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Fast, Medium & Slow Proteins with Gynostemma & Nutrients

Key Points at a Glance

Protein Digestion: Slow and Fast Absorption Impacts Postprandial Metabolism

- fast, medium and slow absorbed proteins have been identified
- absorption rates of amino acids from protein sources ranges from 1.4 g/h (raw egg white) to 8-10 g/h (whey protein isolate)
- fast proteins increase protein synthesis and oxidation and induce no change to protein catabolism
- slow proteins moderately increase protein synthesis and oxidation and induce significant inhibition of protein catabolism
- combining proteins with different absorption speeds may enhance postprandial kinetics (limited clinical data)
- combining slow and fast proteins may prolong and enhance muscle protein synthesis following exercise
- hydrolysis of proteins (e.g. marine collagen) results in faster absorption compared to non-hydrolysed forms
- a combination of fast and slow proteins may prolong the sense of satiety

Protein Types

- fast protein: whey; slow protein: egg, rice, pea, potato, casein
- dietary fish: slow protein; hydrolysed collagen from fish: probably medium to fast protein

Protein Clinical Studies

- individuals with sarcopaenia have low protein intake
- amino acid and micronutrient supplements combined with exercise training may be best way to reduce muscle wasting
- clinical trials support protein/amino acids for treatment of sarcopaenia as well as potential benefit in muscle strength and body composition
- in healthy adults 15-20 g of protein maximises synthesis of muscle proteins, but higher doses required in the elderly; BCAA's, especially leucine are particularly important, but not vital
- high protein intake increased satiety and thermogenesis; results for weight loss mixed
- fast protein may be more satiating in the short term, slow protein in the long term
- high-protein diet improved glycaemic control but not weight loss in obese type 2 diabetics
- replacing carbohydrate with protein may improve blood lipids
- amino acids are necessary for exercise-induced increases to muscle protein synthesis; synthesis plateaus once maximum level is reached (10 g protein stimulates, 20 g is maximum)
- protein supplementation: enhances muscle mass; results inconsistent for performance; may not enhance recovery after exercise

Gynostemma

- used as a folk remedy in southern China as a tonic
- main constituents are saponins: some are closely related to those in *Panax ginseng* root
- clinical trial using heat-processed extract (probably equivalent to 3 g/day of dried leaf) improved anthropometric parameters in obese volunteers

Selected Nutrients

- support the action of protein e.g. magnesium for muscle performance, zinc for muscle growth and wound healing; vitamin D and calcium for malnourishment
- support body systems e.g. chromium for blood glucose regulation; iodine and selenium for thyroid hormone production
- nutrients in diet reduce the risk of obesity (e.g. fibre) and/or chronic diseases such as type 2 diabetes, cardiovascular disease (e.g. magnesium, fibre)
- provide additional nutrition e.g. chia and flax seeds

Indications & Safety

- Muscle wasting conditions e.g. sarcopaenia, cachexia.
- Metabolic syndrome, type 2 diabetes; possibly adjunctive treatment for hyperlipidaemia.
- Weight management.
- Malnourishment, reduced immune function, wound healing, convalescence.
- Sports performance and muscle gain.
- Not advised in those with renal conditions or phenylketonuria.

Gynostemma

Gynostemma pentaphyllum, also known in China as Jiaogulan ("twisting vine orchid"), is a perennial creeping herb and a member of the Cucurbitaceae (cucumber family), that grows in southern China and many countries throughout Asia.

Generally, Gynostemma has not been used by herbalists in the official traditional systems but it has been used as a folk remedy. For example, in the mountainous regions of southern China, the plant's natural habitat, it has been used for an antifatigue effect, for general health and as a rejuvenating elixir – hence has been called an immortality herb.[1] A recent ethnobotanical investigation noted that Gynostemma whole plant decoction is used by the Maonan people in the southwest of China for rheumatism, bronchitis and stomach ache. Due to the remote location, the people in this region have relied upon self-medication and the knowledge of herbs has been passed orally from generation to generation.[2] Records indicate the plant was used as a vegetable in China (during famine) and in Japan.[1] Many herbal products, functional foods and beverages based on Gynostemma, including "total Jiaogulan saponin" tablets, have been available in East Asian markets since the early 1990s.[3,4]

Interest in the herb began seriously in the mid-1970s when a Japanese scientist began researching sweet compounds derived from plants.[1] Saponins were found to be major constituents of Gynostemma.[1] Phytochemical studies have identified a large number of dammarane-type glycosides (called gypenosides), which are closely related to the ginseng saponins. Eight of the gypenosides are identical to the protopanaxadiol-type ginsenosides found in *Panax ginseng* root, including ginsenosides Rb_1, Rc and Rd.[5,6,7] The total saponin content varies according to growing location and time of collection, and was reported in 1993 to be about 2.4% of the dried herb.[6]

Clinical Studies

In vitro research found that two dammarane-type saponins (damulin A and damulin B) which were isolated from *Gynostemma pentaphyllum* leaves, strongly stimulated AMP-activated protein kinase (AMPK).[8] (AMPK is an important sensor and regulator of glucose, lipid and energy metabolism.) At a later date, a double-blind clinical trial was conducted in South Korea to investigate the potential antiobesity effect of Gynostemma.[9] Because the ability of an ethanol extract of Gynostemma leaf to activate AMPK *in vitro* was increased by autoclaving (heating the extract increased the levels of damulin A and B),[10] a heat-processed extract was administered in this trial.[9] Obese, but otherwise healthy volunteers were randomly assigned to Gynostemma and placebo groups. Eighty participants received and completed the 12-week treatment (40 in each group). They were on average: 40 years old, weighed 74.6 kg, had body mass index of 27.5 kg/m^2. The concentrated extract made by extraction with 50% ethanol and heating to 121 ºC for 4 hours, was given at a dosage to match the lower end of range used in folk medicine: 3 g/day of dried leaf. The extract provided 5 mg/day of damulin A. For the trial period, volunteers were asked to maintain their normal diet and physical activity, and diet records indicated their energy and nutrient intakes did not significantly change over the course of the trial. Treatment with Gynostemma significantly decreased many anthropometric parameters from baseline, and these were significant compared to the changes experienced by the placebo group. *See Table 1 and Safety section.*[9]

Several small, controlled trials conducted in Vietnam found that Gynostemma "tea" improved glycaemic control in type 2 diabetics. Body measurements, including BMI, were unchanged.[11,12,13] The dose was potentially very high. A concentrated, non-galenical extract of the whole plant was taken at a dose of 6 g/day for periods ranging from 4 to 12 weeks.

