu10020221. PMID: 29462923; PMCID: PMC5852797.

[23] Eric T Trexler1, Abbie E Smith-Ryan1* and Layne E Norton. Metabolic adaptation to weight loss: implications for the athlete. Trexler et al. Journal of the International Society of Sports Nutrition 2014, 11:7 http://www.jissn.com/content/11/1/7
[24] Jäger R, Kerksick CM, Campbell BI, et al. International Society of Sports Nutrition Position Stand: protein and exercise. *J Int Soc Sports Nutr*. 2017;14:20. Published 2017 Jun 20. doi:10.1186/s12970-017-0177-8
[25] Hector AJ, Phillips SM. Protein Recommendations for Weight Loss in Elite Athletes: A Focus on Body Composition and Performance. *Int J Sport Nutr Exerc Metab*. 2018;28(2):170-177. doi:10.1123/ijsnem.2017-0273
[26] Johnston BC , Kanters S , Bandayrel K , Wu P , Naji F , Siemieniuk RA , Ball GD, et al. Comparison of weight loss among named diet programs in overweight and obese adults: a meta-analysis. JAMA. 2014 Sep 3;312(9):923-33. doi: 10.1001/jama.2014.10397
[27] van Baak MA, Mariman ECM. Dietary Strategies for Weight Loss Maintenance. Nutrients. 2019 Aug 15;11(8):1916. doi: 10.3390/nu11081916. PMID: 31443231; PMCID: PMC6722715
[28] Nancy R Rodriguez and Sharon L Miller. Effective translation of current dietary guidance: understanding and communicating the concepts of minimal and optimal levels of dietary protein. Am J Clin Nutr 2015;101(Suppl):1353S–8S. Printed in USA. _ 2015 American Society for Nutrition 1353S
[29] Yang Y, Breen L, Burd NA, Hector AJ, Churchward-Venne TA, Josse AR, Tarnopolsky MA, Phillips SM. Resistance exercise enhances myofibrillar protein synthesis with graded intakes of whey protein in older men. Br J Nutr. 2012 Nov 28;108(10):1780-8. doi: 10.1017/S0007114511007422. Epub 2012 Feb 7.
[30] Douglas Paddon-Jones, Wayne W Campbell, Paul F Jacques, Stephen B Kritchevsky, Lynn L Moore, Nancy R Rodriguez, and Luc JC van Loon. Protein and healthy aging. Am J Clin Nutr 2015;101(Suppl):1339S–45S.
[31] Lonnie M, Hooker E, Brunstrom JM, Corfe BM, Green MA, Watson AW, Williams EA, Stevenson EJ, Penson S, Johnstone AM. Protein for Life: Review of Optimal Protein Intake, Sustainable Dietary Sources and the Effect on Appetite in Ageing Adults. Nutrients. 2018 Mar 16;10(3):360. doi: 10.3390/nu10030360. PMID: 29547523; PMCID: PMC5872778.
[32] Martone AM, Marzetti E, Calvani R, Picca A, Tosato M, Santoro L, Di Giorgio A, Nesci A, Sisto A, Santoliquido A, Landi F. Exercise and Protein Intake: A Synergistic Approach against Sarcopenia. Biomed Res Int. 2017;2017:2672435. doi: 10.1155/2017/2672435. Epub 2017 Mar 21. PMID: 28421192; PMCID: PMC5379082
[33] Donald K Layman, Tracy G Anthony, Blake B Rasmussen, Sean H Adams, Christopher J Lynch, Grant D Brinkworth, and Teresa A Davis. Defining meal requirements for protein to optimize metabolic roles of amino acids. Am J Clin Nutr 2015;101(Suppl):1330S–8S.
[34] Pasiakos SM. Metabolic advantages of higher protein diets and benefits of dairy foods on weight management, glycemic regulation, and bone. J Food Sci. 2015 Mar;80 Suppl 1:A2-7. doi: 10.1111/1750-3841.12804
[35] Presented at the conference “Protein Summit 2.0: Evaluating the Role of Protein in Public Health,” held in Washington, DC, 2 October 2013
[36] Bauer J, Biolo G, Cederholm T, Cesari M, Cruz-Jentoft A, Morley J, Phillips S, Sieber C, Stehle P, Teta D, et al. 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 2013;14:542–59.
[37] van Zanten ARH, Petit L, De Waele J, Kieft H, de Wilde J, van Horssen P, Klebach M, Hofman Z. Very high intact-protein formula successfully provides protein intake according to nutritional recommendations in overweight critically ill patients: a double-blind randomized trial. Crit Care. 2018 Jun 12;22(1):156. doi: 10.1186/s13054-018-2070-5. PMID: 29895309; PMCID: PMC5998555.
[38] Rizzoli R, Biver E, Bonjour JP, Coxam V, Goltzman D, Kanis JA, Lappe J, Rejnmark L, Sahni S, Weaver C, Weiler H, Reginster JY. Benefits and safety of dietary protein for bone health-an expert consensus paper endorsed by the European Society for Clinical and Economical Aspects of Osteoporosis, Osteoarthritis, and Musculoskeletal Diseases and by the International Osteoporosis Foundation. Osteoporosis Int. 2018 Sep;29(9):1933-1948. doi: 10.1007/s00198-018-4534-5. Epub 2018 May 8. PMID: 29740667.
[39] Tiller NB, Roberts JD, Beasley L, Chapman S, Pinto JM, Smith L, Wiffin M, Russell M, Sparks SA, Duckworth L, O'Hara J, Sutton L, Antonio J, Willoughby DS, Tarpey MD, Smith-Ryan AE, Ormsbee MJ, Astorino TA, Kreider RB, McGinnis GR, Stout JR, Smith JW, Arent SM, Campbell BI, Bannock L. International Society of Sports Nutrition Position Stand: nutritional considerations for single-stage ultra-marathon training and racing. J Int Soc Sports Nutr. 2019 Nov 7;16(1):50. doi: 10.1186/s12970-019-0312-9. PMID: 31699159; PMCID: PMC6839090
[40] Devries MC, Sithamparapillai A, Brimble KS, Banfield L, Morton RW, Phillips SM. Changes in Kidney Function Do Not Differ between Healthy Adults Consuming Higher- Compared with Lower- or Normal-Protein Diets: A Systematic Review and Meta-Analysis. J Nutr. 2018 Nov 1;148(11):1760-1775. doi: 10.1093/jn/nxy197. PMID: 30383278; PMCID: PMC6236074.

[41] Davani-Davari D, Karimzadeh I, Sagheb MM, Khalili H. The Renal Safety of L-Carnitine, L-Arginine, and Glutamine in Athletes and Bodybuilders. J Ren Nutr. 2019 May;29(3):221-234. doi: 10.1053/j.jrn.2018.08.014. Epub 2018 Oct 16. PMID: 30341034.
[42] Trushina EN, Vybornov VD, Riger NA, Mustafina OK, Solntseva TN, Timonin AN, Zilova IS, Radzhabkadiev RM. [The efficiency of branched chain amino acids (BCAA) in the nutrition of combat sport athletes]. Vopr Pitan. 2019;88(4):48-56. Russian. doi: 10.24411/0042-8833-2019-10041. Epub 2019 Jul 15. PMID: 31722141.
[43] Institute of Medicine. Dietary Reference Intakes for energy, carbohydrate, fiber, fat, fatty acids, cholesterol, protein, and amino acids. Washington (DC): National Academies Press; 2005
[44] Fujita S, Dreyer HC, Drummond MJ, Glynn EL, Cadenas JG, Yoshizawa F, Volpi E, Rasmussen BB. Nutrient signaling in the regulation of human muscle protein synthesis. J Physiol 582: 813–823, 2007. [PMCID: PMC2075348] [PubMed: 17478528]
[45] Stuart M. Phillips. A Brief Review of Critical Processes in Exercise-Induced Muscular Hypertrophy. Sports Med (2014) 44 (Suppl 1):S71–S77. DOI 10.1007/s40279-014-0152-3
[46] Stefan M. Pasiakos. Exercise and Amino Acid Anabolic Cell Signaling and the Regulation of Skeletal Muscle Mass. *Nutrients* 2012, *4*, 740-758; doi:10.3390/nu4070740
[47] A Castro LH, S de Araújo FH, M Olimpio MY, B de B Primo R, T Pereira T, F Lopes LA, B S de M Trindade E, Fernandes R, A Oesterreich S. Comparative Meta-Analysis of the Effect of Concentrated, Hydrolyzed, and Isolated Whey Protein Supplementation on Body Composition of Physical Activity Practitioners. Nutrients. 2019 Sep 2;11(9):2047. doi: 10.3390/nu11092047. PMID: 31480653; PMCID: PMC6769754.
[48] Ramdath DD, Padhi EM, Sarfaraz S, Renwick S, Duncan AM. Beyond the Cholesterol-Lowering Effect of Soy Protein: A Review of the Effects of Dietary Soy and Its Constituents on Risk Factors for Cardiovascular Disease. Nutrients. 2017 Mar 24;9(4):324. doi: 10.3390/nu9040324. PMID: 28338639; PMCID: PMC5409663.
[49] Valenzuela PL, Mata F, Morales JS, Castillo-García A, Lucia A. Does Beef Protein Supplementation Improve Body Composition and Exercise Performance? A Systematic Review and Meta-Analysis of Randomized Controlled Trials. Nutrients. 2019 Jun 25;11(6):1429. doi: 10.3390/nu11061429. PMID: 31242624; PMCID: PMC6628355.
[50] Winder Tadeu Silva Ton, Crislaine das Graças de Almeida, Leandro de Morais Cardoso, Yassana Marvila Girondoli, et al. Effect of different protein types on second meal postprandial glycaemia in normal weight and normoglycemic subjects. *(Nutr Hosp.* 2014;29:553-558) DOI:10.3305/NH.2014.29.3.7065
[51] Luiking YC, et al., Protein type and caloric density of protein supplements modulate postprandial amino acid profile through changes in gastrointestinal behaviour: A randomized trial, Clinical Nutrition (2015), http://dx.doi.org/10.1016/j.clnu.2015.02.013
[52] Devries MC , Phillips SM. Supplemental protein in support of muscle mass and health: advantage whey. J Food Sci. 2015 Mar;80 Suppl 1:A8-A15. doi: 10.1111/1750-3841.12802
[53] Nieman DC, Zwetsloot KA, Simonson AJ, Hoyle AT, Wang X, Nelson HK, Lefranc-Millot C, Guérin-Deremaux L. Effects of Whey and Pea Protein Supplementation on Post-Eccentric Exercise Muscle Damage: A Randomized Trial. Nutrients. 2020 Aug 9;12(8):2382. doi: 10.3390/nu12082382. PMID: 32784847; PMCID: PMC7468723.
[54] Oikawa SY, Kamal MJ, Webb EK, McGlory C, Baker SK, Phillips SM. Whey protein but not collagen peptides stimulate acute and longer-term muscle protein synthesis with and without resistance exercise in healthy older women: a randomized controlled trial. Am J Clin Nutr. 2020 Mar 1;111(3):708-718. doi: 10.1093/ajcn/nqz332. PMID: 31919527; PMCID: PMC7049534.
[55] Miller PE , Alexander DD, Perez V. Effects of whey protein and resistance exercise on body composition: a meta-analysis of randomized controlled trials. J Am Coll Nutr. 2014;33(2):163-75. doi: 10.1080/07315724.2013.875365
[56] Tahavorgar A , Vafa M , Shidfar F , Gohari M , Heydari I. Whey protein preloads are more beneficial than soy protein preloads in regulating appetite, calorie intake, anthropometry, and body composition of overweight and obese men. Nutr Res. 2014 Oct;34(10):856-61. doi: 10.1016/j.nutres.2014.08.015. Epub 2014 Sep 2
[57] Giglio BM, Schincaglia RM, da Silva AS, Fazani ICS, Monteiro PA, Mota JF, Cunha JP, Pichard C, Pimentel GD. Whey Protein Supplementation Compared to Collagen Increases Blood Nesfatin Concentrations and Decreases Android Fat in Overweight Women: A Randomized Double-Blind Study. Nutrients. 2019 Sep 2;11(9):2051. doi: 10.3390/nu11092051. PMID: 31480676; PMCID: PMC6770102.
[58] Li M , Liu F . Effect of whey protein supplementation during resistance training sessions on body mass and muscular strength: a meta-analysis. Food Funct. 2019 May 22;10(5):2766-2773. doi: 10.1039/c9fo00182d. PMID: 31041966.

[59] Park Y, Park HY, Kim J, Hwang H, Jung Y, Kreider R, Lim K. Effects of whey protein supplementation prior to, and following, resistance exercise on body composition and training responses: A randomized double-blind placebo-controlled study. J Exerc Nutrition Biochem. 2019 Jun 30;23(2):34-44. doi: 10.20463/jenb.2019.0015. PMID: 31337204; PMCID: PMC6651693.
[60] Kanda A, Nakayama K, Sanbongi C, Nagata M, Ikegami S, Itoh H. Effects of Whey, Caseinate, or Milk Protein Ingestion on Muscle Protein Synthesis after Exercise. Nutrients. 2016 Jun 3;8(6):339. doi: 10.3390/nu8060339. PMID: 27271661; PMCID: PMC4924180.
[61] Gorissen SHM, Trommelen J, Kouw IWK, Holwerda AM, Pennings B, Groen BBL, Wall BT, Churchward-Venne TA, Horstman AMH, Koopman R, Burd NA, Fuchs CJ, Dirks ML, Res PT, Senden JMG, Steijns JMJM, de Groot LCPGM, Verdijk LB, van Loon LJC. Protein Type, Protein Dose, and Age Modulate Dietary Protein Digestion and Phenylalanine Absorption Kinetics and Plasma Phenylalanine Availability in Humans. J Nutr. 2020 Aug 1;150(8):2041-2050. doi: 10.1093/jn/nxaa024. PMID: 32069356; PMCID: PMC7398787.
[62] Brennan J.L., Keerati-U-Rai M., Yin H., Daoust J., Nonnotte E., Quinquis L., St-Denis T., Bolster D.R. Differential Responses of Blood Essential Amino Acid Levels Following Ingestion of High-Quality Plant-Based Protein Blends Compared to Whey Protein-A Double-Blind Randomized, Cross-Over, Clinical Trial. Nutrients. 2019;11:2987. doi: 10.3390/nu11122987. [PMC free article]
[63] Vinicius Fernandes Cruzat, Maurício Krause, Philip Newsholme. Amino acid supplementation and impact on immune function in the context of exercise. *Journal of the International Society of Sports Nutrition* 2014, 11:61 doi:10.1186/s12970-014-0061-8
[64] B. Komar, L. Schwingshackl, Georg Hoffmann. Effects of leucine-rich protein supplements on anthropometric parameter and muscle strength in the elderly: A systematic review and meta-analysis. The Journal of Nutrition, Health & Aging. December 2014
[65] Tang JE[1], Moore DR, Kujbida GW, Tarnopolsky MA, Phillips SM. Ingestion of whey hydrolysate, casein, or soy protein isolate: effects on mixed muscle protein synthesis at rest and following resistance exercise in young men. J Appl Physiol (1985). 2009 Sep;107(3):987-92. doi: 10.1152/japplphysiol.00076.2009. Epub 2009 Jul 9.
[66] Rietsema S, Eelderink C, Joustra ML, van Vliet IMY, van Londen M, Corpeleijn E, Singh-Povel CM, Geurts JMW, Kootstra-Ros JE, Westerhuis R, Navis G, Bakker SJL. Effect of high compared with low dairy intake on blood pressure in overweight middle-aged adults: results of a randomized crossover intervention study. Am J Clin Nutr. 2019 Aug 1;110(2):340-348. doi: 10.1093/ajcn/nqz116. PMID: 31237322; PMCID: PMC6669052.
[67] Holland B., Rahimi Yazdi S., Ion Titapiccolo G., Corredig M. Short communication: Separation and quantification of caseins and casein macropeptide using ion-exchange chromatography. J. Dairy Sci. 2010;93:893–900. doi: 10.3168/jds.2009-2820. [PubMed]
[68] Macedo Mota, Lucio Flavio et al. "Genomic Analysis of Milk Protein Fractions in Brown Swiss Cattle." *Animals : an open access journal from MDPI* vol. 10,2 336. 20 Feb. 2020, doi:10.3390/ani10020336
[69] Ranchordas MK, Burd NA, Godfrey RJ, Senchina DS, Stear SJ, Burke LM, Castell LM: A-Z of nutritional supplements: dietary supplements, sports nutrition foods and ergogenic aids for health and performance: Part 43. Br J Sports Med 2013, 47:399–400
[70] Madureira A.R., Pereira C.I., Gomes A.M.P., Pintado M.E., Xavier Malcata F. Bovine whey proteins–Overview on their main biological properties. Food Res. Int. 2007;40:1197–1211. doi: 10.1016/j.foodres.2007.07.005.
[71] Marshall K: Therapeutic applications of whey protein. Altern Med Rev 2004, 9:136– 156.
[72] The Dairy Processing Handbook. Chapter 15, Whey Processing. Tetra Pak 2020 https://dairyprocessinghandbook.tetrapak.com/chapter/whey-processing
[73] Whey Proteins, From Milk to Medicine. 2019. Chapter 3 - Manufacture of Whey Protein Products: Concentrates, Isolate, Whey Protein Fractions and Microparticulated. Pages 97-122. Academic Press. Science direct. https://doi.org/10.1016/B978-0-12-812124-5.00003-5
[74] Głąb, Tomasz Konrad, and Janusz Boratyński. "Potential of Casein as a Carrier for Biologically Active Agents." *Topics in current chemistry (Cham)* vol. 375,4 (2017): 71. doi:10.1007/s41061-017-0158-z
[75] Mobley, C Brooks et al. "Effects of Whey, Soy or Leucine Supplementation with 12 Weeks of Resistance Training on Strength, Body Composition, and Skeletal Muscle and Adipose Tissue Histological Attributes in College-Aged Males." *Nutrients* vol. 9,9 972. 4 Sep. 2017, doi:10.3390/nu9090972
[76] Bell SJ. Whey protein concentrates with and without immunoglobulins: a review. J Med Food 2000;3:1-13.
[77] Banjare, Indrajeet Singh et al. "Physicochemical Properties and Oxidative Stability of Milk Fortified with Spray-Dried Whey Protein Concentrate-Iron Complex and *In Vitro* Bioaccessibility of the Added Iron." *Food technology and biotechnology* vol. 57,1 (2019): 48-58. doi:10.17113/ftb.57.01.19.5945
[78] Kiewiet MBG, Gros M, van Neerven RJJ, Faas MM, de Vos P. Immunomodulating properties of protein hydrolysates for application in cow's milk allergy. Pediatr Allergy Immunol. 2015 May;26(3):206-217. doi: 10.1111/pai.12354. PMID: 25692325.

