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Proteins: Mechanism of action, physical exercise and athlete's hypocaloric state based on literature review

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Abstract. Athletes and physically active people take nutritional supplements to improve their health, maintain their physical qualities and optimise the recovery process. At the same time, the specifics of their use and the mechanism of action in acute energy deficiency have not yet been definitively investigated, which makes the study of the effect of proteins and amino acids in the hypocaloric state of a person an urgent problem. The purpose of the study was to clarify the role and mechanism of the plastic and regulatory effect of proteins and amino acids with a branched chain on the body of athletes in hypocaloric conditions. The analysis of the literature was carried out using the methods of analysis, synthesis, systematisation and generalisation according to the method of using keywords in the electronic databases PubMed and SPORTDiscus. The peculiarities of the athlete's hypoenergetic condition and dietary protein supplements to preserve the athlete's muscle mass in the conditions of long-term physical tests have been clarified. It was found that with a limited level of calories and a low supply of fat, the need for protein increases almost twice. Taking protein within 2 hours after training or before going to bed stimulates a significant acceleration of muscle protein biosynthesis. The amino acid lysine in the composition of proteins activates the central regulator of anabolism of skeletal muscles – the intracellular multimolecular signalling target of rapamycin complex. Its regulatory signals are propagated by the cyclase system. As a result of the action of the appropriate protein kinase, reactions of phosphorylation of chromatin proteins, participants in processing, splicing, etc. occur. It is assumed that in this way lysine stimulates the rate of synthesis of various regulators and factors of anabolism, remodelling and growth. A hypothetical mechanism of the influence of leucine on the anabolic processes of muscle tissue is proposed in a graphic version. The study will be useful for specialists in biochemistry, sports nutrition, sports and wellness industries

Keywords: protein; lysine; training; target of rapamycin; energy deficit

Introduction

Long-term hypocaloric diets have certain disadvantages because they cause loss of fat and lean mass. A negative balance between them leads to muscle exhaustion, reduces the effectiveness of physical exercises and increases the risk of injuries. At the same time, low body weight is an important success factor in many sports, including gymnastics, martial arts, and figure skating. Athletes can use effective nutritional strategies to reduce body weight, which contribute to the preservation of muscle mass and athletic performance. In this context, increased protein intake prevents muscle

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wasting and supports physical performance during periods of energy restriction. Production and sales of sports nutrition and supplements are growing in the European Union. Athletes may sometimes consume them indiscriminately or choose products that have not been evaluated and approved by scientific evidence. In this sense, information about the interaction of the body, physical activity and nutrition contributes to making informed decisions about sports nutrition.

The International Society of Sports Nutrition presented its position paper on the effects of essential amino acid supplementation on skeletal muscle maintenance and performance. This position was published in the work of A.A. Ferrando *et al.* (2023). Several key provisions are highlighted from the published findings. Essential amino acids play the main role in stimulating the synthesis of muscle protein. Protein is critical for replacing degraded or damaged muscle proteins and increasing functional performance. With age and with energy deficiency, the proportion of leucine must be increased to overcome reduced muscle sensitivity. In the study of M. Holeček (2021) the emphasis is on skeletal muscles, which play a dominant role in the catabolism of branched-chain amino acids (BCAAs; valine, leucine, and isoleucine). The main role of skeletal muscles in the catabolism of these amino acids, according to the author, is determined by their mass and high activity of BCAA-aminotransferase, which is absent in the liver. A decrease in the level of amino acids in liver cirrhosis and an increase in their concentration in diabetes have been shown. The authors of this study considered it expedient to clarify the role of changes in BCAA metabolism in other pathologies. C. Tinline-Goodfellow *et al.* (2023) presented a concise description of the mechanisms underlying the target of rapamycin 1 complex (mTOR) mediated activation of muscle protein synthesis. It is recommended that future research be directed to further study mTORC1 activation at the periphery of skeletal muscle fibres. A group of researchers S. Allard *et al.* (2024) have shown that in mammals, mTORC1 controls cellular metabolism in response to energy availability. Transmitting mTORC1 signalling affects the regulation of energy intake, storage, and utilisation. In the authors' opinion, elucidating the mechanism of transmission of mTORC1 signals from one cell and organ to another can be a key factor for the development of safe methods of using proteins in sports practice and medicine.