Anthropometric Parameters	Gynostemma	Placebo	Significance
total abdominal fat area (cm^2)	-20.90	-2.87	$p < 0.05$
body weight (kg)	-0.98	-0.21	$p < 0.05$
body fat mass (kg)	-1.05	-0.36	$p < 0.0001$
percent body fat (%)	-1.03	0.51	$p < 0.0001$
body mass index (kg/m^2)	-0.37	-0.06	$p < 0.05$
waist circumference (cm)	-2.49	-1.33	$p < 0.05$

Table 1. Changes in selected anthropometric parameters after 12 weeks' intake of Gynostemma leaf extract or placebo in obese volunteers.

Catalysts
• Enzymes are protein molecules which combine with substrates to catalyse chemical reactions in the body. Most physiological processes in the human body are dependent on enzymes, including energy production, contraction of neuromuscular tissue, blood coagulation digestion.
Hormonal messenger
• Hormones can be synthesised from cholesterol (steroid hormones), or amino acids. The following amino acids are precursors to endogenous hormones: - tyrosine is a precursor to catecholamine, and combines with iodine to synthesise thyroid hormones - tryptophan is essential for the production of melatonin in brain tissue - insulin is made up of two polypeptide chains linked by a disulfide bridge - glucagon, parathyroid hormone and calcitonin are structurally a single polypeptide chain
Structural elements
• Contractile proteins (predominately actin and myosin) are essential components of cardiac, skeletal and smooth muscles. • Fibrous proteins include collagen, elastin and keratin and form essential structures of bone, hair, skin, teeth, tendons, cartilage, blood vessels and nails.
Buffers
• Proteins, via their amino acid constituents serve as buffers. They act as either hydrogen donors or acceptors to maintain healthy pH ranges in blood, cells and body tissues.
Fluid balancers
• Proteins attract and hold water into a particular area, therefore contributing to osmotic pressure and fluid balance. Low protein (such as albumin) levels in blood or plasma can result in an inability to retain water and subsequent oedema due to leaking of fluid into interstitial spaces.
Immunoprotectors
• Immunoglobulins and antibodies are protein molecules which play an essential role in immune defences.
Transporters
• Transport proteins combine with vitamins, minerals and other substances, providing a vehicle to carry substances through the body as well as into and out of cells.
Acute phase responders
• In response to acute infection, injury or inflammation, these proteins are released by the liver and serve to protect the body, stimulate the immune system, promote wound healing and remove free iron from the circulation.

Table 2. Functional Roles and Metabolic Activities of Proteins.

Proteins

Protein is a macronutrient comprised of complex three dimensional polypeptide chains. The sequence of amino acids and structure of the protein defines digestibility and function, as well as interaction with other molecules.

Proteins undergo continual catabolism for use as free and branch chain amino acids or resynthesis into new protein structures, according to the needs of the body, and dietary intake. The innumerable protein structures, and rate of resynthesis give rise to diverse metabolic activities, including those outlined in *Table 2*.[14]

Rate of Protein Digestion: Slow and Fast Absorption Impacts Postprandial Metabolism

For dietary carbohydrates, the impact of absorption rate on metabolic and hormonal responses to a meal is well known by the science and health community, and commonly accepted as the glycaemic index (GI).[14] Increasing evidence indicates that, like dietary carbohydrates, the rate of protein digestion and absorption differs significantly between different protein sources, and impacts metabolic and hormonal responses within the body.[15,16] This concept was first described in the literature by Boirie and colleagues, and has been coined "fast" and "slow" proteins.

Studies investigating impacts of protein absorption rate on metabolism have mostly used whey protein (WP) and casein (CAS) as model proteins.[15] Boirie et al demonstrated that in healthy young men given a single protein meal of WP or CAS, WP induced a faster increase in plasma amino acid concentrations, resulted in higher concentrations and also a faster decline; WP was therefore classified a fast protein. Fast protein ingestion was associated with increased protein synthesis and oxidation, but no change to protein catabolism. CAS induced a relatively slower, lower and prolonged increase in plasma amino acids and better nitrogen (protein) retention. It was therefore, classified a slow protein. Slow protein ingestion was associated with a moderate increase of protein synthesis and oxidation and significant inhibition of protein catabolism. Rapidly absorbed protein, despite stimulating greater protein synthesis, also stimulates Protein oxidation, and hence resulting in a lower net protein gain than slowly absorbed protein.[17] *Details of this study can be seen in Table 3*.[16] The amino acid composition, and therefore the nitrogen content, of the two supplemental proteins varies in this study. However, later studies by the same authors administered isonitrogenic doses and found it did not change the outcomes.

Fast Protein (WP)	Slow Protein (CAS)
Postprandial plasma amino acid concentration*	
• Fast, dramatic and short peak in plasma amino acid concentrations. • At 100 min, all 18 amino acids increased significantly ($p < 0.05$) compared to baseline and concentrations had peaked. • At 300 min, concentration returned to basal levels for most amino acids.	• Slow, moderate and prolonged increase in plasma amino acid concentrations. • At 100 min, amino acid concentration increased less with CAS than with WP; 13/18 amino acids had increased significantly ($p < 0.05$). • At 300 min, concentration of most amino acids remained above baseline. This difference was significant for 11/18 amino acids.
Postprandial protein synthesis†	
• Significant increase in protein synthesis. • Total protein synthesis was stimulated by 68% (average from 40-140 min).	• Moderate increase in protein synthesis. • Total protein synthesis was stimulated by 31% (average from 40-140 min).
Postprandial protein oxidationΔ	
• Significant increase in protein oxidation. • At 100 min, total leucine oxidation increased from 0.48 ± 0.13 µmol/kg/min at baseline to 1.50 ± 0.33 µmol/kg/min. • At 420 min, total leucine oxidation returned to baseline. • Exogenous leucine oxidation was completed within 420 min.	• Moderate increase in protein oxidation. • At 100 min, leucine oxidation increased from 0.35 ± 0.11 µmol/kg/min at baseline to 0.78 ± 0.34 µmol/kg/min. • At 420 min, total leucine oxidation remained slightly elevated but was not statistically different from baseline (0.53 ± 0.15 µmol/kg/min). • Exogenous leucine oxidation was not complete within 420 min.
Postprandial whole body protein catabolism	
• No change to net protein catabolism. • Mean endogenous leucine derived from protein breakdown between 120-420 min following ingestion was 1.38 ± 0.04 µmol/kg/min, a non-significant change from baseline.	• Whole body protein catabolism was progressively and durably inhibited. • Mean endogenous leucine derived from protein breakdown between 120-420 min following ingestion was 1.08 ± 0.07 µmol/kg/min, a 34% decrease from baseline ($p < 0.05$).