[79] Kiewiet, M B Gea et al. "Toll-like receptor mediated activation is possibly involved in immunoregulating properties of cow's milk hydrolysates." *PloS one* vol. 12,6 e0178191. 8 Jun. 2017, doi:10.1371/journal.pone.0178191

[80] Capstone Nutrition. 900 S. DEPOT DRIVE OGDEN, UT 84404 (801) 337-9400. https://www.capstonenutrition.com/ NSF Certified and NSF Certified for Sport (NSFCS)

[81] Enattah N, Pekkarinen T, Välimäki MJ, et al. . Genetically defined adult-type hypolactasia and self-reported lactose intolerance as risk factors of osteoporosis in Finnish postmenopausal women. Eur J Clin Nutr 2005;59:1105–11. 10.1038/sj.ejcn.1602219 [PubMed]

[82] Misselwitz B, Fox M. What is normal and abnormal in lactose digestion? Lancet Gastroenterol Hepatol 2017;2:696–7. 10.1016/S2468-1253(17)30180-2 [PubMed]

[83] Misselwitz, Benjamin et al. "Update on lactose malabsorption and intolerance: pathogenesis, diagnosis and clinical management." *Gut* vol. 68,11 (2019): 2080-2091. doi:10.1136/gutjnl-2019-318404

[84] Willoughby D, Hewlings S, Kalman D. Body Composition Changes in Weight Loss: Strategies and Supplementation for Maintaining Lean Body Mass, a Brief Review. Nutrients. 2018 Dec 3;10(12):1876. doi: 10.3390/nu10121876. PMID: 30513859; PMCID: PMC6315740.

[85] Johnston BC , Kanters S , Bandayrel K , Wu P , Naji F , Siemieniuk RA , Ball GD, et al. Comparison of weight loss among named diet programs in overweight and obese adults: a meta-analysis. JAMA. 2014 Sep 3;312(9):923-33. doi: 10.1001/jama.2014.10397

[86] Longland TM, Oikawa SY, Mitchell CJ, Devries MC, Phillips SM. Higher compared with lower dietary protein during an energy deficit combined with intense exercise promotes greater lean mass gain and fat mass loss: a randomized trial. Am J Clin Nutr. 2016 Mar;103(3):738-46. doi: 10.3945/ajcn.115.119339. Epub 2016 Jan 27. PMID: 26817506.

[87] Ge L, Sadeghirad B, Ball GDC, da Costa BR, Hitchcock CL, Svendrovski A, Kiflen R, Quadri K, Kwon HY, Karamouzian M, Adams-Webber T, Ahmed W, Damanhoury S, Zeraatkar D, Nikolakopoulou A, Tsuyuki RT, Tian J, Yang K, Guyatt GH, Johnston BC. Comparison of dietary macronutrient patterns of 14 popular named dietary programmes for weight and cardiovascular risk factor reduction in adults: systematic review and network meta-analysis of randomised trials. BMJ. 2020 Apr 1;369:m696. doi: 10.1136/bmj.m696. Erratum in: BMJ. 2020 Aug 5;370:m3095. PMID: 32238384; PMCID: PMC7190064.

[88] Seimon RV, Wild-Taylor AL, Keating SE, McClintock S, Harper C, Gibson AA, Johnson NA, Fernando HA, Markovic TP, Center JR, Franklin J, Liu PY, Grieve SM, Lagopoulos J, Caterson ID, Byrne NM, Sainsbury A. Effect of Weight Loss via Severe vs Moderate Energy Restriction on Lean Mass and Body Composition Among Postmenopausal Women with Obesity: The TEMPO Diet Randomized Clinical Trial. JAMA Netw Open. 2019 Oct 2;2(10):e1913733. doi: 10.1001/jamanetworkopen.2019.13733. PMID: 31664441; PMCID: PMC6824325.

[89] Hudson JL, Bergia RE 3rd, Campbell WW. Effects of protein supplements consumed with meals, versus between meals, on resistance training-induced body composition changes in adults: a systematic review. Nutr Rev. 2018 Jun 1;76(6):461-468. doi: 10.1093/nutrit/nuy012. PMID: 29697807.

[90] Cava E, Yeat NC, Mittendorfer B. Preserving Healthy Muscle during Weight Loss. Adv Nutr. 2017 May 15;8(3):511-519. doi: 10.3945/an.116.014506. PMID: 28507015; PMCID: PMC5421125.

[91] Bendtsen LQ , Lorenzen JK , Gomes S , Liaset B , Holst JJ , Ritz C , Reitelseder S , Sjödin A , Astrup A. Effects of hydrolyzed casein, intact casein and intact whey protein on energy expenditure and appetite regulation: a randomised, controlled, cross-over study. Br J Nutr. 2014 Oct 28;112(8):1412-22. doi: 10.1017/S000711451400213X. Epub 2014 Sep 5

[92] Dominik H Pesta, and Varman T Samuel. A high-protein diet for reducing body fat: mechanisms and possible caveats. Pesta and Samuel Nutrition & Metabolism 2014, 11:53 http://www.nutritionandmetabolism.com/content/11/1/53

[93] Kjølbæk L, Sørensen LB, Søndertoft NB, Rasmussen CK, Lorenzen JK, Serena A, Astrup A, Larsen LH. Protein supplements after weight loss do not improve weight maintenance compared with recommended dietary protein intake despite beneficial effects on appetite sensation and energy expenditure: a randomized, controlled, double-blinded trial. Am J Clin Nutr. 2017 Aug;106(2):684-697. doi: 10.3945/ajcn.115.129528. Epub 2017 Jul 5. PMID: 28679554.

[94] Sutton EF, Bray GA, Burton JH, Smith SR, Redman LM. No evidence for metabolic adaptation in thermic effect of food by dietary protein. Obesity (Silver Spring). 2016 Aug;24(8):1639-42. doi: 10.1002/oby.21541. Epub 2016 Jun 29. PMID: 27356102; PMCID: PMC4963285.

[95] Dhillon J, Craig BA, Leidy HJ, Amankwaah AF, Osei-Boadi Anguah K, Jacobs A, Jones BL, Jones JB, Keeler CL, Keller CE, McCrory MA, Rivera RL, Slebodnik M, Mattes RD, Tucker RM. The Effects of Increased Protein Intake on Fullness: A Meta-Analysis and Its Limitations. J Acad Nutr Diet. 2016 Jun;116(6):968-83. doi: 10.1016/j.jand.2016.01.003. Epub 2016 Mar 3. PMID: 26947338.

[96] Andersson-Hall U, Pettersson S, Edin F, Pedersen A, Malmodin D, Madsen K. Metabolism and Whole-Body Fat Oxidation Following Postexercise Carbohydrate or Protein Intake. Int J Sport Nutr Exerc Metab. 2018 Jan 1;28(1):37-45. doi: 10.1123/ijsnem.2017-0129. Epub 2018 Jan 17. PMID: 28871893.
[97] Beaudry KM, Devries MC. Nutritional Strategies to Combat Type 2 Diabetes in Aging Adults: The Importance of Protein. Front Nutr. 2019 Aug 28;6:138. doi: 10.3389/fnut.2019.00138. PMID: 31555655; PMCID: PMC6724448.
[98] Camila L P Oliveira et al. A high-protein total diet replacement increases energy expenditure and leads to negative fat balance in healthy, normal-weight adults, *The American Journal of Clinical Nutrition*, Nov 2020 nqaa283, https://doi.org/10.1093/ajcn/nqaa283
[99] Kim JE, Sands L, Slebodnik M, O'Connor L, Campbell W: Effects of high-protein weight loss diets on fat-free mass changes in older adults: a systematic review. FASEB J 2014, 28(1 Suppl):371.5
[100] Mettler S, Mitchell N, Tipton KD: Increased protein intake reduces lean body mass loss during weight loss in athletes. Med Sci Sports Exerc 2010, 42:326–337.
[101] Lopes Gomes D, Moehlecke M, Lopes da Silva FB, Dutra ES, D'Agord Schaan B, Baiocchi de Carvalho KM. Whey Protein Supplementation Enhances Body Fat and Weight Loss in Women Long After Bariatric Surgery: a Randomized Controlled Trial. Obes Surg. 2017 Feb;27(2):424-431. doi: 10.1007/s11695-016-2308-8. PMID: 27885532.
[102] Verreijen AM, Engberink MF, Memelink RG, van der Plas SE, Visser M, Weijs PJ. Effect of a high protein diet and/or resistance exercise on the preservation of fat free mass during weight loss in overweight and obese older adults: a randomized controlled trial. Nutr J. 2017 Feb 6;16(1):10. doi: 10.1186/s12937-017-0229-6. PMID: 28166780; PMCID: PMC5294725.
[103] de Sousa MV, da Silva Soares DB, Caraça ER, Cardoso R. Dietary protein and exercise for preservation of lean mass and perspectives on type 2 diabetes prevention. Exp Biol Med (Maywood). 2019 Sep;244(12):992-1004. doi: 10.1177/1535370219861910. Epub 2019 Jul 15. PMID: 31307203; PMCID: PMC6879776.
[104] Zhou T, Qi L. Vitamin D, genetics, and bone mineral density during weight loss. Curr Opin Clin Nutr Metab Care. 2019 Nov;22(6):465-471. doi: 10.1097/MCO.0000000000000599. PMID: 31577641.
[105] Weiss EP, Jordan RC, Frese EM, Albert SG, Villareal DT. Effects of Weight Loss on Lean Mass, Strength, Bone, and Aerobic Capacity. Med Sci Sports Exerc. 2017 Jan;49(1):206-217. doi: 10.1249/MSS.0000000000001074. PMID: 27580151; PMCID: PMC5161655.
[106] Bosy-Westphal A, Kossel E, Goele K, et al. Contribution of individual organ mass loss to weight loss-associated decline in resting energy expenditure. Am J Clin Nutr. 2009;90(4):993–1001. [PubMed]
[107] Jiang BC, Villareal DT. Weight Loss-Induced Reduction of Bone Mineral Density in Older Adults with Obesity. J Nutr Gerontol Geriatr. 2019 Jan-Mar;38(1):100-114. doi: 10.1080/21551197.2018.1564721. Epub 2019 Feb 22. PMID: 30794099; PMCID: PMC6480356.
[108] Holderbaum M, Casagrande DS, Sussenbach S, Buss C. Effects of very low-calorie diets on liver size and weight loss in the preoperative period of bariatric surgery: a systematic review. Surg Obes Relat Dis. 2018 Feb;14(2):237-244. doi: 10.1016/j.soard.2017.09.531. Epub 2017 Oct 2. PMID: 29239795.
[109] Turicchi J, O'Driscoll R, Finlayson G, Duarte C, Hopkins M, Martins N, Michalowska J, Larsen TM, van Baak MA, Astrup A, Stubbs RJ. Associations between the proportion of fat-free mass loss during weight loss, changes in appetite, and subsequent weight change: results from a randomized 2-stage dietary intervention trial. Am J Clin Nutr. 2020 Mar 1;111(3):536-544. doi: 10.1093/ajcn/nqz331. PMID: 31950141.
[110] Pal S , Radavelli-Bagatini S , Hagger M , Ellis V. Comparative effects of whey and casein proteins on satiety in overweight and obese individuals: a randomized controlled trial. Eur J Clin Nutr. 2014 Sep;68(9):980-6. doi: 10.1038/ejcn.2014.84. Epub 2014 May 7.
[111] King DG, Walker M, Campbell MD, Breen L, Stevenson EJ, West DJ. A small dose of whey protein co-ingested with mixed-macronutrient breakfast and lunch meals improves postprandial glycemia and suppresses appetite in men with type 2 diabetes: a randomized controlled trial. Am J Clin Nutr. 2018 Apr 1;107(4):550-557. doi: 10.1093/ajcn/nqy019. PMID: 29635505.
[112] Chungchunlam SM, Henare SJ, Ganesh S, Moughan PJ. Effect of whey protein and a free amino acid mixture simulating whey protein on measures of satiety in normal-weight women. Br J Nutr. 2016 Nov;116(9):1666-1673. doi: 10.1017/S0007114516003767. Epub 2016 Nov 4. PMID: 27809945.

[113] Mollahosseini M, Shab-Bidar S, Rahimi MH, Djafarian K. Effect of whey protein supplementation on long- and short-term appetite: A meta-analysis of randomized controlled trials. Clin Nutr ESPEN. 2017 Aug;20:34-40. doi: 10.1016/j.clnesp.2017.04.002. Epub 2017 May 8. PMID: 29072167.
[114] Zhubi-Bakija F, Bajraktari G, Bytyçi I, Mikhailidis DP, Henein MY, Latkovskis G, Rexhaj Z, Zhubi E, Banach M; International Lipid Expert Panel (ILEP). The impact of type of dietary protein, animal versus vegetable, in modifying cardiometabolic risk factors: A position paper from the International Lipid Expert Panel (ILEP). Clin Nutr. 2020 May 26:S0261-5614(20)30251-X. doi: 10.1016/j.clnu.2020.05.017. Epub ahead of print. PMID: 32620446.
[115] Rigamonti AE, Leoncini R, Casnici C, Marelli O, Col A, Tamini S, Lucchetti E, Cicolini S, Abbruzzese L, Cella SG, Sartorio A. Whey Proteins Reduce Appetite, Stimulate Anorexigenic Gastrointestinal Peptides and Improve Glucometabolic Homeostasis in Young Obese Women. Nutrients. 2019 Jan 23;11(2):247. doi: 10.3390/nu11020247. PMID: 30678029; PMCID: PMC6412413.
[116] Hector AJ , Marcotte GR , Churchward-Venne TA , Murphy CH , Breen L , von Allmen M , Baker SK, Phillips SM. Whey protein supplementation preserves postprandial myofibrillar protein synthesis during short-term energy restriction in overweight and obese adults. J Nutr. 2015 Feb;145(2):246-52. doi: 10.3945/jn.114.200832. Epub 2014 Dec 17
[117] Verreijen AM , Verlaan S , Engberink MF , Swinkels S , de Vogel-van den Bosch J , Weijs PJ. A high whey protein-, leucine-, and vitamin D-enriched supplement preserves muscle mass during intentional weight loss in obese older adults: a double-blind randomized controlled trial. Am J Clin Nutr. 2015 Feb;101(2):279-86. doi: 10.3945/ajcn.114.090290. Epub 2014 Nov 26
[118] Wirunsawanya K, Upala S, Jaruvongvanich V, Sanguankeo A. Whey Protein Supplementation Improves Body Composition and Cardiovascular Risk Factors in Overweight and Obese Patients: A Systematic Review and Meta-Analysis. J Am Coll Nutr. 2018 Jan;37(1):60-70. doi: 10.1080/07315724.2017.1344591. Epub 2017 Oct 31. PMID: 29087242.
[119] Robert H Coker, Sharon Miller, Scott Schutzler, Nicolaas Deutz, and Robert R Wolfe. Whey protein and essential amino acids promote the reduction of adipose tissue and increased muscle protein synthesis during caloric restriction-induced weight loss in elderly, obese individuals. Nutr J. 2012; 11: 105. Published online 2012 December 11. doi: 10.1186/1475-2891-11-105PMCID: PMC3546025
[120] Morton, Robert W et al. “A systematic review, meta-analysis and meta-regression of the effect of protein supplementation on resistance training-induced gains in muscle mass and strength in healthy adults.” *British journal of sports medicine* vol. 52,6 (2018): 376-384. doi:10.1136/bjsports-2017-097608
[121] Tyler A. Churchward-Venne1, Nicholas A. Burd1, Cameron J. et al. Supplementation of a suboptimal protein dose with leucine or essential amino acids: effects on myofibrillar protein synthesis at rest and following resistance exercise in men. *Physiol* 590.11 (2012) pp 2751–2765
[122] Churchward-Venne TA , Breen L, Di Donato DM, Hector AJ, Mitchell CJ, Moore DR, Stellingwerff T, Breuille D, Offord EA, Baker SK, Phillips SM Leucine supplementation of a low-protein mixed macronutrient beverage enhances myofibrillar protein synthesis in young men: a double-blind, randomized trial. Am J Clin Nutr. 2014 Feb;99(2):276-86. doi: 10.3945/ajcn.113.068775. Epub 2013 Nov 27
[123] Liberman K, Njemini R, Luiking Y, Forti LN, Verlaan S, Bauer JM, Memelink R, Brandt K, Donini LM, Maggio M, Mets T, Wijers SLJ, Sieber C, Cederholm T, Bautmans I. Thirteen weeks of supplementation of vitamin D and leucine-enriched whey protein nutritional supplement attenuates chronic low-grade inflammation in sarcopenic older adults: the PROVIDE study. Aging Clin Exp Res. 2019 Jun;31(6):845-854. doi: 10.1007/s40520-019-01208-4. Epub 2019 May 2. PMID: 31049877; PMCID: PMC6583678.
[124] Xu ZR , Tan ZJ , Zhang Q , Gui QF , Yang YM. The effectiveness of leucine on muscle protein synthesis, lean body mass and leg lean mass accretion in older people: a systematic review and meta-analysis. Br J Nutr. 2014 Sep 19:1-10. [Epub ahead of print]
[125] Kobayashi H. [Amino Acid Nutrition in the Prevention and Treatment of Sarcopenia]. Yakugaku Zasshi. 2018;138(10):1277-1283. Japanese. doi: 10.1248/yakushi.18-00091-4. PMID: 30270272.
[126] Chanet A, Verlaan S, Salles J, Giraudet C, Patrac V, Pidou V, Pouyet C, Hafnaoui N, Blot A, Cano N, Farigon N, Bongers A, Jourdan M, Luiking Y, Walrand S, Boirie Y. Supplementing Breakfast with a Vitamin D and Leucine-Enriched Whey Protein Medical Nutrition Drink Enhances Postprandial Muscle Protein Synthesis and Muscle Mass in Healthy Older Men. J Nutr. 2017 Dec;147(12):2262-2271. doi: 10.3945/jn.117.252510. Epub 2017 Aug 23. PMID: 28835387.
[127] Tipton KD, Elliott TA, Ferrando AA, Aarsland AA, Wolfe RR. Stimulation of muscle anabolism by resistance exercise and ingestion of leucine plus protein. Appl Physiol Nutr Metab. 2009;34(2):151–61.