V.V. Kuibida *et al.* (2022a; 2022b) considered the role of heat shock proteins in adaptation to physical load and growth of bone tissue, which is important for creating optimal methods of the training process and recovery. Viewing metabolism through the prism of the biology of physical activity has proven to be an accessible and practical strategy for gaining new insights into local and systemic regulation of metabolism. Recent methodological developments of J.A.B. Smith *et al.* (2023) improved understanding of the central role of skeletal muscle and nutrition in promoting health. Studying the signalling role of proteins and some amino acids is a new and promising direction of scientific research. The scientific literature is dominated by works devoted to some biochemical, physiological, medical and sports problems of the use of proteins in sports practice. However, it is quite rare to find works in which the mentioned aspects are comprehensively combined with the mechanisms of adaptation of the body in conditions of energy deficiency. This direction of research is relevant and can bring significant results in the theoretical and practical plane. Therefore, the purpose of the article was to find out the plastic and regulatory features of the action of proteins and amino acids under conditions of physical exertion and energy deficit and the methods of their use.

The purpose was achieved through the consistent use of traditional scientific theoretical methods: analysis, synthesis, systematisation and generalisation of data from special literature and Internet sources within the framework of the selected problem. The literature search was carried out on the basis of the above keywords in the PubMed and SPORTDiscus electronic databases. Using the analysis method, the research object, in particular: proteins, branched-chain amino acids, lysine, physical activity, target of rapamycin, energy deficit of the body, mechanism of action, was analysed and broken down into separate component parts. These components were studied in detail, without taking into account their interrelationships. The above-mentioned abstract elements are combined in concrete integrity and interaction by the method of synthesis. As a result of the conducted analysis and synthesis of essential properties, characteristics, features of sports dietary practice and metabolic reactions, the prerequisites for applying the method of systematisation were created. Consequently, this review material is presented in the form of two components or subdivisions: the role of proteins and methods of their use in the nutrition of athletes and physically active people; and the athlete's hypocaloric state and the mechanism of action of branched-chain amino acids. By the final method of generalisation, general signs, properties, trends are recorded and the transition from facts to certain regularities and conclusions is made.

Proteins in the Nutrition of Athletes and Physically Active People

The effectiveness of sports activities depends on the effectiveness of the training process and nutrition strategies, which form the basis of the multi-vector process of adaptation to physical activity. The building and regulatory function of amino acids with a branched chain, primarily leucine, is one of the aspects of the mechanism of adaptation to physical exertion. Current evidence suggests that dietary protein intake is necessary to support metabolic adaptation, repair, remodelling, and protein turnover. In traditional methodological approaches, a protein dose of 1.2 to 2.0 g/kg/day is recommended. Use of a higher dose may be recommended for short periods during intense exercise or when energy intake is reduced.

A significant number of athletes in various sports receive at least one musculoskeletal injury during their sports career. Most often, injuries are recorded during competition periods. A balanced protein diet is of great importance for the recovery and rehabilitation of athletes and physically active people. Eating for recovery after sports injuries has a lot in common with sports nutrition. However, there are differences regarding prevention of the risk or presence of sarcopenia, hypotrophy, or dysphagia. Increased protein consumption with an even distribution of food throughout the day helps to minimise the loss of muscle and strength during immobilisation. Dietary supplement strategies can be helpful in addressing issues related to appropriate caloric intake, timing, and decreased appetite. Rehabilitation nutrition, combined with training, leads to an improvement in the physical condition and acceleration of the return of athletes to the training process after surgery or injury.