Table 3. Key differences in absorption kinetics and metabolic response to ingestion of fast and slow proteins.[16]

Notes: * WP and CAS meals were matched for leucine content, but were not isonitrogenous. Amino acid intake was higher with CAS. Despite this, Cmax was higher with WP. † Difference between groups was not significant although there was a trend for higher protein synthesis with fast protein. Δ Total leucine oxidation increased with both groups, and was significantly higher with fast protein group.

Differing absorption kinetics of slow and fast proteins have been supported by several other studies.[18,19,20,21] A small single-blinded trial compared dietary nitrogen flux and rates of peptide release of WP and CAS in the jejunum of 16 healthy human subjects. Subjects were equipped with a double lumen naso-gastric tube that migrated to the proximal jejunum, and jejunum effluent samples were collected at 30-minute intervals for 6 hours following ingestion of 30 g WP or CAS. Exogenous nitrogen delivery was significantly higher after consumption of WP ($p < 0.0001$), and was complete after 3 hours. CAS-derived dietary nitrogen was progressively emptied for 6 hours during the postprandial period, *as shown in Figure 1*.[18]

It has been proposed that **slow and fast proteins can be combined to prolong muscle protein synthesis following exercise**,[22] and there is some evidence to support this. Clinical application of fast and slow proteins may be different in different populations. Interestingly, postprandial responses to fast and slow proteins have been reported to differ in elderly subjects. One preliminary study found (contrary to what had been observed in young people) fast protein (provided as WP) induced a better postprandial leucine balance than slow protein (provided as CAS).[17]

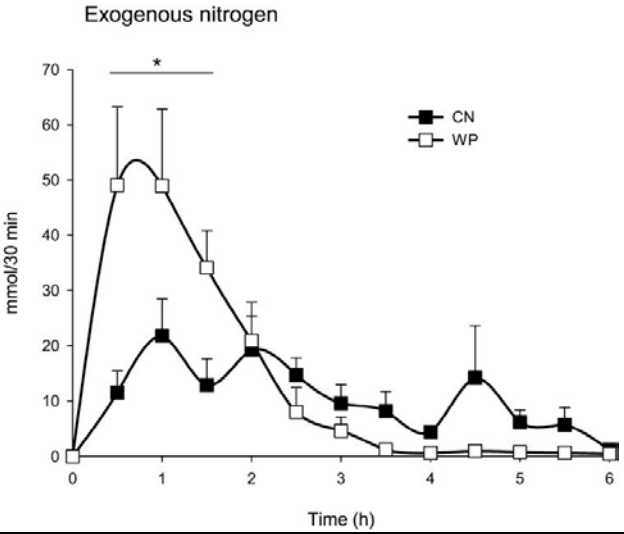

Figure 1. Mean (±SD) contributions of exogenous nitrogen to total nitrogen in the jejunum of subjects after the ingestion of CAS (referred to here as CN) or WP.[18]

Notes: 8 subjects per group. * Significantly different at each time point, $p < 0.05$ (post hoc test).

Data on digestion kinetics of other proteins are reported, although they are generally not classified as fast, medium or slow. Comparison of absorption rate of some proteins can be difficult, as different studies apply different methodologies. Methods may include ingestion of protein with other dietary foods or nutrients, and the study design may employ less accurate methodologies or have design limitations.[21] Despite this, some interpretations can be made with regard to absorption rates of various protein sources as fast, medium and slow proteins.

Absorption of free amino acids, whey protein and hydrolysed proteins appear to be fastest, followed by pea protein and rice protein. Raw and cooked egg white, and potato protein are reported to have the slowest absorption rate. Dietary fish protein was found to have slow absorption, although supplemental marine collagen from fish is a hydrolysed protein; broken down via hydrolysis to enable easier and faster digestion.[23] The absorption rate of marine collagen has not been reported, although, it would be reasonable to expect marine collagen to have medium to fast absorption, as hydrolysis is known to increase the rate of absorption. *Findings on absorption rates and kinetics of various protein sources are detailed in Table 4.*

Trial Details	Results	Ref
Randomised, double blind, crossover, pharmacokinetic study; 10 resistance-trained male volunteers administered 48 g RPI or WPI after 12 h overnight fast, separated by a washout period (7 d)	• WPI and RPI showed a significant difference between Tmax for EAA (RPI 87 ± 7 min, WPI 67 ± 4 min; p = 0.03), NEA (RPI 97 ± 4 min, WPI 71 ± 5 min; p < 0.001) and TAA (RPI 93 ± 4 min, WPI 69 ± 3 min; p < 0.001) with a significantly shorter period of time to reach Cmax levels observed for WPI • WPI was described as a fast protein, whereas RPI was described as a medium to slow protein • no significant differences were detected for AUC with RPI showing a non-significant 6.8% lower TAA concentration in the blood based on AUC • no significant differences were detected for Cmax	24
Randomised, double blind, cross-over study; 20 g protein from WP, pea protein, potato protein, soy protein or casein consumed by 8 healthy adults; digestion kinetics of potato protein studied *in vitro* and compared with reference proteins	• highest digestion found for WP, followed by soy, pea, casein and potato proteins • contrary to whey and casein, potato protein did not result in any changes in plasma insulin or glucose levels	25
17 T2Ds, untreated human subjects; active arm ingested 25 g of protein from 7 protein sources (in addition to 50 g glucose), controls received 50 g glucose only	• egg white, fish protein and beef produced a slow and mild increase in postprandial TAA • soy, turkey and cottage cheese produced a moderate increase in postprandial TAA • gelatine produced a fast increase in postprandial TAA • peripheral glucagon concentration was decreased in glucose alone, and increased following all meals with protein	26
Randomised crossover study, 10 healthy adults; absorption of 25 g of cooked and raw egg protein	• cooked egg protein recovered CO_2 of 68.92% over 6 h (17.23 ± 0.69 g), giving an absorption rate of 2.9 g/h • raw egg protein recovered CO_2 of 32.8% over 6 h (8.2 ± 0.94 g), giving an absorption rate of 1.4 g/h	27
Randomised crossover study, 24 overweight and moderately obese young men and women administered WP, CAS or hydrolysed CAS over 24 hours; EE, appetite regulation and substrate oxidation parameters were measured	• no differences in 24 h on postprandial EE or appetite regulation • lipid oxidation, estimated from RQ, was higher after consumption of WP than after consumption of HC during daytime (p = 0·014) as well as during time after the breakfast meal (p = 0·008) when the food was provided • NEFA concentrations were higher after consumption of WP than after consumption of HC and CAS (p = 0·01) • no overall difference in concentration of insulin or GLP-1 • WP, CAS and HC induced similar effects on EE and appetite regulation, except for lipid oxidation, where RQ values suggest that it is higher after consumption of WP	28
Crossover study, 10 healthy overweight men with an elevated waist circumference (> 94 cm); WP, CAS or LWP incorporated as 15% energy in a high-fat meal	• compared with WP and LWP (which did not differ), CAS markedly reduced postprandial TGs, achieving a 22 ± 10% reduction in the 6-h area under the curve (p < 0.05) • similar trends were shown for plasma chylomicrons (apoB-48; p < 0.05) • no significant differences in other parameters	29
12-wk, randomised, double blind, diet intervention study, 63 adults; 60 g milk protein (WP or CAS) and 63 g milk fat (with high or low MC-SFA content) daily	• apoB-48* response decreased significantly after WP compared with CAS (p = 0.025) independently of fatty acid composition • CAS resulted in significant increase in postprandial GLP-1 response compared with WP (p = 0.003) • no significant differences for other parameters • WP decreased postprandial chylomicron response compared with casein in those with abdominal obesity, indicating a beneficial impact on CVD risk • estimates absorption rates were 8-10 g/h and 6.1 g/h for WP and CAS, respectively	30