[128] Park, Sanghee et al. "Anabolic response to essential amino acid plus whey protein composition is greater than whey protein alone in young healthy adults." *Journal of the International Society of Sports Nutrition* vol. 17,1 9. 10 Feb. 2020, doi:10.1186/s12970-020-0340-5
[129] Borsheim E, Tipton KD, Wolf SE, Wolfe RR. Essential amino acids and muscle protein recovery from resistance exercise. Am J Physiol Endocrinol Metab. 2002;283(4):E648–57.
[130] Kari Johansson, Martin Neovius, and Erik Hemmingsson Effects of anti-obesity drugs, diet, and exercise on weight-loss maintenance after a very-low-calorie diet or low-calorie diet: a systematic review and meta-analysis of randomized controlled trials. Am J Clin Nutr 2014;99:14–23. Printed in USA. _ 2014 American Society for Nutrition
[131] Fatemeh Azizi Soeliman and Leila Azadbakht. Weight loss maintenance: A review on dietary related strategies. J Res Med Sci. 2014 Mar; 19(3): 268–275
[132] Heymsfield SB, van Mierlo CA, van der Knaap HC, Heo M, Frier HI. Weight management using a meal replacement strategy: meta and pooling analysis from six studies. Int J Obes Relat Metab Disord. 2003 May;27(5):537-49.
[133] Douketis JD, Macie C, Thabane L, Williamson DF. Systematic review of long-term weight loss studies in obese adults: clinical significance and applicability to clinical practice. Int J Obes (Lond). 2005 Oct;29(10):1153-67. Review.
[134] A. R. Leeds. Formula food-reducing diets: A new evidence-based addition to the weight management toolbox. 2014 The Author. *Nutrition Bulletin*, 39, 238–246. DOI: 10.1111/nbu.12098 MRs are part of maintenance.
[135] Heymsfield SB. Meal replacements and energy balance. Physiol Behav. 2010;100:90–4.
[136] Phelan S, Wing RR, Brannen A, McHugh A, Hagobian TA, Schaffner A, Jelalian E, Hart CN, Scholl TO, Munoz-Christian K, Yin E, Phipps MG, Keadle S, Abrams B. Randomized controlled clinical trial of behavioral lifestyle intervention with partial meal replacement to reduce excessive gestational weight gain. Am J Clin Nutr. 2018 Feb 1;107(2):183-194. doi: 10.1093/ajcn/nqx043. PMID: 29529157; PMCID: PMC6455030.
[137] Coleman CD, Kiel JR, Mitola AH, Arterburn LM. Comparative effectiveness of a portion-controlled meal replacement program for weight loss in adults with and without diabetes/high blood sugar. Nutr Diabetes. 2017 Jul 10;7(7):e284. doi: 10.1038/nutd.2017.32. PMID: 28692020; PMCID: PMC5549252.
[138] Astbury NM, Piernas C, Hartmann-Boyce J, Lapworth S, Aveyard P, Jebb SA. A systematic review and meta-analysis of the effectiveness of meal replacements for weight loss. Obes Rev. 2019 Apr;20(4):569-587. doi: 10.1111/obr.12816. Epub 2019 Jan 24. PMID: 30675990; PMCID: PMC6849863.
[139] Ard JD, Lewis KH, Rothberg A, Auriemma A, Coburn SL, Cohen SS, Loper J, Matarese L, Pories WJ, Periman S. Effectiveness of a Total Meal Replacement Program (OPTIFAST Program) on Weight Loss: Results from the OPTIWIN Study. Obesity (Silver Spring). 2019 Jan;27(1):22-29. doi: 10.1002/oby.22303. Epub 2018 Nov 13. PMID: 30421863; PMCID: PMC6587830.
[140] Maula A, Kai J, Woolley AK, Weng S, Dhalwani N, Griffiths FE, Khunti K, Kendrick D. Educational weight loss interventions in obese and overweight adults with type 2 diabetes: a systematic review and meta-analysis of randomized controlled trials. Diabet Med. 2020 Apr;37(4):623-635. doi: 10.1111/dme.14193. Epub 2019 Dec 22. PMID: 31785118; PMCID: PMC7154644.
[141] FORM FDA 3537. https://www.fda.gov/media/71632/download DIETARY CONVENTIONAL FOODS OR MEAL REPLACEMENTS (Includes Medical Foods) [21...Products 29. SNACK FOOD ITEMS (FLOUR, MEAL OR VEGETABLE BASE) [21 CFR] https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?fr=105.66 April 2019
[142] Campbell WW , Kim JE , Amankwaah AF , Gordon SL , Weinheimer-Haus EM. Higher Total Protein Intake and Change in Total Protein Intake Affect Body Composition but Not Metabolic Syndrome Indexes in Middle-Aged Overweight and Obese Adults Who Perform Resistance and Aerobic Exercise for 36 Weeks. J Nutr. 2015 Sep;145(9):2076-83. doi: 10.3945/jn.115.213595. Epub 2015 Aug 5
[143] Madonna . Mamerow,4 Joni A. Mettler,4 Kirk L. English. Dietary Protein Distribution Positively Influences 24-h Muscle Protein Synthesis in Healthy Adults. The Journal of Nutrition. First published ahead of print January 29, 2014 as doi: 10.3945/jn.113.185280
[144] Aljuraiban GS, Chan Q, Oude Griep LM, Brown IJ, Daviglus ML, Stamler J, et al, The Impact of Eating Frequency and Time of Intake on Nutrient Quality and Body Mass Index: The INTERMAP Study, a Population-Based Study. J Acad Nutr Diet. 2015 Apr;115(4):528-536.e1. doi: 10.1016/j.jand.2014.11.017. Epub 2015 Jan 22
[145] Scott Lloyd Robinson, Anneliese Lambeth-Mansell, Gavin Gillibrand, et al, A nutrition and conditioning intervention for natural Bodybuilding contest preparation: case study. Journal of the International Society of Sports Nutrition doi:10.1186/s12970-015-0083-x

[146] Phillips SM, Van Loon LJC. Dietary protein for athletes: from requirements to optimum adaptation. J Sports Sci. 2011;29:29–38.
[147] Coleman, Christopher D et al. “Effectiveness of a Medifast meal replacement program on weight, body composition and cardiometabolic risk factors in overweight and obese adults: a multicenter systematic retrospective chart review study.” *Nutrition journal* vol. 14 77. 6 Aug. 2015, doi:10.1186/s12937-015-0062-8
[148] Raynor HA, Anderson AM, Miller GD, Reeves R, Delahanty LM, Vitolins MZ, Harper P, Mobley C, Konersman K, Mayer-Davis E. Partial Meal Replacement Plan and Quality of the Diet at 1 Year: Action for Health in Diabetes (Look AHEAD) Trial. J Acad Nutr Diet. 2015 May;115(5):731-42. doi: 10.1016/j.jand.2014.11.003. Epub 2015 Jan 6.
[149] Murphy CH, Churchward-Venne TA, Mitchell CJ, Kolar NM, Kassis A, Karagounis LG , Burke LM, Hawley JA, Phillips SM. Hypoenergetic diet-induced reductions in myofibrillar protein synthesis are restored with resistance training and balanced daily protein ingestion in older men. Am J Physiol Endocrinol Metab. 2015 Mar 3:ajpendo.00550.2014. doi: 10.1152/ajpendo.00550.2014
[150]Adult weight management evidence-based nutrition practice guideline. American Dietetic Association Evidence Analysis Library Web site. http://www.adaevidencelibrary.com/topic.cfm?cat_2798. Accessed January 2, 2008
[151] Position of the American Dietetic Association: Weight Management. February 2009 Volume 109 Number 2, Journal of the AMERICAN DIETETIC ASSOCIATION
[152] Kamei, Yasutomi et al. “Regulation of Skeletal Muscle Function by Amino Acids.” *Nutrients* vol. 12,1 261. 19 Jan. 2020, doi:10.3390/nu12010261
[153] Biolo, G.; Tipton, K.D.; Klein, S.; Wolfe, R.R. An abundant supply of amino acids enhances the metabolic effect of exercise on muscle protein. Am. J. Physiol. 1997, 273, E122–E129
[154] Areta, J.L.; Burke, L.M.; Ross, M.L.; Camera, D.M.; West, D.W.; Broad, E.M.; Jeacocke, N.A.; Moore, D.R.; Stellingwerff, T.; Phillips, S.M.; et al. Timing and distribution of protein ingestion during prolonged recovery from resistance exercise alters myofibrillar protein synthesis. J. Physiol. 2013, 591, 2319–2331
[155] Witard OC, Garthe I, Phillips SM. Dietary Protein for Training Adaptation and Body Composition Manipulation in Track and Field Athletes. Int J Sport Nutr Exerc Metab. 2019 Mar 1;29(2):165-174. doi: 10.1123/ijsnem.2018-0267. Epub 2019 Feb 25. PMID: 30507259.
[156] Ashida Y, Himori K, Tatebayashi D, Yamada R, Ogasawara R, Yamada T. Effects of contraction mode and stimulation frequency on electrical stimulation-induced skeletal muscle hypertrophy. J Appl Physiol (1985). 2018 Feb 1;124(2):341-348. doi: 10.1152/japplphysiol.00708.2017. Epub 2017 Oct 26. PMID: 29074713.
[157] Hansen M, Oxfeldt M, Larsen AE, Thomsen LS, Rokkedal-Lausch T, Christensen B, Rittig N, De Paoli FV, Bangsbo J, Ørtenblad N, Madsen K. Supplement with whey protein hydrolysate in contrast to carbohydrate supports mitochondrial adaptations in trained runners. J Int Soc Sports Nutr. 2020 Sep 7;17(1):46. doi: 10.1186/s12970-020-00376-3. PMID: 32894140; PMCID: PMC7487963.
[158] Biolo G, Maggi SP, Williams BD, Tipton KD, Wolfe RR. Increased rates of muscle protein turnover and amino acid transport after resistance exercise in humans. Am J Physiol Endocrinol Metab 268: E514–E520, 1995
[159] Vinod Kumar, Philip Atherton, Kenneth Smith, Michael J. Rennie. Human muscle protein synthesis and breakdown during and after exercise. Journal of Applied Physiology Published 1 June 2009 Vol. 106 no. 6, 2026-2039 DOI:10.1152/japplphysiol.91481.2008
[160] Drummond, M.J.; Dreyer, H.C.; Fry, C.S.; Glynn, E.L.; Rasmussen, B.B. Nutritional and contractile regulation of human skeletal muscle protein synthesis and mTORC1 signaling. *J. Appl. Physiol.* 2009, *106*, 1374–1384
[161] Ogasawara R, Kobayashi K, Tsutaki A, Lee K, Abe T, Fujita S, Nakazato K, Ishii N. mTOR signaling response to resistance exercise is altered by chronic resistance training and detraining in skeletal muscle. J Appl Physiol (1985) 114: 934–940, 2013. doi:10.1152/japplphysiol.01161.2012.
[162] Terzis G, Georgiadis G, Stratakos G, Vogiatzis I, Kavouras S, Manta P, Mascher H, Blomstrand E. Resistance exercise-induced increase in muscle mass correlates with p70S6 kinase phosphorylation in human subjects. Eur J Appl Physiol 102: 145–152, 2008. doi:10.1007/s00421-007-0564-y.
[163] Townsend, Jeremy R et al. “The Effect of ProHydrolase® on the Amino Acid and Intramuscular Anabolic Signaling Response to Resistance Exercise in Trained Males.” *Sports (Basel, Switzerland)* vol. 8,2 13. 22 Jan. 2020, doi:10.3390/sports8020013
[164] Kendall J. Condon et al. Nutrient regulation of mTORC1 at a glance 2019. Published by The Company of Biologists Ltd. Journal of Cell Science (2019) 132, jcs222570. doi:10.1242/jcs.222570

[165] Drummond, M. et al. "Regulation of Protein Metabolism in Exercise and Recovery Nutritional and contractile regulation of human skeletal muscle protein synthesis and mTORC 1 signaling." (2009).
[166] Byfield, M.P.; Murray, J.T.; Backer, J.M. hVps34 is a nutrient-regulated lipid kinase required for activation of p70 S6 kinase. *J. Biol. Chem.* 2005, *280*, 33076–33082
[167] Nobukuni, T.; Joaquin, M.; Roccio, M.; Dann, S.G.; Kim, S.Y.; Gulati, P.; Byfield, M.P.; Backer, J.M.; Natt, F.; Bos, J.L.; *et al.* Amino acids mediate mTOR/raptor signaling through activation of class 3 phosphatidylinositol 3OH-kinase. *Proc. Natl. Acad. Sci. USA* 2005, *102*, 14238–14243
[168] MacKenzie, M.G.; Hamilton, D.L.; Murray, J.T.; Taylor, P.M.; Baar, K. mVps34 is activated following high-resistance contractions. *J. Physiol.* 2009, *587*, 253–260.
[169] Gonzalez, A.M.; Ho_man, J.R.; Townsend, J.R.; Jajtner, A.R.; Boone, C.H.; Beyer, K.S.; Baker, K.M.;Wells, A.J.; Mangine, G.T.; Robinson, E.H.T.; et al. Intramuscular MAPK signaling following high volume and high intensity resistance exercise protocols in trained men. Eur. J. Appl. Physiol. 2016, 116, 1663–1670.
[170] Kudrna, R.A.; Fry, A.C.; Nicoll, J.X.; Gallagher, P.M.; Prewitt, M.R. Effect of Three Different Maximal Concentric Velocity Squat Protocols on MAPK Phosphorylation and Endocrine Responses. J. Strength Cond. Res. 2019, 33, 1692–1702
[171] Boppart, M.D.; Aronson, D.; Gibson, L.; Roubeno_, R.; Abad, L.W.; Bean, J.; Goodyear, L.J.; Fielding, R.A. Eccentric exercise markedly increases c-Jun NH2-terminal kinase activity in human skeletal muscle. J. Appl. Physiol. 1999, 87, 1668–1673
[172] Drummond, M.J.; Fry, C.S.; Glynn, E.L.; Timmerman, K.L.; Dickinson, J.M.; Walker, D.K.; Gundermann, D.M.; Volpi, E.; Rasmussen, B.B. Skeletal muscle amino acid transporter expression is increased in young and older adults following resistance exercise. *J. Appl. Physiol.* 2011, *111*, 135–142
[173] Drummond, M.J.; Glynn, E.L.; Fry, C.S.; Timmerman, K.L.; Volpi, E.; Rasmussen, B.B. An increase in essential amino acid availability upregulates amino acid transporter expression in human skeletal muscle. *Am. J. Physiol. Endocrinol. Metab.* 2010, *298*, E1011–E1018
[174] Churchward-Venne, T.A.; Burd, N.A.; Mitchell, C.J.; West, D.W.; Philp, A.; Marcotte, G.R.; Baker, S.K.; Baar, K.; Phillips, S.M. Supplementation of a suboptimal protein dose with leucine or essential amino acids: Effects on myofibrillar protein synthesis at rest and following resistance exercise in men. *J. Physiol.* 2012, *590*, 2751–2765
[175] Gowans G. J., , Hardie D. G. (2014) AMPK: a cellular energy sensor primarily regulated by AMP. Biochem. Soc. Trans. 42, 71–75 10.1042/BST20130244 [PMCID: PMC5703408] [
[176] Kjøbsted, Rasmus et al. "AMPK in skeletal muscle function and metabolism." *FASEB journal : official publication of the Federation of American Societies for Experimental Biology* vol. 32,4 (2018): 1741-1777. doi:10.1096/fj.201700442R
[177] Dibble C. C., Manning B. D. (2013) Signal integration by mTORC1 coordinates nutrient input with biosynthetic output. Nat. Cell Biol. 15, 555–564 10.1038/ncb2763 [PMCID: PMC3743096] [PubMed: 23728461
[178] Inoki K., Zhu T., , Guan K.-L. (2003) TSC2 mediates cellular energy response to control cell growth and survival. Cell 115, 577–590 10.1016/S0092-8674(03)00929-2 [PubMed: 14651849]
[179] Tipton, K.D.; Gurkin, B.E.; Matin, S.; Wolfe, R.R. Nonessential amino acids are not necessary to stimulate net muscle protein synthesis in healthy volunteers. *J. Nutr. Biochem.* 1999, *10*, 89–95
[180] Volpi E, Kobayashi H, Sheffield-Moore M, et al. Essential amino acids are primarily responsible for the amino acid stimulation of muscle protein anabolism in healthy elderly adults. Am J Clin Nutr. 2003;78:250–8.
[181] Nobukuni, T.; Joaquin, M.; Roccio, M.; Dann, S.G.; Kim, S.Y.; Gulati, P.; Byfield, M.P.; Backer, J.M.; Natt, F.; Bos, J.L.; *et al.* Amino acids mediate mTOR/raptor signaling through activation of class 3 phosphatidylinositol 3OH-kinase. *Proc. Natl. Acad. Sci. USA* 2005, *102*, 14238–14243
[182] Tipton KD, Ferrando AA, Phillips SM, Doyle D Jr, Wolfe RR. Postexercise net protein synthesis in human muscle from orally administered amino acids. Am J Physiol. 1999 Apr;276(4 Pt 1):E628-34.
[183] Rennie MJ, Bohé J, Smith K, Wackerhage H, Greenhaff P. Branched-chain amino acids as fuels and anabolic signals in human muscle. J Nutr. 2006 Jan;136(1 Suppl):264S-8S. Review.
[184] Yoshizawa F. Regulation of protein synthesis by branched-chain amino acids in vivo. Biochem Biophys Res Commun. 2004 Jan 9;313(2):417-22. Review.
[185] Tipton KD, Elliott TA, Cree MG, Aarsland AA, Sanford AP, Wolfe RR. Stimulation of net muscle protein synthesis by whey protein ingestion before and after exercise. Am J Physiol Endocrinol Metab. 2007 Jan;292(1):E71-6. Epub 2006 Aug 8.