The post-traumatic recovery of an athlete often leads to a long period of muscle shutdown, which leads to a loss of muscle mass. As a result of the lack of physical activity, the metabolic health and functional capacity of the athlete deteriorates. The most intense loss of muscle mass occurs during the first two weeks of not using the muscles. The degree of loss of post-traumatic muscle mass determines the level and duration of the necessary rehabilitation. Atrophy of muscle failure is associated with a decrease in the basal rate of protein synthesis and the development of anabolic resistance to food consumption. In this situation, the protein content of the food is crucial for stimulating the rate of its synthesis in the muscles. Increasing your daily intake of protein/amino acids of a specific type at specific times can limit the loss of muscle mass and strength.

Skeletal muscle mass is one of the indicators of human longevity and health. Human muscle mass is determined by the balance of two cellular processes – the synthesis of muscle protein and its breakdown. The balance between the processes of anabolism and catabolism is sensitive to the characteristics of physical activity and protein nutrition. Understanding the molecular mechanisms underlying this process shortens the way to the informed use of nutritional technology in sports and in overcoming age-related anabolic resistance of skeletal muscle. Knowledge of the molecular mechanisms of protein and amino acid effects facilitates the delineation of recommendations for future research directions that may advance knowledge in this field.

In 2023, the use of nutritional supplements by athletes was among the most significant problems. A group of researchers led by C.C. Carey *et al.* (2023) found that among 405 participants in the experiment, 52% take nutritional supplements to improve health, and 35% to maintain their physical qualities or recovery. A ranking of the processes and qualities of athletes and active people, which are significantly dependent on sports nutrition, has been compiled: more effective muscle recovery; increasing endurance; increasing strength and power. Except for performance in the peak power range, a protein intake of 1.6 g/kg/day appears to be sufficient to maximise gains in lean mass, muscle strength, performance, and aerobic capacity as when consuming 1.6 or 3.2 g/kg/day for 16 weeks. This daily amount of protein is effective and safely tolerated by young, healthy adults. R. Bagheri *et al.* (2023) recommended daily protein intake in excess of recommended doses to maximise metabolic adaptation to exercise.

In the study of E.R. Goldstein *et al.* (2023) after training, master class athletes consumed one of three beverages during a 2-hour recovery period: placebo (electrolytes and water), carbohydrates (1.2 g/kg), or a mixture of carbohydrates and proteins (0.8 g/kg + 0.4 g/kg). It has been established that the use of carbohydrates and a carbohydrate-protein mixture is equally effective in enhancing glycogen resynthesis during the 2-hour recovery period. Water and electrolytes alone were not effective in restoring endurance during the second bout of strenuous exercise. J. Zhang *et al.* (2023) studied 427 young people (217 women) aged 21 to 35 years who did 240 min of aerobic and resistance training per week. Strength training was found to be positively associated with intake of protein, vitamins B2, B3, B5, B6, and B12, and percent of total calories from protein, and negatively associated with percent of total calories from carbohydrate. Time spent in aerobic exercise was positively associated with fibre, pectin, and vitamin B6 intake and negatively associated with percentage of total calories from protein. It is assumed that it is worth paying attention to the negative relationship between physical exercises and carbohydrate consumption.

Milk proteins contain essential amino acids (with a sufficient amount of leucine), 80% casein and 20% whey protein. Whey protein belongs to the “fast” proteins, and casein – to the “slow” proteins. In an 8-week experiment by H. Yapici *et al.* (2023), 8 exercises/training were performed with a volume of 3 sets of 8 repetitions and a rest interval between them of 120 s. Two experimental groups participated in the experiment: first group performed combined strength training + consumption of 500 ml of chocolate milk (30 g of protein), second group – strength training without consumption of chocolate milk. The participants of the first group had higher indicators of muscle thickness, lean body mass, maximum strength, length of the jump against the movement, and peak power than the representatives of the second group. The authors concluded that chocolate milk should be considered as a promising post-workout nutritional supplement. M. Ummels *et al.* (2023) evaluated the nutritional value and absorption kinetics of processed barley/rice proteins extracted from brewer’s waste grain and compared them with pea proteins. F. Li *et al.* (2021) reported that isolated soy protein supplementation (40 g/day, 5 days per week, 8 weeks) combined with aerobic exercise (8 weeks at an intensity of 40-65% heart rate reserve) improved body composition, anthropometric characteristics, and cardiovascular pulmonary endurance in women.