Crossover study, 12 T2Ds; WP, cod (fish), CAS, gluten compared for effect in response to high-fat meal; blood samples collected over 8 h after ingestion of test meal (100 g butter + 45 g carbohydrate + 45 g of respective protein)†	• incremental area under the curve for triglyceride was significantly lower for WP than all other proteins • RP response was lower with WP compared to CAS and cod in the chylomicron-rich fraction • RP response was higher with WP compared to cod and gluten in the chylomicron-poor fraction • free fatty acids and glucose response were lower with WP • no significant differences in insulin, glucagon, GLP-1 and glucose-dependent insulinotropic peptide responses • the data suggest that as a supplement to a fat-rich meal in patients with T2D, WP seems to outperform other proteins in terms of postprandial lipaemia improvement, possibly because of the formation of fewer chylomicrons or increased clearance of chylomicrons	31
Randomised placebo controlled trial, 68 physically active men; 30 g SMP, CAS or placebo twice daily or 3 times daily on training days for 10 wk	• after 4 and 10 weeks, muscle fatigue was significantly lower ($p < 0.05$) in SMP group (−326.8 ± 114.1 W and −296.6 ± 130.1 W, respectively) compared to placebo (−439.2 ± 153.9 W and −479.2 ± 138.1 W, respectively) and CAS (−415.1 ± 165.1 W and −413.7 ± 139.4 W, respectively) • maximal muscle power, strength, endurance and thickness were not statistically different between groups • authors described SMP as a fast protein, and CAS as a slow protein, and suggested the reduced muscle fatigue may be attributed to absorption speed	32

Table 4. Selected studies reporting absorption rate and kinetics of various protein sources.

Abbreviations: apoB-48: apolipoprotein B-48; AUC: area under the concentration versus time curve; CAS: casein; Cmax: maximum plasma concentration; CO_2: carbon dioxide; CVD: cardiovascular disease; d: day; EAA: essential amino acids; EE: energy expenditure; FFAs: free fatty acids; GIP: gastric inhibitory polypeptide; GLP-1: glucagon-like peptide-1; h: hour; HC: hydrolysed casein; LWP: α-lactalbumin-enriched whey protein; MC: micellar caseins; MC-SFA: medium-chain saturated fatty acids; MSPI: milk soluble protein isolate; NEA: non-essential amino acids; NEFA: non-esterified fatty acids; NPPU: net postprandial protein utilisation; PBV: postprandial biological value; RID: real ileal digestibility; RP: retinyl palmitate; RPI: rice protein isolate; RQ: respiratory quotient; SMP: solubilised milk protein; Tmax: time to reach maximum concentration; TAA: total amino acids; T2Ds: type 2 diabetics; TGs: triglycerides; TMP: total milk protein; wk: week; WP: whey protein WPI: whey protein isolate

Notes: * ApoB-48 reflects chylomicrons of intestinal origin. † Study did not measure plasma amino acid and absorption. However, time to effect on parameters measured may indicate relative absorption rates.

Clinical Studies

Muscle Wasting Conditions Including Sarcopaenia and Cachexia

Loss of skeletal muscle mass and strength is a common phenomenon. It exists as sarcopaenia in otherwise healthy ageing populations, and cachexia in response to chronic illnesses, where it is also accompanied by weight loss. Muscle wasting may also be seen in malnutrition, lack of physical activity and as a side effect of some drugs.[33]

Pathophysiologic mechanisms contributing to sarcopaenia include, atrophy of muscle fibres (especially of the fast Type II fibres), a decrease in motor units, and an accumulation of fat within the muscle. Reduced anabolic drive in ageing may lead to a decrease in muscle synthesis. This is because ageing is associated with lower testosterone levels, insulin-like growth factor 1 (IGF-1), and insulin resistance. Testosterone, insulin, and IGF-1 are potent activators of the Akt pathway, which stimulates muscle protein synthesis. These hormones also inhibit forkhead box proteins (Fox-0), which decrease protein degradation. Many of these mechanisms are common to cachexia, although in cachexia, inflammation, increased energy expenditure and impaired intestinal barrier function have been implicated. Disease related symptoms, mental health effects and medications may reduce appetite, further potentiating a hyper-catabolic state in cachexia.[33]

A 2015 review of nutrition and sarcopaenia included a cross-sectional study combining five datasets (n = 900) that found 77% of all participants had lower than recommended protein intake.[34] Another 2015 review on muscle wasting and treatment options in ageing and chronic illness reported exercise training was the treatment of choice, and the provision of **nutritional supplements incorporating essential amino acids, protein and micronutrients combined with exercise training may be the best method of attenuating muscle wasting**. This review included only two trials on nutritional supplementation and wasting.[33]

A third 2015 review of 67 studies, including epidemiological and controlled studies, investigated nutritional management of sarcopaenia. Eighteen articles, including 8 randomised controlled trials and 2 cross-sectional studies, covered research on amino acids and protein. Adequate intake of protein and amino acids was reported as essential in sarcopaenia prevention and intervention. Two studies involving healthy adults found a dose of approximately 15-20 g of protein (7.5 g essential amino acids) is sufficient to maximise the synthesis of muscle proteins, although higher doses are required in the elderly (25 to 30 g of protein per meal, containing approximately 2.5 to 2.8 g of leucine).[35]

These findings were supported by a study comparing various doses of amino acids on muscle protein synthesis (MPS) in healthy young and elderly men. Consumption of 5 g and 10 g of EAA's in the young and elderly respectively resulted in equivalent stimulation of MPS.[36] The dose response curve of MPS in elderly and young muscle following protein digestion (at rest) *can be seen in Figure 2*.[37]

There are a significant number of studies investigating the use of protein and amino acids in muscle wasting. *Results of a random selection of these studies are detailed in Table 5*.