[186] Rennie MJ, Edwards RH, Halliday D, Matthews DE, Wolman SL, Millward DJ. Muscle protein synthesis measured by stable isotope techniques in man: the effects of feeding and fasting. Clin Sci (Lond) 63: 519–523, 1982
[187] Phillips, S.M.; Glover, E.I.; Rennie, M.J. Alterations of protein turnover underlying disuse atrophy in human skeletal muscle. *J. Appl. Physiol*. 2009, *107*, 645–654.
[188] Phillips SM. The science of muscle hypertrophy: making dietary protein count. Proc Nutr Soc. 2011 Feb;70(1):100-3. Epub 2010 Nov 22.
[189] Hudson JL, Kim JE, Paddon-Jones D, Campbell WW. Within-day protein distribution does not influence body composition responses during weight loss in resistance-training adults who are overweight. Am J Clin Nutr. 2017 Nov;106(5):1190-1196. doi: 10.3945/ajcn.117.158246. Epub 2017 Sep 13. PMID: 28903957; PMCID: PMC5657287.
[190] Witard, O.C., Wardle, S.L., Macnaughton, L.S., Hodgson, A.B., & Tipton, K.D. (2016). Protein considerations for optimizing skeletal muscle mass in healthy young and older adults. Nutrients, 8(4), 181. PubMed ID: 27023595 doi:10.3390/nu8040181
[191] Kerksick, C.M., Arent, S., Schoenfeld, B.J. *et al.* International society of sports nutrition position stand: nutrient timing. *J Int Soc Sports Nutr* 14, 33 (2017). https://doi.org/10.1186/s12970-017-0189-4
[192] Gieske BT, Stecker RA, Smith CR, Witherbee KE, Harty PS, Wildman R, Kerksick CM. Metabolic impact of protein feeding prior to moderate-intensity treadmill exercise in a fasted state: a pilot study. J Int Soc Sports Nutr. 2018 Nov 29;15(1):56. doi: 10.1186/s12970-018-0263-6. PMID: 30497484; PMCID: PMC6267781.
[193] Arent SM, Cintineo HP, McFadden BA, Chandler AJ, Arent MA. Nutrient Timing: A Garage Door of Opportunity? Nutrients. 2020 Jun 30;12(7):1948. doi: 10.3390/nu12071948. PMID: 32629950; PMCID: PMC7400240.
[194] Churchward-Venne, T.A.; Burd, N.A.; Phillips, S.M. Nutritional regulation of muscle protein synthesis with resistance exercise: Strategies to enhance anabolism. Nutr. Metab. 2012, 9, 40.
[195] John L. Ivy, PhD, Lisa M. Ferguson-Stegall, PhD. Nutrient Timing. The Means to Improved Exercise Performance, Recovery, and Training Adaptation, American Journal of Lifestyle Medicine *July/August 2014* 8: *246*-259, first published on *October 7, 2013.*
[196] Naderi, Alireza et al. "Timing, Optimal Dose and Intake Duration of Dietary Supplements with Evidence-Based Use in Sports Nutrition." *Journal of exercise nutrition & biochemistry* vol. 20,4 (2016): 1-12. doi:10.20463/jenb.2016.0031
[197] Phillips SM, Tipton KD, Aarsland A, Wolf SE, Wolfe RR. Mixed muscle protein synthesis and breakdown after resistance exercise in humans. Am J Physiol. 1997 Jul;273(1 Pt 1):E99-107.
[198] Wolfe, R.R.; Miller, S.L. Amino acid availability controls muscle protein metabolism. *Diabetes Nutr. Metab.* 1999, *12*, 322–328
[199] Stark M, Lukaszuk J, Prawitz A, Salacinski A. Protein timing and its effects on muscular hypertrophy and strength in individuals engaged in weight-training. J Int Soc Sports Nutr. 2012 Dec 14;9(1):54. doi: 10.1186/1550-2783-9-54
[200] Cribb P, Hayes A. Effects of supplement timing and resistance exercise on skeletal muscle hypertrophy. Med Sci Sports Exerc. 2006;38(11):1918–1925. doi:10.1249/01.mss.0000233790.08788.3e.
[201] Guimarães-Ferreira L, Cholewa JM, Naimo MA, Zhi XI, Magagnin D, de Sá RB, Streck EL, Teixeira Tda S, Zanchi NE. Synergistic effects of resistance training and protein intake: practical aspects. Nutrition. 2014 Oct;30(10):1097-103. doi: 10.1016/j.nut.2013.12.017. Epub 2014 Jan 10
[202] Damas, Felipe et al. "Resistance training-induced changes in integrated myofibrillar protein synthesis are related to hypertrophy only after attenuation of muscle damage." *The Journal of physiology* vol. 594,18 (2016): 5209-22. doi:10.1113/JP272472
[203] Kraemer, William J et al. "Growth Hormone(s), Testosterone, Insulin-Like Growth Factors, and Cortisol: Roles and Integration for Cellular Development and Growth With Exercise." *Frontiers in endocrinology* vol. 11 33. 25 Feb. 2020, doi:10.3389/fendo.2020.00033
[204] MacKrell JG, Yaden BC, Bullock H, Chen K, Shetler P, Bryant HU, et al. Molecular targets of androgen signaling that characterize skeletal muscle recovery and regeneration. Nucl Recept Signal. (2015) 13:e005. doi: 10.1621/nrs.13005
[205] Abdulla, H.; Smith, K.; Atherton, P.J.; Idris, I. Role of insulin in the regulation of human skeletal muscle protein synthesis and breakdown: A systematic review and meta-analysis. Diabetologia 2016, 59, 44–55.
[206] Pasiakos, S.M.; McClung, H.L.; McClung, J.P.; Margolis, L.M.; Andersen, N.E.; Cloutier, G.J.; Pikosky, M.A.; Rood, J.C.; Fielding, R.A.; Young, A.J. Leucine-enriched essential amino acid supplementation during moderate steady state exercise enhances postexercise muscle protein synthesis. *Am. J. Clin. Nutr.* 2011, *94*, 809–818.
[207] Haeusler RA., McGraw TE. & Accili D. Biochemical and cellular properties of insulin receptor signaling. Nat. Rev. Mol. Cell Biol 19, 31–44 (2018).

[208] Park, Yeram et al. "Effects of whey protein supplementation prior to, and following, resistance exercise on body composition and training responses: A randomized double-blind placebo-controlled study." *Journal of exercise nutrition & biochemistry* vol. 23,2 (2019): 34-44. doi:10.20463/jenb.2019.0015
[209] Tipton KD, Rasmussen BB, Miller SL, Wolf SE, Owens-Stovall SK, Petrini BE, Wolfe RR. Timing of amino acid-carbohydrate ingestion alters anabolic response of muscle to resistance exercise. *Am J Physiol Endocrinol Metab.* 2001;281:E197-206
[210] Kim, I.Y.; Deutz, N.E.P.; Wolfe, R.R. Update on maximal anabolic response to dietary protein. Clin. Nutr. 2017
[211] Atherton, P.J.; Etheridge, T.; Watt, P.W.; Wilkinson, D.; Selby, A.; Rankin, D.; Smith, K.; Rennie, M.J. Muscle full effect after oral protein: Time-dependent concordance and discordance between human muscle protein synthesis and mTORC1 signaling. *Am. J. Clin. Nutr.* 2010, *92*, 1080–1088
[212] Dreyer HC et al (2008) Leucine-enriched essential amino acid and carbohydrate ingestion following resistance exercise enhances mTOR signaling and protein synthesis in human muscle. Am J Physiol Endocrinol Metab 294:E392–E400
[213] Bohe, J.; Low, J.F.; Wolfe, R.R.; Rennie, M.J. Latency and duration of stimulation of human muscle protein synthesis during continuous infusion of amino acids. *J. Physiol.* 2001, *532*, 575–579
[214] Burke, L.M.; van Loon, L.J.C.; Hawley, J.A. Postexercise muscle glycogen resynthesis in humans. J. Appl. Physiol. 2017, 122, 1055–1067
[215] Cermak N, Res P, de Groot L, Saris H, van Loon L. 2012. Protein supplementation augments the adaptive response of skeletal muscle to resistance-type exercise training: a meta-analysis. Am J Clin Nutr 96:1454–64.
[216] West, Daniel W D et al. "Whey Protein Supplementation Enhances Whole Body Protein Metabolism and Performance Recovery after Resistance Exercise: A Double-Blind Crossover Study." *Nutrients* vol. 9,7 735. 11 Jul. 2017, doi:10.3390/nu9070735
[217] Daniel WD West, Nicholas A Burd, Vernon G Coffey, Steven K Baker, Louise M Burke, John A Hawley, Daniel R Moore, Trent Stellingwerff, and Stuart M Phillips. Rapid aminoacidemia enhances myofibrillar protein synthesis and anabolic intramuscular signaling responses after resistance exercise. Am J Clin Nutr 2011;94:795–803. Printed in USA. _ 2011 American Society for Nutrition
[218] Miller SL, Tipton KD, Chinkes DL, Wolf SE, Wolfe RR. Independent and combined effects of amino acids and glucose after resistance exercise. Med Sci Sports Exerc. 2003 Mar;35(3):449-55.
[219] Tipton KD, Rasmussen BB, Miller SL, Wolf SE, Owens-Stovall SK, Petrini BE, Wolfe RR. Timing of amino acid-carbohydrate ingestion alters anabolic response of muscle to resistance exercise. Am J Physiol Endocrinol Metab. 2001 Aug;281(2):E197- 206. 70-[220] Tipton KD, Elliott TA, Cree MG, Aarsland AA, Sanford AP, Wolfe RR. Stimulation of net muscle protein synthesis by whey protein ingestion before and after exercise. Am J Physiol Endocrinol Metab. 2007 Jan;292(1):E71-6. Epub 2006 Aug 8.
[221] Tipton KD, Elliott TA, Cree MG, Wolf SE, Sanford AP, Wolfe RR. Ingestion of casein and whey proteins result in muscle anabolism after resistance exercise. Med Sci Sports Exerc. 2004 Dec;36(12):2073-81.
[222] Cooke MB, Rybalka E, Stathis CG, Cribb PJ, Hayes A. Whey protein isolate attenuates strength decline after eccentrically induced muscle damage in healthy individuals. J Int Soc Sports Nutr. 2010 Sep 22;7:30.
[223] Moore DR, Atherton PJ, Rennie MJ, Tarnopolsky MA, Phillips SM. Resistance exercise enhances mTOR and MAPK signaling in human muscle over that seen at rest after bolus protein ingestion. Acta Physiol (Oxf). 2011 Mar;201(3):365-72. doi: 10.1111/j.1748-1716.2010.02187.x. Epub 2010 Nov 9.
[224] Bos C, Metges CC, Gaudichon C, Petzke KJ, Pueyo ME, Morens C, Everwand J, Benamouzig R, Tomé D. Postprandial kinetics of dietary amino acids are the main determinant of their metabolism after soy or milk protein ingestion in humans. J Nutr. 2003 May;133(5):1308-15.
[225] Dangin M, Boirie Y, Garcia-Rodenas C, Gachon P, Fauquant J, Callier P, Ballèvre O, Beaufrère B. The digestion rate of protein is an independent regulating factor of postprandial protein retention. Am J Physiol Endocrinol Metab. 2001 Feb;280(2):E340-8.
[226] Phillips SM, Tang JE, Moore DR. The role of milk- and soy-based protein in support of muscle protein synthesis and muscle protein accretion in young and elderly persons. J Am Coll Nutr. 2009 Aug;28(4):343-54. Review.
[227] Snijders T , Res PT , Smeets JS , van Vliet S , van Kranenburg J , Maase K , Kies AK , Verdijk LB , van Loon LJ Protein Ingestion before Sleep Increases Muscle Mass and Strength Gains during Prolonged Resistance-Type Exercise Training in Healthy Young Men. J Nutr. 2015 Apr 29. pii: jn208371. [Epub ahead of print]
[228] Snijders T, Trommelen J, Kouw IWK, Holwerda AM, Verdijk LB, van Loon LJC. The Impact of Pre-sleep Protein Ingestion on the Skeletal Muscle Adaptive Response to Exercise in Humans: An Update. Front Nutr. 2019 Mar 6;6:17. doi: 10.3389/fnut.2019.00017. PMID: 30895177; PMCID: PMC6415027.

[229] FAO/WHO Expert Consultation. 1991. Protein quality evaluation report of the Joint FAO/WHO Expert Consultation held in Bethseda, Md., USA, in 1989. FAO Food and Nutrition Paper 51, Rome.
[230] Pennings B, Boirie Y, Senden J, Gijsen A, Kuipers H, van Loon L. 2011. Whey protein stimulates postprandial muscle protein accretion more effectively than do casein and casein hydrolysate in older men. Am J Clin Nutr 93(5):997–1005.
[231] Burd N, Yang Y, Moore DR, Tang JE, Tarnopolsky MA, Phillips S. 2012. Greater stimulation of myofibrillar protein synthesis with ingestion of whey protein isolate v. micellar casein at rest and after resistance exercise in elderly men. Br J Nutr 108:958–62.
[232] Pennings B, Boirie Y, Senden J, Gijsen A, Kuipers H, van Loon L. 2011. Whey protein stimulates postprandial muscle protein accretion more effectively than do casein and casein hydrolysate in older men. Am J Clin Nutr 93(5):997–1005.
[233] Burd N, Yang Y, Moore DR, Tang JE, Tarnopolsky MA, Phillips S. 2012. Greater stimulation of myofibrillar protein synthesis with ingestion of whey protein isolate v. micellar casein at rest and after resistance exercise in elderly men. Br J Nutr 108:958–62.
[234] Luiking Y, Deutz N, Jakel M, Soeters P. 2005. Casein and soy protein meals differentially affect whole-body and splanchnic protein metabolism in healthy humans. J Nutr 135:1080–7.
[235] Fouillet H, Juillet B, Gaudichon C, Mariotti F, Tome D, Bos C. 2009. Absorption kinetics are a key factor regulating postprandial protein metabolism in response to qualitative and quantitative variations in protein intake. Am J Physiol Regul Integr Comp Physiol 297(6):R1691–705
[236] Phillips, S.M. The impact of protein quality on the promotion of resistance exercise-induced changes in muscle mass. *Nutr Metab (Lond)* 13, 64 (2016). https://doi.org/10.1186/s12986-016-0124-8
[237] Rutherfurd SM, Moughan PJ. Available versus digestible dietary amino acids. Br J Nutr. 2012;108 Suppl 2:S298–305
[238] Anthony J, Yoshizawa F, Anthony T, Vary T, Jefferson L, Kimball S. 2000. Leucine stimulates translation initiation in skeletal muscle of postabsorptive rats via a rapamycin-sensitive pathway. J Nutr 130:2413–9.
[239] Lynch CJ, Halle B, Fujii H, Vary T, Wallin R, Damuni Z, Hutson S. Potential role of leucine metabolism in the leucine-signaling pathway involving mTOR. Am J Physiol Endocrinol Metab 2003;285:E854–63.
[240] Li F, Yin Y, Kong X, Wu G. Leucine nutrition in animals and humans: mTOR signaling and beyond. Amino Acids 2011;41:1185–93
[241] Kimball SR, Jefferson L. Signaling pathways and molecular mechanisms through which branched-chain amino acids mediate translational control of protein synthesis. J Nutr 2006;136(Suppl):227S–31S
[242] Takegaki, Junya et al. "The Effect of Leucine-Enriched Essential Amino Acid Supplementation on Anabolic and Catabolic Signaling in Human Skeletal Muscle after Acute Resistance Exercise: A Randomized, Double-Blind, Placebo-Controlled, Parallel-Group Comparison Trial." *Nutrients* vol. 12,8 2421. 12 Aug. 2020, doi:10.3390/nu12082421
[243] Hulmi JJ, Lockwood CM, Stout JR. Effect of protein/essential amino acids and resistance training on skeletal muscle hypertrophy: A case for whey protein. Nutr Metab (Lond). 2010 Jun 17;7:51.
[244] Hulmi JJ, Tannerstedt J, Selänne H, Kainulainen H, Kovanen V, Mero AA. Resistance exercise with whey protein ingestion affects mTOR signaling pathway and myostatin in men. J Appl Physiol. 2009 May;106(5):1720-9. Epub 2009 Mar 19.
[245] Moore DR, Tang JE, Burd NA, Rerecich T, Tarnopolsky MA, Phillips SM. Differential stimulation of myofibrillar and sarcoplasmic protein synthesis with protein ingestion at rest and after resistance exercise. J Physiol. 2009 Feb 15;587(Pt 4):897-904. Epub 2009 Jan 5.
[246] West D, Burd N, Coffey V, Baker S, Burke L, Hawley J, Moore DR, Stellingwerff T, Phillips S. 2011. Rapid aminoacidemia enhances myofibrillar protein synthesis and anabolic intramuscular signaling responses after resistance exercise. Am J Clin Nutr 94(3):795–803.
[247] Boirie Y, Dangin M, Gachon P, Vasson M, Maubois J, Beaufrere B. 1997. Slow and fast dietary proteins differently modulate postprandial protein accretion. Proc Nath Acad Sci USA 94:14930–5.
[248] Yang Y, Churchward-Venne T, Burd N, Breen L, Tarnopolsky MA, Phillips S. 2012b. Myofibrillar protein synthesis following ingestion of soy protein isolate at rest and after resistance exercise in elderly men. Nutr Metab (Lond) 9:57.
[249] Fouillet H, Mariotti F, Gaudichon C, Bos C, Tome D. 2002. Peripheral and splanchnic metabolism of dietary nitrogen are differently affected by the protein source in humans as assessed by compartmental modeling. J Nutr 132(1):125–33.
[250] Luc J.C. van Loon Application of Protein or Protein Hydrolysates to Improve Postexercise Recovery. Int J Sport Nutr Exerc Metab. 2007 August: 17(Supp): S104-S117
[251] Gabriella A.M. Ten Have ; Marielle P.K.J. Engelen ; Yvette C. Luiking ; Nicolaas E.P. Deutz. Absorption Kinetics of Amino Acids, Peptides, and Intact Proteins. Int J Sport Nutr Exerc Metab. 2007 August: 17(Supp): S23-S36