The traditional dietary intake of protein for adults is 0.8-0.9 g/kg/day body weight. These guidelines differ from the authoritative recommendations of the Academy of Nutrition and Dietetics, Dietitians of Canada, and the American College of Sports Medicine of 1.2-2.0 g/kg/day. Athletes training for endurance and strength are recommended to consume 1.2-1.7 g/kg/day. Population protein requirements are still based on nitrogen balance. However, this approach is quite debatable. Achieving a balanced intake and excretion of nitrogen is not essential for athletes, for whom the main goal is the successful performance of physical exercises. Nutritional strategies are necessary, for example, for people with type 1 diabetes to control glycemia after exercise. F.R. Muntis *et al.* (2023) monitored glucose levels in 112 adolescents aged 14.5 years and 36.6% overweight or obese. Intensive insulin therapy led to an increase in body weight. Adolescents were recommended to participate in 60 min/day of physical activity of medium and intense power.

C. Ruiz-Castellano *et al.* (2021) provided recommendations for diet, nutrition, and nutritional supplementation for the weight loss phase of strength-trained athletes. Protein intake (2.2-3.0 g/kg/day) should be spread throughout the day (3-6 meals) to ensure sufficient protein in each meal (0.40-0.55 g/kg body weight/food) and eat within 2-3 hours before and after training. Military personnel experience long periods of energy deficits during combat and training, making them susceptible to harmful weight loss. Research results of R.E. Anderson *et al.* (2023) support the recommendation that more than 0.8 g/kg/day is needed to moderate the effect of energy deficit on lean body mass reduction, whereas intakes of up to 1.6 g/kg/day may be better. However, precise recommendations cannot be made because the severity and duration of energy deficits varies between studies. Longer and larger studies are needed to elucidate the role of protein during energy deficits in the military.

Consumption of compounds rich in amino acids improves collagen content in tendons and their biomechanical properties. N.W.C. Campbell *et al.* (2023) found: if an oral bolus of essential amino acids is not combined with exercise, it leads to only a moderate increase in the concentration of amino acids in the peritendinous Achilles tendon; in combination with exercise with a gap in the consumption of essential amino acids resulted in a higher concentration of amino acids in the tendons compared to no exercise.

D. Mercer *et al.* (2020) calculated the average need for protein consumption for female athletes in the period before menopause. It is close to the needs of those who train in the mode of aerobic endurance (1.28-1.63 g/kg/day), engage in resistance exercises (1.49 g/kg/day) and periodic physical exertion (1.41 g/kg/day) lasting ~60-90 min. Pre- and post-workout protein intake of 0.32-0.38 g/kg has been shown to be beneficial for female athletes performing resistance exercise and periodic exercise. However, in further studies, researchers need to identify the influence of the phase of the menstrual cycle or the use of hormonal contraceptives on the need for protein.

Nutrition has a significant impact on injury prevention and recovery due to its impact on the athlete's overall physical and psychological well-being and improved tissue healing. In particular, amino acid and protein intake, antioxidants, creatine, and omega-3 have been studied for their role in preventing muscle loss and anabolic resistance and healing injuries. An adequate nutritional strategy plays an important role in recovery and rehabilitation after injury, and nutritional interventions should become part of standard practice in post-injury care (Smith-Ryan *et al.*, 2020).

A group of researchers led by P. Rodriguez-Lopez *et al.* (2022) analysed and screened commercial protein supplements for sports practice and identified the types of tested protein products. Among them, whey (milk) is a protein of high biological value with a high concentration of branched amino acids, including leucine. There are three types of protein: concentrated (with 70-80% protein and a small amount of carbohydrates and fats); isolated (with 90% protein and a minimum amount of carbohydrates and fats); hydrolysed (a protein in which long polypeptide chains are broken into short peptides, due to which their absorption occurs faster). Casein (milk) is a protein of high biological value, which makes up 80% of milk protein. Due to the acidic environment of the stomach, it forms blood clots that slow down its absorption. Egg albumin is a protein of high biological value without fat or carbohydrates. Soy protein has a high biological value, its digestion is fast. It is usually low in leucine unless it is additionally fortified. Other plant proteins have lower biological value unless multiple sources are mixed or fortified.