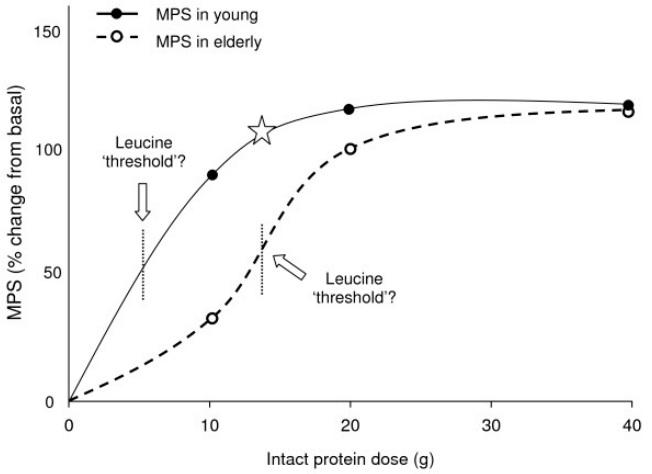

Figure 2. Dose response curve of MPS in elderly and young muscle (at rest) following protein ingestion.[37]

Trial Details	Results	Ref
Sarcopaenia		
Randomised double blind placebo-controlled; 53 males, 12-wk guided resistance training programme (3 x/wk) and collagen peptides (15 g/d, sourced from pork†) or placebo (silica)	• following the training programme, all participants showed significantly higher levels for FFM, BM, IQS of the right leg and SMC with significantly lower levels for FM, but the effect was significantly more pronounced in those receiving collagen peptides compared to placebo: - FFM (4·2 kg vs 2·9 kg, respectively; p < 0·05) - IQS (16·5 Nm vs 7·3 Nm, respectively; p < 0·05) - FM (-5·4 kg vs -3·5 kg, respectively; p < 0·05)	38
Randomised, open label, crossover, placebo-controlled trial; 41 volunteers aged 66-84 years, EAA (8 g, twice a day) or placebo	• significant increases in whole-body lean mass in all areas were seen after 6 months and more consistently after 18 months of oral nutritional supplementation with EAA • fasting blood glucose, serum insulin, and insulin resistance by HOMA index significantly decreased with EAA • significant reductions in serum TNF-alpha and a significant increase in serum IGF-1 and IGF-1/TNF-alpha	39
Muscle wasting and reduced physical activity		
Randomised, placebo controlled trial; 100 elderly volunteers (65 years or older) with reduced physical activity, 12 g/day AAs or placebo for 3 months	• AAs significantly increased 6-min walk distance (p < 0.001) • AAs significantly increased maximal isometric muscle strength (p < 0.001) • peak stress LVEF was higher in AA group (p < 0.01) • left ventricular response to exercise normalised in 75% of patients with abnormal response at baseline, but remained unchanged in the placebo group	40
Obesity		
Randomised, double blind; 80 older adults, 13-wk weight loss program; hypocaloric diet (2600 kcal/d), resistance training 3 x/wk; WP/leucine (equivalent 20.7 g/2.8 g per serve), vitamin D (equivalent 800 IU per serve) and other macro- and micronutrients, 10 serves/wk or an isocaloric control	• 13-wk change in appendicular muscle mass, was different in the intervention and control groups (+0.4 ± 1.2 kg and -0.5 ± 2.1 kg, respectively; p = 0.03) • both intervention and control groups decreased in body weight (-3.4 ± 3.6 kg and -2.8 ± 2.8 kg; both p < 0.001) and fat mass (-3.2 ± 3.1 kg and -2.5 ± 2.4 kg; both p < 0.001), with no differences between groups • muscle strength and function improved over time without significant differences between groups	41
Stable chronic heart failure with muscle loss		
Randomised, double-blind trial; 38 patients, 8 g/day EAA or placebo for 2 months	• body weight increased by > 1 kg in 80% of the active arm (mean 2.96 kg), and in 30% of the control group (mean 2.3 kg) with a significant difference found between the groups (p < 0.05) • changes in arm muscle area, nitrogen balance and insulin resistance by HOMA index were similar between the two groups • plasma lactate and pyruvate levels increased in controls (p<0.01 for both), and decreased in the active arm (p < 0.01 and p < 0.02 respectively) • active arm, but not controls improved exercise output, peak oxygen consumption and walking test	42

Elderly malnourishment		
Randomised, controlled trial; 210 patients, 24 g protein, 500 mg calcium and 400 IU vitamin D daily in addition to telephone counselling with a dietician compared to usual care for 3 months	• body weight increased more in the active arm than in the control group; this was significant for the highest body weight category only (mean difference 3.4 kg, 95% CI = 0.6–8.1) • no significant differences were reported for functional limitations (such as climbing stairs, getting up from and sitting down in a chair), physical performance, physical activities, fat-free mass or handgrip strength but there were significantly fewer falls in the supplemented group	43

Table 5: Selected randomised trials on protein, amino acid and nutritional supplementation in muscle wasting.

Abbreviations: AA: amino acids; BM: bone mass; EAA: essential amino acids; FFM: fat-free mass; FM: fat mass; HOMA: homeostasis model assessment; IGF-1: insulin-like growth factor-1; IQS: isokinetic quadriceps strength; LVEF: left ventricular ejection fraction; SMC: sensory motor control; TNF-alpha: tumour necrosis factor-alpha; WP: whey protein

Note: † Information about proprietary product provided to the author from the manufacturer.

Branched-chain amino acids (BCAAs), especially leucine are of particular relevance in muscle wasting conditions due to their stimulatory effect on MPS. Leucine is the only BCAA shown to independently stimulate MPS.[44,45] Two studies, published in1999 and 2004, found that 2-3 g or up to 0.05 g/kg bodyweight of leucine was required to maximise MPS,[46,47] with one study reporting greater muscle stimulation from 3.5 g leucine, compared to 1.8 g.[35]

However, other factors in addition to leucine and BCAA's influence MPS. This is evident in the results of a randomised controlled trial of hydrolysed collagen peptides on muscle mass and muscle function following resistance training in sarcopaenia (*described in Table 5*). The study reported significant improvements in body composition and muscle function parameters in the hydrolysed collagen group, despite a relatively low BCAA and leucine content.[38]

There is a growing body of evidence to suggest that metabolic factors associated with obesity and diabetes induce the progression of sarcopaenia (sarcopaenic obesity). Calorie restriction has been suggested for sarcopaenic obesity, although safety and effectiveness has not been established Ensuring optimal dietary protein would be beneficial to support muscle mass, body composition and satiety.[34] Other intervention to support metabolic health would be valuable.