[252] Volek JS, Volk BM, Gomez AL, Kunces LJ, Kupchak BR, Freidenreich DJ, et al. Whey protein supplementation during resistance training augments lean body mass. J Am Coll Nutr. 2013;32(2):122–35.
[253] Farup J , Rahbek SK, Knudsen IS, de Paoli F, Mackey AL, Vissing K. Whey protein supplementation accelerates satellite cell proliferation during recovery from eccentric exercise. Amino Acids. 2014 Nov;46(11):2503-16. doi: 10.1007/s00726-014-1810-3. Epub 2014 Jul 26
[254] Farup J , Rahbek SK , Riis S , Vendelbo MH , Paoli Fd , Vissing K. Influence of exercise contraction mode and protein supplementation on human skeletal muscle satellite cell content and muscle fiber growth. J Appl Physiol (1985). 2014 Oct 15;117(8):898-909. doi: 10.1152/japplphysiol.00261.2014. Epub 2014 Aug 7.
[255] Farup J , Rahbek SK, Vendelbo MH, Matzon A, Hindhede J, Bejder A, Ringgard S, Vissing K. Whey protein hydrolysate augments tendon and muscle hypertrophy independent of resistance exercise contraction mode. Scand J Med Sci Sports. 2014 Oct;24(5):788-98. doi: 10.1111/sms.12083. Epub 2013 May 7.
[256] Smiles WJ , Areta JL , Coffey VG , Phillips SM , Moore DR , Stellingwerff T , Burke LM , Hawley JA, Camera DM. Modulation of autophagy signaling with resistance exercise and protein ingestion following short-term energy deficit. Am J Physiol Regul Integr Comp Physiol. 2015 Sep;309(5):R603-12. doi: 10.1152/ajpregu.00413.2014. Epub 2015 Jul 1.
[257] P. T. Reidy, D. K. Walker, J. M. Dickinson, D. M. Gundermann, et al. Soy-dairy protein blend and whey protein ingestion after resistance exercise increases amino acid transport and transporter expression in human skeletal muscle. J Appl Physiol (1985). 2014 Jun 1; 116(11): 1353–1364. Published online 2014 Apr 3. doi: 10.1152/japplphysiol.01093.2013 PMCID: PMC4044402
[258] Cameron J Mitchell, Paul A Della Gatta, Aaron C Petersen, David Cameron-Smith, and James F Markworth. Soy protein ingestion results in less prolonged p70S6 kinase phosphorylation compared to whey protein after resistance exercise in older men. Journal of the International Society of Sports Nutrition (2015) 12:6 DOI 10.1186/s12970-015-0070-2
[259] Oikawa SY, McGlory C, D'Souza LK, Morgan AK, Saddler NI, Baker SK, Parise G, Phillips SM. A randomized controlled trial of the impact of protein supplementation on leg lean mass and integrated muscle protein synthesis during inactivity and energy restriction in older persons. Am J Clin Nutr. 2018 Nov 1;108(5):1060-1068. doi: 10.1093/ajcn/nqy193. PMID: 30289425.
[260] Anisimova, Aleksandra S et al. "Protein synthesis and quality control in aging." *Aging* vol. 10,12 (2018): 4269-4288. doi:10.18632/aging.101721
[261] Burd, N.A.; Mitchell, C.J.; Churchward-Venne, T.A.; Phillips, S.M. Bigger weights may not beget bigger muscles: Evidence from acute muscle protein synthetic responses after resistance exercise. *Appl. Physiol. Nutr. Metab.* 2012, *37*, 551–554
[262] Mitchell, C.J.; Churchward-Venne, T.A.; West, D.D.; Burd, N.A.; Breen, L.; Baker, S.K.; Phillips, S.M. Resistance exercise load does not determine training-mediated hypertrophic gains in young men. *J. Appl. Physiol.* 2012, doi:10.1152/japplphysiol.00307.2012
[263] Burd, N.A.; Holwerda, A.M.; Selby, K.C.; West, D.W.; Staples, A.W.; Cain, N.E.; Cashaback, J.G.; Potvin, J.R.; Baker, S.K.; Phillips, S.M. Resistance exercise volume affects myofibrillar protein synthesis and anabolic signaling molecule phosphorylation in young men. *J. Physiol.* 2010, *588*, 3119–3130
[264] Egan B., Zierath J. R. (2013) Exercise metabolism and the molecular regulation of skeletal muscle adaptation. Cell Metab. 17, 162–184 10.1016/j.cmet.2012.12.012
[265]Bassel-Duby R., , Olson E. N. (2006) Signaling pathways in skeletal muscle remodeling. Annu. Rev. Biochem. 75, 19–37 10.1146/annurev.biochem.75.103004.142622
[266]Flück M. (2006) Functional, structural and molecular plasticity of mammalian skeletal muscle in response to exercise stimuli. J. Exp. Biol. 209, 2239–2248 10.1242/jeb.02149 [PubMed: 16731801] [CrossRef:10.1242/jeb.02149]
[267]Stecker et al. Timing of ergogenic aids and micronutrients on muscle and exercise performance. Journal of the International Society of Sports Nutrition (2019) 16:37 https://doi.org/10.1186/s12970-019-0304-9
[268] Mette Hansen, Jens Bangsbo, Jørgen Jensen, Bo Martin Bibby, and Klavs Madsen. Effect of Whey Protein Hydrolysate on Performance and Recovery of Top-Class Orienteering Runners. DOI: http://dx.doi.org/10.1123/ijsnem.2014-0083
[269] Cartee, Gregory D et al. "Exercise Promotes Healthy Aging of Skeletal Muscle." *Cell metabolism* vol. 23,6 (2016): 1034-1047. doi:10.1016/j.cmet.2016.05.007
[270] Ahtiainen, J.P., Pakarinen, A., Alen, M., Kraemer, W.J., & Hakkinen, K. (2003). Muscle hypertrophy, hormonal adaptations and strength development during strength training in strength-trained and untrained men. European Journal of Applied Physiology, 89(6), 555–563. PubMed ID: 12734759 doi:10.1007/s00421-003-0833-3
[271] Flann, K.L., LaStayo, P.C., McClain, D.A., Hazel, M., & Lindstedt, S.L. (2011). Muscle damage and muscle remodeling: No pain, no gain? The Journal of Experimental Biology, 214(Pt. 4), 674–679. PubMed ID: 21270317 doi:10.1242/jeb.050112

[272] Ribeiro, A.S., Tomeleri, C.M., Souza, M.F., Pina, F.L., Schoenfeld, B.J., Nascimento, M.A., : : : Cyrino, E.S. (2015). Effect of resistance training on C-reactive protein, blood glucose and lipid profile in older women with differing levels of RT experience. Age, 37(6), 109. PubMed ID: 26499819 doi:10.1007/s11357-015-9849-y
[273] Desbrow et al. Nutrition for Special Populations: Young, Female, and Masters Athletes. International Journal of Sport Nutrition and Exercise Metabolism, 2019, 29, 220-227. https://doi.org/10.1123/ijsnem.2018-0269
[274] Philp A, Hamilton DL, Baar K. Signals mediating skeletal muscle remodeling by resistance exercise: PI3-kinase independent activation of mTORC1. J Appl Physiol. 2011;110:561–8
[275] Dickinson JM, Gundermann DM, Walker DK, Reidy PT, Borack MS, Drummond MJ, Arora M, Volpi E, Rasmussen BB. Leucine-enriched amino acid ingestion after resistance exercise prolongs myofibrillar protein synthesis and amino acid transporter expression in older men. J Nutr. 2014 Nov;144(11):1694-702. doi: 10.3945/jn.114.198671. Epub 2014 Sep 3
[276] Burke et al. International Association of Athletics Federations Consensus Statement 2019: Nutrition for Athletics. Volume 29 Issue 2. March 2019 DOI: https://doi.org/10.1123/ijsnem.2019-0065
[277] Knapik, Joseph J et al. "Prevalence of Dietary Supplement Use by Athletes: Systematic Review and Meta-Analysis." *Sports medicine (Auckland, N.Z.)* vol. 46,1 (2016): 103-123. doi:10.1007/s40279-015-0387-7
[278] Garthe I, Maughan RJ. Athletes and Supplements: Prevalence and Perspectives. Int J Sport Nutr Exerc Metab.2018 Mar 1;28(2):126-138. doi: 10.1123/ijsnem.2017-0429. Epub 2018 Mar 26.
[279] Cheng S, Degens H, Evans M, Cheng SM, Selänne H, Rittweger J, Heinonen A, Suominen H, Strandberg T, Alen M, Korhonen MT Gerontology What Makes a 97-Year-Old Man Cycle 5,000 km a Year? *2016; 62(5):508-12.*
[280] Korhonen MT, Cristea A, Alén M, Häkkinen K, Sipilä S, Mero A, Viitasalo JT, Larsson L, Suominen H. Aging, muscle fiber type, and contractile function in sprint-trained athletes. *J Appl Physiol (1985). 2006 Sep; 101(3):906-17.*
[281] Wall BT, Gorissen SH, Pennings B, Koopman R, Groen BB, Verdijk LB, van Loon LJ. Aging Is Accompanied by a Blunted Muscle Protein Synthetic Response to Protein Ingestion. *PLoS One. 2015; 10(11):e0140903.*
[282]Rawson et al. Dietary Supplements for Health, Adaptation, and Recovery in Athletes. Int J Sport Nutr Exerc Metab. 2018 Mar 1;28(2):188-199. doi: 10.1123/ijsnem.2017-0340. Epub 2018 Feb 19.
[283] Cuthbertson D, Smith K, Babraj J, Leese G, Waddell T, Atherton P, Wackerhage H, Taylor PM, Rennie MJ. Anabolic signaling deficits underlie amino acid resistance of wasting, aging muscle. FASEB J. 2005 Mar;19(3):422-4. Epub 2004 Dec 13.
[284] Deutz NE , Bauer JM , Barazzoni R , Biolo G , Boirie Y , Bosy-Westphal, et al. Protein intake and exercise for optimal muscle function with aging: Recommendations from the ESPEN Expert Group. Clin Nutr. 2014 Apr 24. pii: S0261-5614(14)00111-3. doi: 10.1016/j.clnu.2014.04.007. [Epub ahead of print]
[285] Deer RR , Volpi E. Protein intake and muscle function in older adults. Curr Opin Clin Nutr Metab Care. 2015 May;18(3):248-53. doi: 10.1097/MCO.0000000000000162.
[286] Phillips BE, Hill DS, Atherton PJ. Regulation of muscle protein synthesis in humans. Curr Opin Clin Nutr Metab Care. 2012.
[287] Hruby, Adela et al. "Protein Intake and Functional Integrity in Aging: The Framingham Heart Study Offspring." *The journals of gerontology. Series A, Biological sciences and medical sciences* vol. 75,1 (2020): 123-130. doi:10.1093/gerona/gly201
[288] Otsuka R, Kato Y, Tange C, Nishita Y, Tomida M, Imai T, Ando F, Shimokata H, Arai H. Protein intake per day and at each daily meal and skeletal muscle mass declines among older community dwellers in Japan. Public Health Nutr. 2020 Apr;23(6):1090-1097. doi: 10.1017/S1368980019002921. Epub 2019 Oct 14. PMID: 31608843.
[289] Foscolou A, Magriplis E, Tyrovolas S, Chrysohoou C, Sidossis L, Matalas AL, Rallidis L, Panagiotakos D. The association of protein and carbohydrate intake with successful aging: a combined analysis of two epidemiological studies. Eur J Nutr. 2019 Mar;58(2):807-817. doi: 10.1007/s00394-018-1693-2. Epub 2018 Apr 23. PMID: 29687264
[290] Richter M, Baerlocher K, Bauer JM, Elmadfa I, Heseker H, Leschik-Bonnet E, Stangl G, Volkert D, Stehle P; on behalf of the German Nutrition Society (DGE). Revised Reference Values for the Intake of Protein. Ann Nutr Metab. 2019;74(3):242-250. doi: 10.1159/000499374. Epub 2019 Mar 22. PMID: 30904906; PMCID: PMC6492513
[291] Phillips SM, Chevalier S, Leidy HJ. Protein "requirements" beyond the RDA: implications for optimizing health. Appl Physiol Nutr Metab. 2016 May;41(5):565-72. doi: 10.1139/apnm-2015-0550. Epub 2016 Feb 9. PMID: 26960445.
[292] Franzke B, Neubauer O, Cameron-Smith D, Wagner KH. Dietary Protein, Muscle and Physical Function in the Very Old. Nutrients. 2018 Jul 20;10(7):935. doi: 10.3390/nu10070935. PMID: 30037048; PMCID: PMC6073115.

[293] Oliveira CLP, Dionne IJ, Prado CM. Are Canadian protein and physical activity guidelines optimal for sarcopenia prevention in older adults? Appl Physiol Nutr Metab. 2018 Dec;43(12):1215-1223. doi: 10.1139/apnm-2018-0141. Epub 2018 Jul 10. PMID: 29990445.
[294] Traylor DA, Gorissen SHM, Phillips SM. Perspective: Protein Requirements and Optimal Intakes in Aging: Are We Ready to Recommend More Than the Recommended Daily Allowance? Adv Nutr. 2018 May 1;9(3):171-182. doi: 10.1093/advances/nmy003. PMID: 29635313; PMCID: PMC5952928.
[295] Bauer J, Morley JE, Schols AMWJ, Ferrucci L, Cruz-Jentoft AJ, Dent E, Baracos VE, Crawford JA, Doehner W, Heymsfield SB, Jatoi A, Kalantar-Zadeh K, Lainscak M, Landi F, Laviano A, Mancuso M, Muscaritoli M, Prado CM, Strasser F, von Haehling S, Coats AJS, Anker SD. Sarcopenia: A Time for Action. An SCWD Position Paper. J Cachexia Sarcopenia Muscle. 2019 Oct;10(5):956-961. doi: 10.1002/jcsm.12483. Epub 2019 Sep 15. PMID: 31523937; PMCID: PMC6818450.
[296] Sattler FR. Growth hormone in the aging male. Best Pract Res Clin Endocrinol Metab. 2013 Aug;27(4):541-55. doi: 10.1016/j.beem.2013.05.003. Epub 2013 Jun 18. PMID: 24054930; PMCID: PMC3940699.
[297] Bartke A (2005) Minireview: role of the growth hormone/insulin like growth factor system in mammalian aging. Endocrinology 146(9):3718–3723
[298] Tan KT, Ang SJ, Tsai SY. Sarcopenia: Tilting the Balance of Protein Homeostasis. Proteomics. 2020 Mar;20(5-6):e1800411. doi: 10.1002/pmic.201800411. Epub 2019 Dec 4. PMID: 31722440.
[299] Sonntag WE, Ramsey M, Carter CS (2005) Growth hormone and insulin-like growth factor-1 (IGF-1) and their influence on cognitive aging. Ageing Res Rev 4(2):195–212
[300] Colon, Gabriela et al. "The enigmatic role of growth hormone in age-related diseases, cognition, and longevity". *GeroScience* vol. 41,6 (2019): 759-774. doi:10.1007/s11357-019-00096-w
[301] Nielsen J, Jensen RB, Afdeling AJ (2015) Growth hormone deficiency in children. Ugeskr Laeger 177(26):1260–1263
[302] Devesa J, Almenglo C, Devesa P (2016a) Multiple effects of growth hormone in the body: is it really the hormone for growth? Clin Med Insights Endocrinol Diabetes 9:47–71
[303] Larsson, Lars et al. "Sarcopenia: Aging-Related Loss of Muscle Mass and Function." *Physiological reviews* vol. 99,1 (2019): 427-511. doi:10.1152/physrev.00061.2017
[304] Sugihara Junior P, Ribeiro AS, Nabuco HCG, Fernandes RR, Tomeleri CM, Cunha PM, Venturini D, Barbosa DS, Schoenfeld BJ, Cyrino ES. Effects of Whey Protein Supplementation Associated With Resistance Training on Muscular Strength, Hypertrophy, and Muscle Quality in Preconditioned Older Women. Int J Sport Nutr Exerc Metab. 2018 Sep 1;28(5):528-535. doi: 10.1123/ijsnem.2017-0253. Epub 2018 Jun 22. PMID: 29252039.
[305] Nilsson MI, Mikhail A, Lan L, Di Carlo A, Hamilton B, Barnard K, Hettinga BP, Hatcher E, Tarnopolsky MG, Nederveen JP, Bujak AL, May L, Tarnopolsky MA. A Five-Ingredient Nutritional Supplement and Home-Based Resistance Exercise Improve Lean Mass and Strength in Free-Living Elderly. Nutrients. 2020 Aug 10;12(8):2391. doi: 10.3390/nu12082391. PMID: 32785021; PMCID: PMC7468764.
[306] McKendry, J.; Currier, B.S.; Lim, C.; Mcleod, J.C.; Thomas, A.C.; Phillips, S.M. Nutritional Supplements to Support Resistance Exercise in Countering the Sarcopenia of Aging. *Nutrients* 2020, *12*, 2057
[307] Robinson, S M et al. "Does nutrition play a role in the prevention and management of sarcopenia?" *Clinical nutrition (Edinburgh, Scotland)* vol. 37,4 (2018): 1121-1132. doi:10.1016/j.clnu.2017.08.016
[308] Liao CD, Tsauo JY, Wu YT, Cheng CP, Chen HC, Huang YC, Chen HC, Liou TH. Effects of protein supplementation combined with resistance exercise on body composition and physical function in older adults: a systematic review and meta-analysis. Am J Clin Nutr. 2017 Oct;106(4):1078-1091. doi: 10.3945/ajcn.116.143594. Epub 2017 Aug 16. PMID: 28814401.
[309] Kobayashi S, Suga H, Sasaki S; Three-generation Study of Women on Diets and Health Study Group. Diet with a combination of high protein and high total antioxidant capacity is strongly associated with low prevalence of frailty among old Japanese women: a multicenter cross-sectional study. Nutr J. 2017 May 12;16(1):29. doi: 10.1186/s12937-017-0250-9. PMID: 28499379; PMCID: PMC5429552.
[310] Bo Y, Liu C, Ji Z, Yang R, An Q, Zhang X, You J, Duan D, Sun Y, Zhu Y, Cui H, Lu Q. A high whey protein, vitamin D and E supplement preserves muscle mass, strength, and quality of life in sarcopenic older adults: A double-blind randomized controlled trial. Clin Nutr. 2019 Feb;38(1):159-164. doi: 10.1016/j.clnu.2017.12.020. Epub 2018 Jan 9. PMID: 29395372.

[311] Boutry-Regard C, Gerard VP, Denis B, Toshio M. Supplementation with Whey Protein, Omega-3 Fatty Acids and Polyphenols Combined with Electrical Muscle Stimulation Increases Muscle Strength in Elderly Adults with Limited Mobility: A Randomized Controlled Trial. Nutrients. 2020 Jun 23;12(6):1866. doi: 10.3390/nu12061866. PMID: 32585837; PMCID: PMC7353259.
[312] Yoshida D, Ohara T, Hata J, Shibata M, Hirakawa Y, Honda T, Uchida K, Takasugi S, Kitazono T, Kiyohara Y, Ninomiya T. Dairy consumption and risk of functional disability in an elderly Japanese population: the Hisayama Study. Am J Clin Nutr. 2019 Jun 1;109(6):1664-1671. doi: 10.1093/ajcn/nqz040. PMID: 31075788.
[313] Li CY, Fang AP, Ma WJ, Wu SL, Li CL, Chen YM, Zhu HL. Amount Rather than Animal vs Plant Protein Intake Is Associated with Skeletal Muscle Mass in Community-Dwelling Middle-Aged and Older Chinese Adults: Results from the Guangzhou Nutrition and Health Study. J Acad Nutr Diet. 2019 Sep;119(9):1501-1510. doi: 10.1016/j.jand.2019.03.010. Epub 2019 May 9. PMID: 31080071.
[314] Park Y, Choi JE, Hwang HS. Protein supplementation improves muscle mass and physical performance in undernourished prefrail and frail elderly subjects: a randomized, double-blind, placebo-controlled trial. Am J Clin Nutr. 2018 Nov 1;108(5):1026-1033. doi: 10.1093/ajcn/nqy214. PMID: 30475969.
[315] Nowson CA, Service C, Appleton J, Grieger JA. The Impact of Dietary Factors on Indices of Chronic Disease in Older People: A Systematic Review. J Nutr Health Aging. 2018;22(2):282-296. doi: 10.1007/s12603-017-0920-5. PMID: 29380857.
[316] Houston DK, Tooze JA, Garcia K, Visser M, Rubin S, Harris TB, Newman AB, Kritchevsky SB; Health ABC Study. Protein Intake and Mobility Limitation in Community-Dwelling Older Adults: the Health ABC Study. J Am Geriatr Soc. 2017 Aug;65(8):1705-1711. doi: 10.1111/jgs.14856. Epub 2017 Mar 17. PMID: 28306154; PMCID: PMC5555791.
[317] Mitchell CJ, Milan AM, Mitchell SM, Zeng N, Ramzan F, Sharma P, Knowles SO, Roy NC, Sjödin A, Wagner KH, Cameron-Smith D. The effects of dietary protein intake on appendicular lean mass and muscle function in elderly men: a 10-wk randomized controlled trial. Am J Clin Nutr. 2017 Dec;106(6):1375-1383. doi: 10.3945/ajcn.117.160325. Epub 2017 Nov 1. PMID: 29092886.
[318] Adela Hruby, Paul F Jacques, Dietary Protein and Changes in Biomarkers of Inflammation and Oxidative Stress in the Framingham Heart Study Offspring Cohort, *Current Developments in Nutrition*, Volume 3, Issue 5, May 2019, nzz019, https://doi.org/10.1093/cdn/nzz019
[319] Nabuco HCG, Tomeleri CM, Fernandes RR, Sugihara Junior P, Venturini D, Barbosa DS, Deminice R, Sardinha LB, Cyrino ES. Effects of pre- or post-exercise whey protein supplementation on oxidative stress and antioxidant enzymes in older women. Scand J Med Sci Sports. 2019 Aug;29(8):1101-1108. doi: 10.1111/sms.13449. Epub 2019 May 27. PMID: 31050066.
[320] Rondanelli M, Klersy C, Terracol G, Talluri J, Maugeri R, Guido D, Faliva MA, Solerte BS, Fioravanti M, Lukaski H, Perna S. Whey protein, amino acids, and vitamin D supplementation with physical activity increases fat-free mass and strength, functionality, and quality of life and decreases inflammation in sarcopenic elderly. Am J Clin Nutr. 2016 Mar;103(3):830-40. doi: 10.3945/ajcn.115.113357. Epub 2016 Feb 10. PMID: 26864356.
[321] S *Breen L, Phillips SM. S*keletal muscle protein metabolism in the elderly: Interventions to counteract the 'anabolic resistance' of aging. *Nutr Metab (Lond). 2011 Oct 5; 8():68.*
[322] Barclay, Richie D et al. "The Role of the IGF-1 Signaling Cascade in Muscle Protein Synthesis and Anabolic Resistance in Aging Skeletal Muscle." *Frontiers in nutrition* vol. 6 146. 10 Sep. 2019, doi:10.3389/fnut.2019.00146
[323] Dawson BM[1], Axford S. Nutrition as a part of healthy aging and reducing cardiovascular risk: improving functionality in later life using quality protein, with optimized timing and distribution. Semin Thromb Hemost. 2014 Sep;40(6):695-703. doi: 10.1055/s-0034-1389081. Epub 2014 Aug 24.
[324] Gilmartin, Sarah et al. "Whey for Sarcopenia; Can Whey Peptides, Hydrolysates or Proteins Play a Beneficial Role?" *Foods (Basel, Switzerland)* vol. 9,6 750. 5 Jun. 2020, doi:10.3390/foods9060750
[325] Björkman MP, Suominen MH, Kautiainen H, Jyväkorpi SK, Finne-Soveri HU, Strandberg TE, Pitkälä KH, Tilvis RS. Effect of Protein Supplementation on Physical Performance in Older People With Sarcopenia-A Randomized Controlled Trial. J Am Med Dir Assoc. 2020 Feb;21(2):226-232.e1. doi: 10.1016/j.jamda.2019.09.006. Epub 2019 Nov 14. PMID: 31734121.
[326] Amasene M, Besga A, Echeverria I, Urquiza M, Ruiz JR, Rodriguez-Larrad A, Aldamiz M, Anaut P, Irazusta J, Labayen I. Effects of Leucine-Enriched Whey Protein Supplementation on Physical Function in Post-Hospitalized Older Adults Participating in 12-Weeks of Resistance Training Program: A Randomized Controlled Trial. Nutrients. 2019 Oct 1;11(10):2337. doi: 10.3390/nu11102337. PMID: 31581591; PMCID: PMC6835698.
[327] Moran C, Scotto di Palumbo A, Bramham J, Moran A, Rooney B, De Vito G, Egan B. Effects of a Six-Month Multi-Ingredient Nutrition Supplement Intervention of Omega-3 Polyunsaturated Fatty Acids, vitamin D, Resveratrol, and Whey Protein on

Cognitive Function in Older Adults: A Randomised, Double-Blind, Controlled Trial. J Prev Alzheimers Dis. 2018;5(3):175-183. doi: 10.14283/jpad.2018.11. PMID: 29972210.
[328] Kirk B, Mooney K, Amirabdollahian F, Khaiyat O. Exercise and Dietary-Protein as a Countermeasure to Skeletal Muscle Weakness: Liverpool Hope University - Sarcopenia Aging Trial (LHU-SAT). Front Physiol. 2019 Apr 25;10:445. doi: 10.3389/fphys.2019.00445. PMID: 31133863; PMCID: PMC6524700.
[329] Verreijen AM, Engberink MF, Houston DK, Brouwer IA, Cawthon PM, Newman AB, Tylavsky FA, Harris TB, Weijs PJM, Visser M. Dietary protein intake is not associated with 5-y change in mid-thigh muscle cross-sectional area by computed tomography in older adults: the Health, Aging, and Body Composition (Health ABC) Study. Am J Clin Nutr. 2019 Mar 1;109(3):535-543. doi: 10.1093/ajcn/nqy341. PMID: 30850837; PMCID: PMC6408207.
[330] Bhasin S, Apovian CM, Travison TG, Pencina K, Moore LL, Huang G, Campbell WW, Li Z, Howland AS, Chen R, Knapp PE, Singer MR, Shah M, Secinaro K, Eder RV, Hally K, Schram H, Bearup R, Beleva YM, McCarthy AC, Woodbury E, McKinnon J, Fleck G, Storer TW, Basaria S. Effect of Protein Intake on Lean Body Mass in Functionally Limited Older Men: A Randomized Clinical Trial. JAMA Intern Med. 2018 Apr 1;178(4):530-541. doi: 10.1001/jamainternmed.2018.0008. PMID: 29532075; PMCID: PMC5885156.
[331] Stijn Soenen, Caroline Giezenaar, Amy T Hutchison, Michael Horowitz, Ian Chapman, Natalie D Luscombe-Marsh, Effects of intraduodenal protein on appetite, energy intake, and antropyloroduodenal motility in healthy older compared with young men in a randomized trial, *The American Journal of Clinical Nutrition*, Volume 100, Issue 4, October 2014, Pages 1108–1115, https://doi.org/10.3945/ajcn.114.087981
[332] Yeung SE, Hilkewich L, Gillis C, Heine JA, Fenton TR. Protein intakes are associated with reduced length of stay: a comparison between Enhanced Recovery After Surgery (ERAS) and conventional care after elective colorectal surgery. Am J Clin Nutr. 2017 Jul;106(1):44-51. doi: 10.3945/ajcn.116.148619. Epub 2017 May 3. PMID: 28468890.
[333] Lancha AH Jr, Zanella R Jr, Tanabe SG, Andriamihaja M, Blachier F. Dietary protein supplementation in the elderly for limiting muscle mass loss. Amino Acids. 2017 Jan;49(1):33-47. doi: 10.1007/s00726-016-2355-4. Epub 2016 Nov 2. PMID: 27807658.
[334] Nabuco HCG, Tomeleri CM, Fernandes RR, Sugihara Junior P, Cavalcante EF, Cunha PM, Antunes M, Nunes JP, Venturini D, Barbosa DS, Burini RC, Silva AM, Sardinha LB, Cyrino ES. Effect of whey protein supplementation combined with resistance training on body composition, muscular strength, functional capacity, and plasma-metabolism biomarkers in older women with sarcopenic obesity: A randomized, double-blind, placebo-controlled trial. Clin Nutr ESPEN. 2019 Aug;32:88-95. doi: 10.1016/j.clnesp.2019.04.007. Epub 2019 May 13. PMID: 31221297.
[335] Wilkinson DJ, Bukhari SSI, Phillips BE, Limb MC, Cegielski J, Brook MS, Rankin D, Mitchell WK, Kobayashi H, Williams JP, Lund J, Greenhaff PL, Smith K, Atherton PJ. Effects of leucine-enriched essential amino acid and whey protein bolus dosing upon skeletal muscle protein synthesis at rest and after exercise in older women. Clin Nutr. 2018 Dec;37(6 Pt A):2011-2021. doi: 10.1016/j.clnu.2017.09.008. Epub 2017 Sep 23. PMID: 29031484; PMCID: PMC6295981.
[336] Kang L, Gao Y, Liu X, Liang Y, Chen Y, Liang Y, Zhang L, Chen W, Pang H, Peng LN. Effects of whey protein nutritional supplement on muscle function among community-dwelling frail older people: A multicenter study in China. Arch Gerontol Geriatr. 2019 Jul-Aug;83:7-12. doi: 10.1016/j.archger.2019.03.012. Epub 2019 Mar 17. PMID: 30921603.
[337] Lin CC, Shih MH, Chen CD, Yeh SL. Effects of adequate dietary protein with whey protein, leucine, and vitamin D supplementation on sarcopenia in older adults: An open-label, parallel-group study. Clin Nutr. 2020 Aug 27:S0261-5614(20)30432-5. doi: 10.1016/j.clnu.2020.08.017. Epub ahead of print. PMID: 32928579.
[338] Bauer JM , Verlaan S , Bautmans I , Brandt K, et al. Effects of a Vitamin D and Leucine-Enriched Whey Protein Nutritional Supplement on Measures of Sarcopenia in Older Adults, the PROVIDE Study: A Randomized, Double-Blind, Placebo-Controlled Trial. J Am Med Dir Assoc. 2015 Sep 1;16(9):740-7. doi: 10.1016/j.jamda.2015.05.021. Epub 2015 Jul 10.
[339] Hashemilar M, Khalili M, Rezaeimanesh N, Sadeghi Hokmabadi E, Rasulzade S, Shamshirgaran SM, Taheraghdam A, Farhoudi M, Shaafi S, Shakouri SK, Savadi Osgouei D. Effect of Whey Protein Supplementation on Inflammatory and Antioxidant Markers, and Clinical Prognosis in Acute Ischemic Stroke (TNS Trial): A Randomized, Double Blind, Controlled, Clinical Trial. Adv Pharm Bull. 2020 Jan;10(1):135-140. doi: 10.15171/apb.2020.018. Epub 2019 Dec 11. PMID: 32002373; PMCID: PMC6983999.
[340] Niitsu M , Ichinose D , Hirooka T , Mitsutomi K , Morimoto Y , Sarukawa J , Nishikino S , Yamauchi K , Yamazaki. Effects of combination of whey protein intake and rehabilitation on muscle strength and daily movements in patients with hip fracture in the early postoperative period. Clin Nutr. 2015 Jul 17. pii: S0261-5614(15)00181-8. doi: 10.1016/j.clnu.2015.07.006. [Epub ahead of print]