Weight Management, Metabolic Effects

A high proportion of dietary energy from protein has been reported in a number of studies to increase weight loss, and prevent regaining weight following losses. Other studies have reported mixed results.[48,49,50] Proteins have been demonstrated to induce satiety, increase secretion of gastrointestinal hormones, reduce hunger and increase diet-induced thermogenesis. A 2013 review of protein supplementation in weight management identified a number of potential mechanisms for these effects, including:[48]

- Protein is an effective secretagogue of cholecystokinin (CCK), glucagon-like peptide 1 (GLP-1), and peptide YY, hormones are secreted in the gut in response to food intake which act as satiety signals.
- Protein suppresses plasma ghrelin, commonly known as the hunger hormone.
- Increasing protein intake enhances the satiating effect of circulating leptin in the central nervous system.
- Tryptophan (one of the EAAs) is a precursor of the neurotransmitter serotonin, which acts as an anorexigenic signal in the brain, therefore stimulating satiety. (Egg white is relatively rich in tryptophan.)
- Leucine and lysine are ketogenic amino acids, and it has been shown that appetite decreases under ketogenic conditions.
- Protein may stimulate incretin hormones such as GLP-1 thereby stabilising blood sugar levels postprandially.

A review of the effects of high-protein diets on the thermogenesis, satiety and weight loss found higher protein intake increases thermogenesis and satiety compared to lower protein intake. The review included 48 randomised controlled trials with 50 outcome comparisons. Reduced energy intake was reported, although weight loss and fat loss were inconsistent. Studies investigating these effects over periods longer than 6 months were limited. Trial participants included healthy volunteers as well as patients with metabolic and weight issues. Results from studies that compared high-protein diet with a lower protein diet, as well as high-protein versus high-carbohydrate diet, were described. (As a general rule, high-protein diets included protein intakes that met or exceeded 25% of dietary energy. Some of the trials in this review evaluated diets exceeding 60% protein.)[51]

- Fifteen studies investigated thermogenesis, with all showing significant thermogenic effects with higher protein intake.
- Thermogenesis was reported as 20-35% of energy intake for proteins, compared to 5-15% in carbohydrates.
- Eleven of fourteen studies on satiety found that protein preload (ranging from 29-75% of protein in meal) significantly increased subjective ratings of satiety.

- Seven of fifteen studies of higher protein diets in weight loss reported significant benefits. Higher protein diets may facilitate weight loss when compared to a lower protein diet in the short term (within 6 months). Long-term results were limited.
- Three of 10 studies reported a statistically significant greater fat loss with higher versus lower protein diets.
- Macronutrient ratios and study designs varied between groups, with studies of longer duration yielding better results.
- Increasing proteins and decreasing carbohydrates was seen to improve blood lipids, and epidemiologic studies demonstrated that higher protein diets are associated with lower blood pressure and reduced risk of coronary heart disease.

A 2013 review of randomised clinical trials examined the metabolic effects of fast and slow proteins. Fast protein, provided as WP, was reported to be more satiating in the short term (90-180 minutes), whereas slow protein, provided as CAS, may be more satiating in the same participants when followed over a longer period of time (330 minutes). Effect on energy expenditure, body weight, and body composition were conflicting, possibly due to complexities and limitations in the study design, or confounding factors which may influence protein stimulated metabolic effects. In addition, the review discussed studies indicating that. Fast protein (provided as WP) was found to stimulate secretion of the incretin hormones glucagon-like peptide-1 and glucose-dependent insulinotropic polypeptide to a greater extent than other proteins.[48] Whether this effect was due to the faster rate of absorption of WP or other factors such as amino acid profile was not explored.

A 2013 systematic review and meta-analysis of 20 randomised, controlled trials evaluated the effect of various dietary approaches to T2D. The low carbohydrate (9 trials), low GI (3 trials), Mediterranean (4 trials) and high-protein (2 trials) diets resulted in significant improvements in glycaemic control: glycated haemoglobin (HbA_{1c}) reductions of 0.12% ($p = 0.04$), 0.14% ($p = 0.008$), 0.47% ($p < 0.00001$) and 0.28% ($p < 0.00001$), respectively, compared with their respective control diets. An increase in HDL-cholesterol was seen in all diets except the high-protein diet. The high-protein diet studies enrolled overweight and/or obese adults, and both trials were 12 months in duration. Protein content of the high-protein diets was approximately 30%, although one trial compared a high-protein diet with a low-protein diet and the other study compared a high-protein diet with a high-carbohydrate diet. Neither study showed any significant differences in glycaemic control, but pooled data showed significantly lower HbA_{1c} concentrations in the high-protein group, (as per above). There was no effect on weight loss or lipids.[52]

Building Muscle, Sports Recovery and Performance

Resistance training simultaneously stimulates catabolism and anabolism in active muscle fibres. The difference between these mechanisms is net protein balance (NPB). When NPB is positive, anabolism outweighs catabolism, and there is an increase in muscle mass. Protein consumption following resistance training potentiates anabolic effects, and is required to provide amino acid precursors necessary to support exercise-induced increases to muscle protein synthesis, positive NPB, and subsequent increased muscle mass, *as represented in Figure 3*.[53,54]

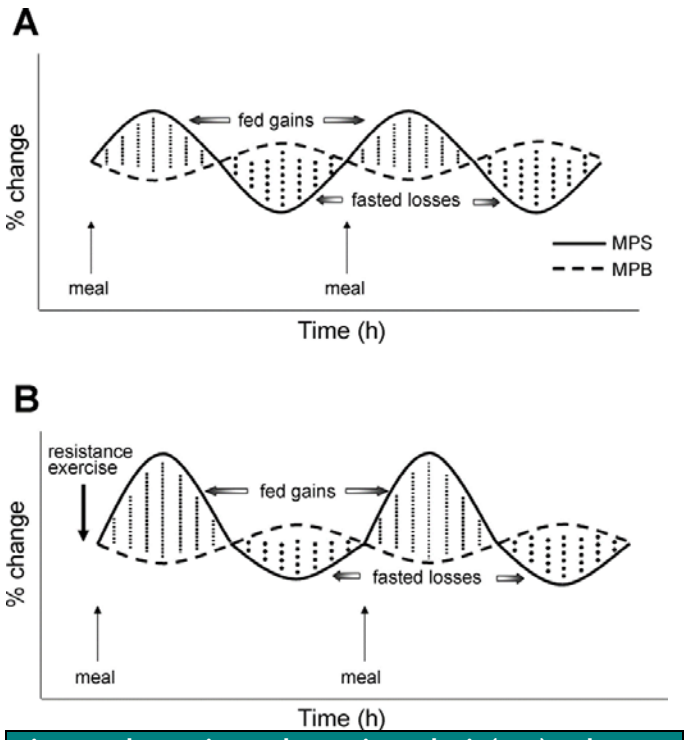

Figure 3. Changes in muscle protein synthesis (MPS) and muscle protein breakdown (MPB) in response to amino acid feeding.[53]