[341] Wallace TC, Frankenfeld CL. Dietary Protein Intake above the Current RDA and Bone Health: A Systematic Review and Meta-Analysis. J Am Coll Nutr. 2017 Aug;36(6):481-496. doi: 10.1080/07315724.2017.1322924. Epub 2017 Jul 7. PMID: 28686536.
[342] Kerstetter JE , Bihuniak JD , Brindisi J , Sullivan RR , Mangano KM , Larocque S , Kotler BM , Simpson CA , Cusano AM , Gaffney-Stomberg E, et al. The Effect of a Whey Protein Supplement on Bone Mass in Older Caucasian Adults. J Clin Endocrinol Metab. 2015 Jun;100(6):2214-22. doi: 10.1210/jc.2014-3792. Epub 2015 Apr 6.
[343] Groenendijk I, den Boeft L, van Loon LJC, de Groot LCPGM. High Versus low Dietary Protein Intake and Bone Health in Older Adults: a Systematic Review and Meta-Analysis. Comput Struct Biotechnol J. 2019 Jul 22;17:1101-1112. doi: 10.1016/j.csbj.2019.07.005. PMID: 31462966; PMCID: PMC6704341
[344] Hill TR, Verlaan S, Biesheuvel E, Eastell R, Bauer JM, Bautmans I, Brandt K, Donini LM, Maggio M, Mets T, Seal CJ, Wijers SL, Sieber C, Cederholm T, Aspray TJ; PROVIDE Consortium. A Vitamin D, Calcium and Leucine-Enriched Whey Protein Nutritional Supplement Improves Measures of Bone Health in Sarcopenic Non-Malnourished Older Adults: The PROVIDE Study. Calcif Tissue Int. 2019 Oct;105(4):383-391. doi: 10.1007/s00223-019-00581-6. Epub 2019 Jul 23. PMID: 31338563
[345] Teixeira FJ, Santos HO, Howell SL, Pimentel GD. Whey protein in cancer therapy: A narrative review. Pharmacol Res. 2019 Jun;144:245-256. doi: 10.1016/j.phrs.2019.04.019. Epub 2019 Apr 18. PMID: 31005617.
[346] Cereda E, Turri A, Klersy C, Cappello S, Ferrari A, Filippi AR, Brugnatelli S, Caraccia M, Chiellino S, Borioli V, Monaco T, Stella GM, Arcaini L, Benazzo M, Grugnetti G, Pedrazzoli P, Caccialanza R. Whey protein isolate supplementation improves body composition, muscle strength, and treatment tolerance in malnourished advanced cancer patients undergoing chemotherapy. Cancer Med. 2019 Nov;8(16):6923-6932. doi: 10.1002/cam4.2517. Epub 2019 Sep 30. PMID: 31568698; PMCID: PMC6853834.
[347] Layman DK, Lönnerdal B, Fernstrom JD. Applications for α-lactalbumin in human nutrition. Nutr Rev. 2018 Jun 1;76(6):444-460. doi: 10.1093/nutrit/nuy004. PMID: 29617841; PMCID: PMC5934683.
[348] Sahathevan S, Se CH, Ng S, Khor BH, Chinna K, Goh BL, Gafor HA, Bavanandan S, Ahmad G, Karupaiah T. Clinical efficacy and feasibility of whey protein isolates supplementation in malnourished peritoneal dialysis patients: A multicenter, parallel, open label randomized controlled trial. Clin Nutr ESPEN. 2018 Jun;25:68-77. doi: 10.1016/j.clnesp.2018.04.002. PMID: 29779821.
[349] Owen PJ, Armbrecht G, Bansmann M, Zange J, Pohle-Fröhlich R, Felsenberg D, Belavý DL. Whey protein supplementation with vibration exercise ameliorates lumbar paraspinal muscle atrophy in prolonged bed rest. J Appl Physiol (1985). 2020 Jun 1;128(6):1568-1578. doi: 10.1152/japplphysiol.00125.2020. Epub 2020 Apr 23. PMID: 32324477.
[350] Alyasin S, Nabavizadeh SH, Esmaeilzadeh H, Heydari ST, Mosavat SH, Parvizi MM, Hashemi SM, Hashempur MH. Efficacy of oral supplementation of whey protein in patients with contact dermatitis: A pilot randomized double-blind placebo-controlled clinical trial. Dermatol Ther. 2020 Sep 2:e14260. doi: 10.1111/dth.14260. Epub ahead of print. PMID: 32876987.
[351] Bohl M , Bjørnshave A , Rasmussen KV , Schioldan AG , Amer B , Larsen MK , Dalsgaard TK , Holst JJ , Herrmann A , O'Neill S, et al. Dairy proteins, dairy lipids, and postprandial lipemia in persons with abdominal obesity (DairyHealth): a 12-wk, randomized, parallel-controlled, double-blinded, diet intervention study. Am J Clin Nutr. 2015 Apr;101(4):870-8. doi: 10.3945/ajcn.114.097923. Epub 2015 Jan 14
[352] Jakubowicz D , Froy O, Ahrén B, Boaz M, Landau Z, Bar-Dayan Y, Ganz T, Barnea M, Wainstein J. Incretin, insulinotropic and glucose-lowering effects of whey protein pre-load in type 2 diabetes: a randomised clinical trial. Diabetologia. 2014 Sep;57(9):1807-11. doi: 10.1007/s00125-014-3305-x. Epub 2014 Jul 10.
[353] Paul J. Arciero, Daniel Baur, Scott Connelly, and Michael J. Ormsbee. Timed-daily ingestion of whey protein and exercise training reduces visceral adipose tissue mass and improves insulin resistance: the PRISE study. *J Appl Physiol* 117: 1–10, 2014. First published May 15, 2014; doi:10.1152/japplphysiol.00152.2014.
[354] Ling-Mei Zhou, Jia-Ying Xu, Chun-Ping Rao, Shufen Han, Zhongxiao Wan and Li-Qiang Qin. Effect of Whey Supplementation on Circulating C-Reactive Protein: A Meta-Analysis of Randomized Controlled Trials. *Nutrients* 2015, *7*, 1131-1143; doi:10.3390/nu7021131
[355] Zhang Z, Xu G, Yang F, Zhu W, Liu X. Quantitative analysis of dietary protein intake and stroke risk. Neurology. 2014 Jul 1;83(1):19-25. doi: 10.1212/WNL.0000000000000551. Epub 2014 Jun 11.
[356] Fekete ÁA, Giromini C, Chatzidiakou Y, Givens DI, Lovegrove JA. Whey protein lowers blood pressure and improves endothelial function and lipid biomarkers in adults with prehypertension and mild hypertension: results from the chronic Whey2Go randomized controlled trial. Am J Clin Nutr. 2016 Dec;104(6):1534-1544. doi: 10.3945/ajcn.116.137919. Epub 2016 Oct 26. PMID: 27797709; PMCID: PMC5118733.

[357] Badely M, Sepandi M, Samadi M, Parastouei K, Taghdir M. The effect of whey protein on the components of metabolic syndrome in overweight and obese individuals; a systematic review and meta-analysis. Diabetes Metab Syndr. 2019 Nov-Dec;13(6):3121-3131. doi: 10.1016/j.dsx.2019.11.001. Epub 2019 Nov 15. PMID: 31790966.
[358] Gjevestad GO, Holven KB, Rundblad A, Flatberg A, Myhrstad M, Karlsen K, Mutt SJ, Herzig KH, Ottestad I, Ulven SM. Increased protein intake affects pro-opiomelanocortin (POMC) processing, immune function and IGF signaling in peripheral blood mononuclear cells of home-dwelling old subjects using a genome-wide gene expression approach. Genes Nutr. 2019 Nov 28;14:32. doi: 10.1186/s12263-019-0654-6. PMID: 31798754; PMCID: PMC6883584.
[359] Gjevestad GO, Hamarsland H, Raastad T, Ottestad I, Christensen JJ, Eckardt K, Drevon CA, Biong AS, Ulven SM, Holven KB. Gene expression is differentially regulated in skeletal muscle and circulating immune cells in response to an acute bout of high-load strength exercise. Genes Nutr. 2017 Mar 3;12:8. doi: 10.1186/s12263-017-0556-4. PMID: 28270867; PMCID: PMC5335818.
[360] Mizubuti YGG, Vieira ELM, Silva TA, d'Alessandro MO, Generoso SV, Teixeira AL, Lima AS, Correia MITD. Comparing the effects of whey and casein supplementation on nutritional status and immune parameters in patients with chronic liver disease: a randomised double-blind controlled trial. Br J Nutr. 2020 Aug 18:1-12. doi: 10.1017/S0007114520003219. Epub ahead of print. PMID: 32807252.
[361] Bumrungpert A, Pavadhgul P, Nunthanawanich P, Sirikanchanarod A, Adulbhan A. Whey Protein Supplementation Improves Nutritional Status, Glutathione Levels, and Immune Function in Cancer Patients: A Randomized, Double-Blind Controlled Trial. J Med Food. 2018 Jun;21(6):612-616. doi: 10.1089/jmf.2017.4080. Epub 2018 Mar 12. PMID: 29565716.
[362] Hellinga, Anneke H et al. "In Vitro Induction of Trained Innate Immunity by bIgG and Whey Protein Extracts." *International journal of molecular sciences* vol. 21,23 9077. 28 Nov. 2020, doi:10.3390/ijms21239077
[363] Buey B, Bellés A, Latorre E, Abad I, Pérez MD, Grasa L, Mesonero JE, Sánchez L. Comparative effect of bovine buttermilk, whey, and lactoferrin on the innate immunity receptors and oxidative status of intestinal epithelial cells. Biochem Cell Biol. 2020 Jun 13. doi: 10.1139/bcb-2020-0121. Epub ahead of print. PMID: 32538128.
[364] Ha E, Zemel MB: Functional properties of whey, whey components, and essential amino acids: mechanisms underlying health benefits for active people (review). *J Nutr Biochem* 2003, 14:251–258
[365] Josse AR, Atkinson SA, Tarnopolsky MA, Phillips SM: Increased consumption of dairy foods and protein during diet- and exercise-induced weight loss promotes fat mass loss and lean mass gain in overweight and obese premenopausal women. *J Nutr* 2011, 141:1626–1634
[366] Falkowski, Mateusz et al. "Whey Protein Concentrate WPC-80 Improves Antioxidant Defense Systems in the Salivary Glands of 14-Month Wistar Rats." *Nutrients* vol. 10,6 782. 17 Jun. 2018, doi:10.3390/nu10060782
[367] Cury-Boaventura MF, Levada-Pires AC, Folador A, Gorjao R, Alba-Loureiro TC, Hirabara SM, Peres FP, Silva PR, Curi R, Pithon-Curi TC: Effects of exercise on leukocyte death: prevention by hydrolyzed whey protein enriched with glutamine dipeptide. *Eur J Appl Physiol* 2008, 103:289–294
[368] Witard OC, Turner JE , Jackman SR , Kies AK , Jeukendrup AE , Bosch JA , Tipton KD. High dietary protein restores overreaching induced impairments in leukocyte trafficking and reduces the incidence of upper respiratory tract infection in elite cyclists. Brain Behav Immun. 2014 Jul;39:211-9. doi: 10.1016/j.bbi.2013.10.002. Epub 2013 Oct 10
[369] Groen, B.B.; Horstman, A.M.; Hamer, H.M.; de Haan, M.; van Kranenburg, J.; Bierau, J.; Poeze, M.; Wodzig, W.K.; Rasmussen, B.B.; van Loon, L.J. Post-prandial protein handling: You are what you just ate. PLoS ONE 2015, 10, e0141582.
[370] Martin Kohlmeier, Department of Nutrition, UNC. Nutrient Metabolism, Structures, Functions and Genes, Second Edition. 2015 Elsevier Ltd. Chapter 3 Absorption, Transportation and Retention, page 51. ISBN: 978-0-12-387784-0
[371] Ahmed Ismaeel, Suzy Weems, and Darryn S. Willoughby. A Comparison of the Nutrient Intakes of Macronutrient-Based Dieting and Strict Dieting Bodybuilders. International Journal of Sport Nutrition and Exercise Metabolism, 2018, 28, 502-508 https://doi.org/10.1123/ijsnem.2017-0323
[372] Witard, O.C.; Jackman, S.R.; Breen, L.; Smith, K.; Selby, A.; Tipton, K.D. Myofibrillar muscle protein synthesis rates subsequent to a meal in response to increasing doses of whey protein at rest and after resistance exercise. Am. J. Clin. Nutr. 2014, 99, 86–95
[373] Moore, D.R.; Robinson, M.J.; Fry, J.L.; Tang, J.E.; Glover, E.I.; Wilkinson, S.B.; Prior, T.; Tarnopolsky, M.A.; Phillips, S.M. Ingested protein dose response of muscle and albumin protein synthesis after resistance exercise in young men. *Am. J. Clin. Nutr.* 2009, *89*, 161–168

[374] Macnaughton, L.S.;Wardle, S.L.;Witard, O.C.; McGlory, C.; Hamilton, D.L.; Jeromson, S.; Lawrence, C.E.; Wallis, G.A.; Tipton, K.D. The response of muscle protein synthesis following whole-body resistance exercise is greater following 40 g than 20 g of ingested whey protein. Physiol. Rep. 2016, 4, e128932
[375] Schoenfeld, Brad Jon, and Alan Albert Aragon. "How much protein can the body use in a single meal for muscle-building? Implications for daily protein distribution." *Journal of the International Society of Sports Nutrition* vol. 15 10. 27 Feb. 2018, doi:10.1186/s12970-018-0215-1
[376] Stokes T, Hector AJ, Morton RW, McGlory C, Phillips SM. Recent Perspectives Regarding the Role of Dietary Protein for the Promotion of Muscle Hypertrophy with Resistance Exercise Training. Nutrients. 2018 Feb 7;10(2):180. doi: 10.3390/nu10020180. PMID: 29414855; PMCID: PMC5852756.
[377] Jenner, Sarah L et al. "Dietary Intakes of Professional and Semi-Professional Team Sport Athletes Do Not Meet Sport Nutrition Recommendations-A Systematic Literature Review." *Nutrients* vol. 11,5 1160. 23 May. 2019, doi:10.3390/nu11051160
[378] Pasiakos SM , McLellan TM, Lieberman HR. The effects of protein supplements on muscle mass, strength, and aerobic and anaerobic power in healthy adults: a systematic review. Sports Med. 2015 Jan;45(1):111-31. doi: 10.1007/s40279-014-0242-2.
[379] MacKenzie K, Slater G, King N, Byrne N. The Measurement and Interpretation of Dietary Protein Distribution During a Rugby Preseason. Int J Sport Nutr Exerc Metab. 2015 Aug;25(4):353-8. doi: 10.1123/ijsnem.2014-0168. Epub 2015 Feb 12
[380] Spendlove J , Mitchell L, Gifford J, Hackett D, Slater G, Cobley S, O'Connor H. Dietary Intake of Competitive Bodybuilders. Sports Med. 2015 Jul;45(7):1041-63. doi: 10.1007/s40279-015-0329-4.
[381] Doering TM, Reaburn PR, Phillips SM, Jenkins DG ."Post-Exercise Dietary Protein Strategies to Maximize Skeletal Muscle Repair and Remodeling in Masters Endurance Athletes: A Review" by *International Journal of Sport Nutrition and Exercise Metabolism* © 2015 Human Kinetics, Inc
[382] Pablo M. García-Rovés,[†] Pedro García-Zapico, Ángeles M. Patterson, and Eduardo Iglesias-Gutiérrez. Nutrient Intake and Food Habits of Soccer Players: Analyzing the Correlates of Eating Practice. Nutrients. 2014 Jul; 6(7): 2697–2717. Published online 2014 Jul 18. doi: 10.3390/nu6072697. PMCID: PMC4113765
[383] Tran, Lee et al. "A new method to measure muscle protein synthesis in humans by endogenously introduced d9-leucine and using blood for precursor enrichment determination." *Physiological reports* vol. 3,8 (2015): e12479. doi:10.14814/phy2.12479
[384] Bohe, J.; Low, A.;Wolfe, R.R.; Rennie, M.J. Human muscle protein synthesis is modulated by extracellular, not intramuscular amino acid availability: A dose-response study. J. Physiol. 2003, 552, 315–324
[385]Moore, D.R.; Churchward-Venne, T.A.; Witard, O.; Breen, L.; Burd, N.A.; Tipton, K.D.; Phillips, S.M. Protein ingestion to stimulate myofibrillar protein synthesis requires greater relative protein intakes in healthy older versus younger men. J. Gerontol. A Biol. Sci. Med. Sci. 2015, 70, 57–62.
[386] Symons TB, Sheffield-Moore M, Wolfe RR, Paddon-Jones D. A moderate serving of high-quality protein maximally stimulates skeletal muscle protein synthesis in young and elderly subjects. Journal of the American Dietetic Association. 2009 Sep; 109(9):1582–6. Epub 2009/08/25.eng. [PubMed: 19699838]
[387] Nicolaas EP Deutz and Robert R Wolfe. Is there a maximal anabolic response to protein intake with a meal? Clin Nutr. 2013 April ; 32(2): 309–313. doi:10.1016/j.clnu.2012.11.018.
[388] Chow LS, Albright RC, Bigelow ML, Toffolo G, Cobelli C, Nair KS. Mechanism of insulin's anabolic effect on muscle: measurements of muscle protein synthesis and breakdown using aminoacyl-tRNA and other surrogate measures. Am J Physiol Endocrinol Metab. 2006 Oct;291(4):E729-36. Epub 2006 May 16.
[389] Wolfe RR. Protein supplements and exercise. Am J Clin Nutr. 2000 Aug; 72(2 Suppl):551S–7S. [PubMed: 10919959]
[390] Elango R, Humayun MA, Ball RO, Pencharz PB. Protein requirement of healthy school-age children determined by the indicator amino acid oxidation method. Am J Clin Nutr. 2011 Dec; 94(6):1545–52. [PubMed: 22049165]
[391] Humayun MA, Elango R, Ball RO, Pencharz PB. Reevaluation of the protein requirement in young men with the indicator amino acid oxidation technique. Am J Clin Nutr. 2007 Oct; 86(4):995–1002. [PubMed: 17921376]
[392] Pennings B, Groen B, de Lange A, Gijsen AP, Zorenc AH, Senden JM, et al. Amino acid absorption and subsequent muscle protein accretion following graded intakes of whey protein in elderly men. Am J Physiol Endocrinol Metab. 2012 Apr 15; 302(8):E992–9. [PubMed: 22338070]
[393] Hartman, J.W.; Moore, D.R.; Phillips, S.M. Resistance training reduces whole-body protein turnover and improves net protein retention in untrained young males. Appl. Physiol. Nutr. Metab. 2006, 31, 557–564
[394] Moore, D.R.; Del Bel, N.C.; Nizi, K.I.; Hartman, J.W.; Tang, J.E.; Armstrong, D.; Phillips, S.M. Resistance