Note: Image B: Changes in MPS and MPB in response to resistance exercise and feeding. Chronic application of these anabolic stimuli results in muscle hypertrophy.
Abbreviations: MPS: muscle protein synthesis; MPB: muscle protein breakdown

A 2015 systematic review of 32 studies evaluated the effects of protein supplements alone, or in combination with carbohydrate, on exercise performance, metrics of body composition, or measures of aerobic or anaerobic power. The review found **protein supplementation may enhance muscle mass and performance** when the training stimulus is adequate (e.g. frequency, volume, duration), and dietary intake is consistent with recommendations for physically active individuals. Enhanced performance was reported as enhanced gains to muscle strength, muscle hypertrophy as well as increases to aerobic and anaerobic power. Several weeks of protein supplementation were reported to be required for benefits to be seen, particularly in untrained individuals.[55]

A second 2015 systematic review investigated combined protein-carbohydrate supplementation during exercise, and the effects on endurance performance. The review concluded protein intake during exercise demonstrated an ergogenic effect on endurance performance when assessed by time to exhaustion. Twenty studies were evaluated. Results were inconsistent, with 13 studies finding significant improvements with combined protein-carbohydrate supplementation compared to carbohydrate only or placebo. Better outcomes were reported when the caloric content of sports drinks was increased by adding protein rather than reducing carbohydrate content to match the amount of calories. Methodological differences may account for the inconsistent results.[56]

Conflicting results on the effects of protein-carbohydrate supplements on endurance performance have continued with a number of other studies.[57,58] One review reported that at least three studies demonstrated carbohydrate-protein ingestion improves endurance performance to a greater extent than carbohydrate alone, whilst two studies showed no significant benefit.[57] The varied outcomes of these studies may be attributed to the form and quantity administered, inclusion criteria for studies selected for reviews, trained or untrained status of the subjects, as well as methodological differences including exercise protocols, duration of supplementation, and metrics measured.[58]

A randomised, double-blind, crossover trial demonstrated 10 g WP containing 4.2 g EAA (in combination with carbohydrate) was a sufficient dose to induce MPS after resistance exercise in trained young men.[59] However, protein and leucine stimulate MPS to potentially induce benefits in sports recovery and performance in a dose-dependent manner. Doses reported for maximum stimulation are higher, and once a maximum stimulation point is reached, MPS plateaus.[47,53]

A study conducted in 2009 investigated the dose-response of leucine for muscle protein synthesis in six young healthy men following resistance exercise. They were administered single doses of 0, 5, 10, 20 and 40 g of egg protein after an intense bout of leg-based resistance exercise. Maximum MPS was seen with 1.7 g leucine (from 20 g egg protein). No significant difference in MPS was seen between 1.7 g leucine and the higher, 3.4 g dose (from 40 g egg protein).[60]

A randomised, double-blind trial with 24 resistance trained males investigated the effect of 48 g of rice protein (RP) or WP (isocaloric and isonitrogenous) on training days, immediately after exercise. Volunteers trained 3 days per week for 8 weeks as a part of a daily resistance-training program. The 48 g dose provided 3.84 g and 5.52 g of leucine for RP and WP respectively. Muscle thickness, weight lifted, and perceived recovery, soreness and readiness to train were recorded at baseline and weeks 4 and 8. Both WP and RP induced significant increases in lean body mass, muscle mass, strength and power and decreases in fat mass at 4 and 8 weeks, with no significant differences between the two groups. The study did not investigate dose-response, yet benefits from protein in resistance-trained males are evident and similar for both doses of leucine.[47]

A double-blind, randomised, clinical trial by Reidy and colleagues concluded that a fast and slow protein blend (PB, as soy and WP) ingested following exercise is capable of prolonging blood aminoacidaemia, mTORC1 signalling, and protein synthesis in human skeletal muscle and is an effective post-exercise nutritional supplement. PB resulted in prolonged positive net protein balance compared to WP alone. Nineteen young adults were studied before and for a 5-hour period after high-intensity exercise. Volunteers ingested 19 g PB or 18 g WP, consumed 1 hour post exercise. Compared to baseline, positive net phenylalanine balance during post exercise recovery was found at 20, 40 min and 20, 40, 60, 80, 100, 120 minutes for WP and PB respectively ($p < 0.05$). Amino acid transport into the cell and transporter expression was similar for both groups.[54] Other slow proteins combined with WP are likely to exert similar effects.

Selected Nutrients

Magnesium

Magnesium is involved in over 300 enzymatic reactions that cover a wide range of metabolic activities including energy production, neuronal activity, cardiac excitability, synthesis of essential molecules, ion transport across cell membranes, cell signalling and cell migration.[61,62,63]

A cross-sectional study found a significant, independent and strong relation between serum magnesium concentration and muscle performance in 1138 older persons.[64]

A 2006 review notes marginal magnesium deficiency impairs exercise performance in athletes, and magnesium supplementation or increased dietary intake of magnesium may have beneficial effects on exercise performance in magnesium-deficient individuals.[65]

Serum and dietary magnesium is inversely associated with the risk of developing cardiovascular disease and T2D (dietary only).[66,67]

A 2006 review of the potential of magnesium in T2D reported a beneficial effect on glucose levels and insulin sensitivity in non-diabetic individuals such as those who were overweight or insulin resistant. Randomised, controlled trials in T2D individuals yielded inconsistent results regarding effect on glycaemic control.[67]

Absorption rates differ significantly between various magnesium salts, and may affect required dose, and efficacy. Magnesium citrate has demonstrated superior bioavailability, high mineral content, purity, solubility and organoleptic properties, when compared to salts such as magnesium oxide.[68,69]

Zinc

Zinc is an essential trace mineral with divergent function, attributable to its status as a component of metalloenzymes from every class.[14] Zinc is found in high protein foods, complexed with amino acids,[14] and plays an essential role in proper muscle growth, metabolic processes, immune function, wound healing, energy substrate utilisation and antioxidant defences.[14,61]

The role of zinc in tissue and muscle growth is related primarily to its function in regulating protein synthesis. Zinc is essential for carboxypeptidases A and B, enzymes necessary for protein absorption in the duodenum; enzyme activity is reportedly decreased in zinc deficiency.[14]

Zinc is involved with the synthesis, storage and secretion of insulin and therefore influences carbohydrate metabolism.[14,70,71]

Matrix metalloproteinases are zinc dependant enzymes which function in wound healing, degrading components of the extracellular matrix to allow for remodelling of extracellular matrix proteins and tissue repair.[14]

Chromium, Iodine, Selenium

Chromium has a role in blood glucose regulation and is thought to potentiate the action of insulin, probably by facilitating the binding of insulin to its cellular receptor, by enhancing insulin-dependent functions, or both.[72,73]

As with other minerals, the rate of chromium excretion during periods of physical exertion are also increased, suggesting that maintaining optimal chromium status is difficult if dietary intakes fail to keep up with increased demand.

Iodine and selenium are essential trace minerals necessary for thyroid hormones synthesis.[74] The effects of thyroid hormones on metabolism are many and varied, and include stimulating metabolic rate, enhancing muscle contraction and enhancing lipolysis. Selenium also has activity in antioxidant defences and immune function.[14]

Cholecalciferol, Biotin, Nicotinamide

Vitamin D has various biological functions, including neuromuscular maintenance, regulating cellular growth, differentiation, proliferation; and modulating immune function. Additionally, vitamin D appears to enhance the secretion and action of insulin.[74]

Biotin is a B vitamin that functions as a cofactor for enzymes involved in carboxylation reactions. Biotin is important for energy metabolism by its role inducing enzymes involved in gluconeogenesis, fatty acid synthesis, amino acid metabolism and glucose utilisation.[14,74]

Vitamin B3 as nicotinamide adenine dinucleotide (NAD) plays a role in energy production via its function in the electron transport chain and therefore synthesis of adenosine triphosphate (ATP).[14]

Chia, Flax, Fibre

Chia seed (*Salvia hispanica*) and flax seed (*Linum usitatissimum*) are well-known sources of high quality nutrients, and traditionally played an important role in the diet of pre-Hispanic Mexicans, and ancient Egyptians respectively. The nutritional value of the seeds of chia and flax are largely attributed to their protein, fibre, omega-3 fatty acids (including alpha-linolenic acid), antioxidant and mineral content.[75,76,77]

The role of fibre in human health includes benefits on risk factors for cardiovascular disease, weight management, immune function and colonic health.[78] Epidemiological and clinical studies demonstrate that intake of dietary fibre is inversely related to obesity, type 2 diabetes, cancer and cardiovascular disease.[79] Adequate intake of dietary fibre for Australians is 25 g and 30 g for women and men respectively.[80]

Pectin is a soluble polysaccharide, soluble fibre and prebiotic which dissolves in water forming viscous gels. Pectin bypasses enzymatic digestion of the small intestine and is easily fermented by colonic microflora, which is likely responsible for pectin's ability to stimulate certain lactobacillus and bifidobacterium species *in vitro*.[79]

Medium Chain Triglycerides

Digestion of medium chain triglycerides (MCTs) does not require bile salts or re-esterification by intestinal cells, and MCTs are absorbed directly into the portal circulation. In contrast, long chain triglycerides (LCTs) are incorporated into chylomicrons following absorption into the intestinal cells and transported through the lymphatic system into the blood stream where lipases hydrolyse LCTs into smaller molecules. Compared to LCTs, MCTs are metabolised faster by the body, have lower calories and have less storage in adipose tissue.[81]

MCT is commonly derived from coconut oil and palm kernel oil, with coconut oil being the richer source.[82] MCT has beneficial organoleptic properties including neutral flavour, reduced rancidity and enhanced palatability.[83]

Safety

Proteins

Caution is advised for individuals with existing renal conditions and young children. In populations with established renal disease, it has been shown that limiting protein to the RDI may slow the progression of disease.[51] Elderly subjects with severe renal impairment (estimated glomerular filtration rate < 30 mL/min/1.73 m^2) who are not on dialysis must limit their protein intake.[35]

Egg protein is contraindicated in individuals with known allergy to egg. WP is contraindicated in individuals with known allergy to milk. Phenylalanine is contraindicated in individuals who suffer from phenylketonuria.

Gynostemma

Animal studies and information from China indicates that Gynostemma herb and total gypenoside extracts have low toxicity. In teratogenicity tests, there were no observable effects on mice embryos when the parent mice were fed with gypenosides at levels of 20% and 50% of LD_{50}. The offspring showed normal development and reproductive ability. No carcinogenicity or mutagenicity was observed in other testing for the gypenosides.[6]

A Thai trial investigated the safety of three doses of concentrated extract of Gynostemma in healthy volunteers. After 8 weeks of treatment, no major adverse events were reported. Laboratory results indicated no toxic effects on the liver, kidney or haematology (including platelet count). Blood glucose was unchanged or remained within normal limits.[84] The daily doses of the extract corresponded to about 0.4, 1.6 and 3.2 g/day of dried aerial parts.[84,85] However, it is known that herb harvested in Thailand has lower amounts of gypenosides compared to herb grown in China.[86]

No adverse effects on liver, kidney, haematology or vital signs were found in obese volunteers, treated with extract corresponding to 3 g/day of dried leaf.[9]

A small number of patients experienced mild adverse effects (vomiting, diarrhoea/constipation, abdominal tension, dizziness, blurred vision, tinnitus) in a large, uncontrolled trial conducted in China. Patients were treated with 2.5–3 g of Gynostemma powder, prepared as tablets or capsules, and administered 3 times a day for 10 days.[6]

Supportive Formulation

Combining proteins absorbed at differing rates may enhance postprandial kinetics. Fast proteins have been shown to induce significantly greater stimulation of muscle protein synthesis, whereas slow proteins inhibit postprandial muscle catabolism. Combining slow and fast proteins has been reported to prolong and enhance MPS following exercise.

Protein, Gynostemma and nutrients complement each other to support the following actions and systems:

- muscle health and strength;
- metabolic processes and normal glucose metabolism;
- immune cells and processes;
- satiety.

Indications

- Muscle wasting conditions including sarcopaenia and cachexia.
- Metabolic syndrome including type 2 diabetes, and possibly as adjunctive therapy for hyperlipidaemia.
- Maintaining healthy weight.
- Physically active individuals, training for sport events and athletes.
- Malnourishment, convalescence.
- Wound healing: postsurgical, accidents and injury.

Author: Abbie Harris.
Gynostemma information was provided by Michelle Morgan

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