training reduces fasted- and fed-state leucine turnover and increases dietary nitrogen retention in previously untrained young men. J. Nutr. 2007, 137, 985–991
[395] Fahey, T.D. (1998). Anabolic-androgenic steroids: mechanism of action and effects on performance. In: Encyclopedia of Sports Medicine and Science, T.D.Fahey (Editor). Internet Society for Sport Science: http://sportsci.org. 7 March 1998
[396] van Wayjen RG. Metabolic effects of anabolic steroids. Wien Med Wochenschr. 1993;143(14-15):368-75. PMID: 8256449
[397] Kicman, A T. "Pharmacology of anabolic steroids." *British journal of pharmacology* vol. 154,3 (2008): 502-21. doi:10.1038/bjp.2008.165
[398] Long, C.L.; Birkhahn, R.H.; Geiger, J.W.; Blakemore, W.S. Contribution of skeletal muscle protein in elevated rates of whole-body protein catabolism in trauma patients. Am. J. Clin. Nutr. 1981, 34, 1087–1093
[399] Institute of Medicine. Dietary Reference Intakes for energy, carbohydrates, fiber, fat, protein, and amino acids (macronutrients). Washington (DC): The National Academies Press; 2002/2005.
[400] Van Elswyk, Mary E et al. "A Systematic Review of Renal Health in Healthy Individuals Associated with Protein Intake above the US Recommended Daily Allowance in Randomized Controlled Trials and Observational Studies." *Advances in nutrition (Bethesda, Md.)* vol. 9,4 (2018): 404-418. doi:10.1093/advances/nmy026
[401] Devries MC, Sithamparapillai A, Brimble KS, Banfield L, Morton RW, Phillips SM. Changes in Kidney Function Do Not Differ between Healthy Adults Consuming Higher- f with Lower- or Normal-Protein Diets: A Systematic Review and Meta-Analysis. J Nutr. 2018 Nov 1;148(11):1760-1775. doi: 10.1093/jn/nxy197. PMID: 30383278; PMCID: PMC6236074.
[402] Phillips, Stuart M et al. "Optimizing Adult Protein Intake During Catabolic Health Conditions." *Advances in nutrition (Bethesda, Md.)* vol. 11,4 (2020): S1058-S1069. doi:10.1093/advances/nmaa047
[403] Bauer J, Biolo G, Cederholm T, Cesari M, Cruz-Jentoft A, Morley J, Phillips S, Sieber C, Stehle P, Teta D, et al. 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 2013;14:542–59.
[404] Darling AL, Millward D, Torgerson D, Hewitt C, Lanham-New S. Dietary protein and bone health: a systematic review and meta-analysis. Am J Clin Nutr 2009;90:1674–92.
[405] Papageorgiou M, Merminod F, Chevalley T, van Rietbergen B, Ferrari S, Rizzoli R, Biver E. Associations between age-related changes in bone microstructure and strength and dietary acid load in a cohort of community-dwelling, healthy men and postmenopausal women. Am J Clin Nutr. 2020 Oct 1;112(4):1120-1131. doi: 10.1093/ajcn/nqaa191. PMID: 32678420.
[406] Heaney RP, Layman D. Amount and type of protein influences bone health. Am J Clin Nutr 008;87(Suppl):1567S–70S.
[407]Nakayama AT, Lutz LJ, Hruby A, Karl JP, McClung JP, Gaffney-Stomberg E. A dietary pattern rich in calcium, potassium, and protein is associated with tibia bone mineral content and strength in young adults entering initial military training. Am J Clin Nutr. 2019 Jan 1;109(1):186-196. doi: 10.1093/ajcn/nqy199. PMID: 30615068.
[408] Jeukendrup, A.E. Carbohydrate and exercise performance: The role of multiple transportable carbohydrates. Curr. Opin. Clin. Nutr. Metab. Care 2010, 13, 452–457. ISSN 2072-6643
[409] Lindsay B. Baker, Ian Rollo, Kimberly W. Stein and Asker E. Jeukendrup. Acute Effects of Carbohydrate Supplementation on Intermittent Sports Performance. Nutrients 2015, 7, 5733-5763; doi:10.3390/nu7075249
[410] Maunder E, Podlogar T, Wallis GA. Postexercise Fructose-Maltodextrin Ingestion Enhances Subsequent Endurance Capacity. Med Sci Sports Exerc. 2018 May;50(5):1039-1045. doi: 10.1249/MSS.0000000000001516. PMID: 29232314.
[411] Malek AM, Hunt KJ, DellaValle DM, Greenberg D, St Peter JV, Marriott BP. Reported consumption of low-calorie sweetener in foods, beverages, and food and beverage additions by US Adults: NHANES 2007–2012. Curr Dev Nutr 2018;2(9):nzy054.
[412] Sheiham A., James W.P.T. A reappraisal of the quantitative relationship between sugar intake and dental caries: The need for new criteria for developing goals for sugar intake. BMC Public Health. 2014;14:863. doi: 10.1186/1471-2458-14-863
[413] Chazelas, et al. Sugary drink consumption and risk of cancer: results from NutriNet-Santé prospective cohort. *BMJ* 2019;365:l2408. http://dx.doi.org/10.1136/bmj.l2408
[414] Dunford et al. Types and Amounts of Nonnutritive Sweeteners Purchased by US Households: A Comparison of 2002 and 2018 Nielsen Homescan Purchases. Journal of the Academy of Nutrition and Dietetics. Volume 120, Issue 10, October 2020, Pages 1662-1671.e10
[415] Al-Waili, Noori et al. "Antibiotic, pesticide, and microbial contaminants of honey: human health hazards." *The Scientific World Journal* vol. 2012 (2012): 930849. doi:10.1100/2012/930849

[416] Gupta, Ruchi S et al. "Prevalence and Severity of Food Allergies Among US Adults." *JAMA network open* vol. 2,1 e185630. 4 Jan. 2019, doi:10.1001/jamanetworkopen.2018.5630
[417] Charlotte Debras, Eloi Chazelas, Bernard Srour, Emmanuelle Kesse-Guyot, Chantal Julia, Laurent Zelek, Cédric Agaësse, Nathalie Druesne-Pecollo, Pilar Galan, Serge Hercberg, Paule Latino-Martel, Mélanie Deschasaux, Mathilde Touvier, Total and added sugar intakes, sugar types, and cancer risk: results from the prospective NutriNet-Santé cohort, *The American Journal of Clinical Nutrition*, Volume 112, Issue 5, November 2020, Pages 1267–1279, https://doi.org/10.1093/ajcn/nqaa246
[418] Popkin BM, Hawkes C. Sweetening of the global diet, particularly beverages: Patterns, trends, and policy responses. Lancet Diabetes Endocrinol. 2016;4(2):174-186
[419] Vyth EL, Steenhuis I, Roodenburg A, Brug J, Seidell JC. Front-of-pack nutrition label stimulates healthier product development: A quantitative analysis. Int J Behav Nutr Phys Act. 2010;7(1):65.
[420] Grummon, et al. Sugar-Sweetened Beverage Health Warnings and Purchases: A Randomized Controlled Trial. Am J Prev Med. 2019 Nov;57(5):601-610. doi: 10.1016/j.amepre.2019.06.019. Epub 2019 Oct 2
[421] Lee MM, Falbe J, Schillinger D, Basu S, McCulloch CE, Madsen KA. Sugar-Sweetened Beverage Consumption 3 Years After the Berkeley, California, Sugar-Sweetened Beverage Tax. Am J Public Health. 2019 Apr;109(4):637-639. doi: 10.2105/AJPH.2019.304971. Epub 2019 Feb 21. PMID: 30789776; PMCID: PMC6417561.
[422] Lohner S, Toews I, Meerpohl JJ. Health outcomes of non-nutritive sweeteners: analysis of the research landscape. Nutr J. 2017;16(1):55
[423] Jensen, P.N., Howard, B.V., Best, L.G. *et al.* Associations of diet soda and non-caloric artificial sweetener use with markers of glucose and insulin homeostasis and incident diabetes: the Strong Heart Family Study. *Eur J Clin Nutr* 74, 322–327 (2020). https://doi.org/10.1038/s41430-019-0461-6
[424] Hirahatake KM, Jacobs DR, Shikany JM, Jiang L, Wong ND, Steffen LM, Odegaard AO. Cumulative intake of artificially sweetened and sugar-sweetened beverages and risk of incident type 2 diabetes in young adults: the Coronary Artery Risk Development In Young Adults (CARDIA) Study. Am J Clin Nutr. 2019 Sep 1;110(3):733-741. doi: 10.1093/ajcn/nqz154. PMID: 31374564; PMCID: PMC6736196.
[425] Thomson P, Santibañez R, Aguirre C, Galgani JE, Garrido D. Short-term impact of sucralose consumption on the metabolic response and gut microbiome of healthy adults. Br J Nutr. 2019 Oct 28;122(8):856-862. doi: 10.1017/S0007114519001570. Epub 2019 Sep 13. PMID: 31258108.
[426] EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS), Aguilar, F., Crebelli, R., Di Domenico, A., Dusemund, B., Frutos, M. J., Galtier, P., Gott, D., Gundert-Remy, U., Lambré, C., Leblanc, J. C., Lindtner, O., Moldeus, P., Mosesso, P., Parent-Massin, D., Oskarsson, A., Stankovic, I., Waalkens-Berendsen, I., Woutersen, R. A., Wright, M., … Mortensen, A. (2017). Statement on the validity of the conclusions of a mouse carcinogenicity study on sucralose (E 955) performed by the Ramazzini Institute. *EFSA journal. European Food Safety Authority*, *15*(5), e04784. https://doi.org/10.2903/j.efsa.2017.4784
[427] US Food & Drug Administration. Additional Information about High- Intensity Sweeteners Permitted for Use in Food in the United States [Internet]. 2018 (accessed 30 December 2020). Available from: https://bit.ly /31sxNJr. https://www.fda.gov/food/food-additives-petitions/additional-information-about-high-intensity-sweeteners-permitted-use-food-united-states
[428] Fitch C, Keim KS; Academy of Nutrition and Dietetics. Position of the Academy of Nutrition and Dietetics: use of nutritive and nonnutritive sweeteners. J Acad Nutr Diet. 2012 May;112(5):739-58. doi: 10.1016/j.jand.2012.03.009. Epub 2012 Apr 25.
[429] Policy Statement from The American Academy of Pediatrics. The Use of Nonnutritive Sweeteners in Children. PEDIATRICS Volume 144, number 5, November 2019:e20192765.
[430] E 955. EFSA Journal. http://onlinelibrary.wiley.com/journal/10.1002/(ISSN)1831-4732
[431] European Food Safety Authority. Scientific topic: Sweeteners [Internet]. 2019 (accessed 7 July 2019). Available from: http://www. efsa.europa.eu/en/topics/topic/sweeteners.
[432] Pepino MY. Metabolic effects of non-nutritive sweeteners. Physiol Behav 2015;152(Pt B):450–5.
[433] Bruyère Olivier, Ahmed H. Serge, Atlan Catherine, Belegaud Jacques, et al. Review of the nutritional benefits and risks related to intense sweeteners. Olivier et al. Archives of Public Health (2015) 73:41.
DOI 10.1186/s13690-015-0092-x
[434] Evert et al. Nutrition Therapy for Adults With Diabetes or Prediabetes: A Consensus Report. Diabetes Care 2019 May; 42(5): 731-754.https://doi.org/10.2337/dci19-0014

[435] Higgins KA, Mattes RD. A randomized controlled trial contrasting the effects of 4 low-calorie sweeteners and sucrose on body weight in adults with overweight or obesity. Am J Clin Nutr. 2019 May 1;109(5):1288-1301. doi: 10.1093/ajcn/nqy381. PMID: 30997499.
[436] Dereń K, Weghuber D, Caroli M, Koletzko B, Thivel D, Frelut ML, Socha P, Grossman Z, Hadjipanayis A, Wyszyńska J, Mazur A. Consumption of Sugar-Sweetened Beverages in Paediatric Age: A Position Paper of the European Academy of Paediatrics and the European Childhood Obesity Group. Ann Nutr Metab. 2019;74(4):296-302. doi: 10.1159/000499828. Epub 2019 Apr 23. PMID: 31013493.
[437] Gardner et al. Nonnutritive Sweeteners: Current Use and Health Perspectives. A Scientific Statement From the American Heart Association and the American Diabetes Association. *Circulation* July 24, 2012. DOI: 10.1161/CIR.0b013e31825c42ee
[438] Qi et al. Sugar-Sweetened Beverages and Genetic Risk of Obesity. N Engl J Med 2012; 367:1387-1396.DOI: 10.1056/NEJMoa1203039. October 11, 2012
[439] Pepin A, Stanhope KL, Imbeault P. Are Fruit Juices Healthier Than Sugar-Sweetened Beverages? A Review. Nutrients. 2019 May 2;11(5):1006. doi: 10.3390/nu11051006. PMID: 31052523; PMCID: PMC6566863.
[440] Ruanpeng D, Thongprayoon C, Cheungpasitporn W, Harindhanavudhi T. Sugar and artificially sweetened beverages linked to obesity: a systematic review and meta-analysis. QJM. 2017 Aug 1;110(8):513-520. doi: 10.1093/qjmed/hcx068. PMID: 28402535.
[441] Martyn D, Darch M, Roberts A, Lee HY, Yaqiong Tian T, Kaburagi N, Belmar P. Low-/No-Calorie Sweeteners: A Review of Global Intakes. Nutrients. 2018 Mar 15;10(3):357. doi: 10.3390/nu10030357. PMID: 29543782; PMCID: PMC5872775.
[442] NCBI Bookshelf. National Center for Biotechnology Information, U.S. National Library of Medicine8600 Rockville Pike, Bethesda MD, 20894 USA https://www.ncbi.nlm.nih.gov/books/NBK278991/table/diet-treatment-obes.table20nut/
[443] Nabors LO. Sweet choices: sugar replacements for foods and beverages. Food Technol. 2002;56:28–32.
[444] Belton, Kerry et al. "A Review of the Environmental Fate and Effects of Acesulfame-Potassium." *Integrated environmental assessment and management* vol. 16,4 (2020): 421-437. doi:10.1002/ieam.4248
[445] Food and Drug Administration: http://www.fda.gov/Food/IngredientsPackagingLabeling/FoodAdditivesIngredients/ucm397725.htm#AceK
[446] Food and Drug Administration (2006) Food labeling: health claims; dietary noncariogenic carbohydrate sweeteners and dental caries. Fed Reg 71:15559–15564 http://frwebgate.access.gpo.gov/cgi-bin/getpage.cgi?position=all&page=15559&dbname=2006_register. Accessed 10 June 2010
[447447] US Pharmacopeia. Sucralose Monograph. (Accessed December 30, 2020. http://www.pharmacopeia.cn/v29240/usp29nf24s0_m78575.html
[448] Arora S, Singh VP, Sharma V, Wadhwa BK, George V, Singh AK, Sharma GS. Analysis of sucralose and its storage stability in barfi. J Food Sci Technol. 2009;46:114–117.
[449] Berry, Colin et al. "Sucralose Non-Carcinogenicity: A Review of the Scientific and Regulatory Rationale." *Nutrition and cancer* vol. 68,8 (2016): 1247-1261. doi:10.1080/01635581.2016.1224366
[450] Sanchari Chattopadhyay, Utpal Raychaudhuri, and Runu Chakraborty Artificial sweeteners – a review. J Food Sci Technol. 2014 Apr; 51(4): 611–621. Published online 2011 Oct 21. doi: 10.1007/s13197-011-0571-1 PMCID: PMC3982014
[451] FDA Food Additives GRAS https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?fr=182.1745
[452] Casaburi et al. Carboxymethyl cellulose with tailored degree of substitution obtained from bacterial cellulose, Food Hydrocolloids, Volume 75,2018. Pages 147-156,ISSN 0268-005X,https://doi.org/10.1016/j.foodhyd.2017.09.002.(http://www.sciencedirect.com/science/article/pii/S0268005X17307853)
[453] Peter F. Stanbury, Stephen J. Hall. Media for industrial fermentations. Principles of Fermentation Technology (Third Edition), 2017. Sodium Carboxymethyl Cellulose. Lignocellulose Biorefinery Engineering.
[454] Sara Ranjbar, Sara Movahhed, Nabiollah Nematti and Roghayeh Sokotifar. Evaluation of the Effect of Carboxy Methyl Cellulose on Sensory Properties of Gluten-Free Cake. Research Journal of Applied Sciences, Engineering and Technology 4(19): 3819-3821, 2012. ISSN: 2040-7467. © Maxwell Scientific Organization, 2012
[455] Badwaik HR, Giri TK, Nakhate KT, Kashyap P, Tripathi DK. Xanthan gum and its derivatives as a potential bio-polymeric carrier for drug delivery system. Curr Drug Deliv. 2013 Oct;10(5):587-600.
[456] Palaniraj A., Jayaraman V. Production, recovery and applications of xanthan gum by *Xanthomonas campestris*. J. Food Eng. 2011;106:1–12. doi: 10.1016/j.jfoodeng.2011.03.035.

[457] Rosalam S., England R. Review of xanthan gum production from unmodified starches by *Xanthomonas comprestris* sp. Enzyme Microb. Technol. 2006;39:197–207. doi: 10.1016/j.enzmictec.2005.10.019
[458] Hu, Xiaolong et al. "Characterization and Antioxidant Activity of a Low-Molecular-Weight Xanthan Gum." *Biomolecules* vol. 9,11 730. 12 Nov. 2019, doi:10.3390/biom9110730
[459] The Food and Drug Administration. PART 172 -- FOOD ADDITIVES PERMITTED FOR DIRECT ADDITION TO FOOD FOR HUMAN CONSUMPTION. Sec. 172.695 Xanthan gum. https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?fr=172.695
[460] EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS) et al. "Re-evaluation of xanthan gum (E 415) as a food additive." *EFSA journal. European Food Safety Authority* vol. 15,7 e04909. 14 Jul. 2017, doi:10.2903/j.efsa.2017.4909
[461] Razavi,M.,Nyamathulla,S.,andKarimian,H.(2014).Hydrogelpolysaccharides of tamarind and xanthan to formulate hydro-dynamically balanced matrix tablets of famotidine. *Molecules* 19,13909–13931.doi:10.3390/molecules1909 13909
[462] Jagdale,S.C.,andPawar,C.R.(2014).Application of design of experiment for polyox and xanthan gum coated floating pulsatile delivery of sumatriptan succinate in migraine treatment. *Biomed.Res.Int*. 2014:547212.doi: 10.1155/2014/547212
[463] Feskanich D, Willett WC, Stampfer MJ, Colditz GA. Protein consumption and bone fractures in women. Am J Epidemiol. 1996 Mar 1;143(5):472-9.
[464] Munger RG, Cerhan JR, Chiu BC. Prospective study of dietary protein intake and risk of hip fracture in postmenopausal women. Am J Clin Nutr. 1999 Jan; 69(1):147-52.
[465] Wengreen HJ, Munger RG, West NA, Cutler DR, Corcoran CD, Zhang J, Sassano NE. Dietary protein intake and risk of osteoporotic hip fracture in elderly residents of Utah. J Bone Miner Res. 2004 Apr; 19(4):537-45. Epub 2004 Feb 9.
[466] Institute of Medicine. Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein, and Amino Acids. Washington, DC: National Academies Press, 2005.
[467] Martin WF, Armstrong LE, Rodriguez NR. Dietary protein intake and renal function. Nutr Metab (Lond). 2005 Sep 20; 2:25.
[468] McKenzie S, Phillips SM, Carter SL, Lowther S, Gibala MJ, Tarnopolsky MA. Endurance exercise training attenuates leucine oxidation and BCOAD activation during exercise in humans. Am J Physiol Endocrinol Metab. 2000 Apr; 278(4):E580-7.
[469] Miller BF, Olesen JL, Hansen M, Døssing S, Crameri RM, Welling RJ, Langberg H, Flyvbjerg A, Kjaer M, Babraj JA, Smith K, Rennie MJ. Coordinated collagen and muscle protein synthesis in human patella tendon and quadriceps muscle after exercise. J Physiol. 2005 Sep 15; 567(Pt 3):1021-33. Epub 2005 Jul 7.