Protein supplements have certain features and characteristics, in particular, a sufficient amount of branched chain amino acids (BCAA – leucine, isoleucine, valine) and high biological value, digestibility and bioavailability. They correspond to the classification of the Australian Institute of Sport as isolated protein supplements from a certain source: whey, casein, egg albumin, plant sources (soy, hemp, peas, chickpeas, rice, etc.). There are no special regulations for protein supplements that regulate the production and marketing of this type of sports nutrition. Supplement usage patterns for different purposes, dosages, and regimens do not always align with the protein that works best. In addition, supplements may contain substances that are classified as doping according to World Anti-Doping Agency. They contain substances that are not prohibited, but reduce their quality. In particular, this is furosine, which appears in whey proteins during their preparation at high temperatures due to the Maillard reaction. R.J. Maughan *et al.* (2018) and P. Rodriguez-Lopez *et al.* (2022) noted that in some places supplements may contain enzyme and allergen impurities and ingredients that lead to absorption or doping problems. The content of BCAA in the blood plasma can signal certain diseases. Significant changes in the content of combinations of amino acids leucine/isoleucine/valine, etc. were revealed in blood plasma of patients with coronary heart disease with atrial fibrillation.

Hypocaloric State of the Athlete and the Mechanism of Action of Amino Acids with a Branched Chain

It is advisable to consider in more detail the hypocaloric condition and the mechanism of action of amino acids. In cases of energy restriction or sudden inactivity resulting from injury, there is a need for increased protein intake to 2.0 g/kg/day or higher (Burke *et al.*, 2023). T.J. O'Leary *et al.* (2023) found that short periods of severe energy deprivation and heavy exercise (in the form of 36-hour military field exercises) can suppress bone formation for at least 96 hours. Inhibition of bone formation in men and women did not differ. Protein feeding did not compensate for decreased bone formation during severe energy deficits.

In the studies of C. Hiroux *et al.* (2023) exogenous ketosis did not affect body composition but was effective in weight control, maintained declines in physical capacity, and improved general stress parameters. Protein requirements increased when athletes became calorie restricted or low fat. In 2014, the team led by E.R. Helms *et al.* (2014) studied the effect of dietary protein on the body composition of energy-restricted athletes subjected to strength training. It was concluded that the need for protein for them is 2.3-3.1 g/kg, taking into account the limitation of caloric content and leanness.

Targeted weight loss in obese older adults is a risk factor for accelerated muscle loss. In the experiment A.M. Verreijen *et al.* (2017) were involved 100 overweight and obese adults aged 55-80. The effect of a high-protein diet and/or physical exercise on the preservation of fat-free mass during weight loss was studied. During the 10-week weight loss programme, all subjects followed a hypocaloric diet and performed strength exercises 3 times a week: the first group consumed food with a high (1.3 g/kg) and the second with a normal (0.8 g/kg) protein content. It was concluded that in the group with the combined use of a high-protein diet and strength exercises, the index of fat-free body mass increased significantly more.

Consuming high doses of protein during energy deficits combined with intense exercise results in effective muscle gain and fat loss. In the experiment of T.M. Longland *et al.* (2016) used hypoenergetic diets (~ 40% of requirement) that provided 33 ± 1 kcal/kg. Young men of group I consumed a small amount (1.2 g/kg/day), and men of group II consumed an increased amount (2.4 g/kg/day) of protein. All participants of the experiment performed resistance exercises in combination with high-intensity interval training for 4 weeks 6 times a week. It was concluded that during marked energy deficit, intake of 2.4 g protein kg/day was more effective than intake of 1.2 g protein kg/day in lean body mass gain and fat loss in combination with high volume resistance and anaerobic exercises.

Energy deficiency leads to a decrease in the formation of bone tissue, increased resorption, or a combination of them. Physiological adaptation to low energy availability includes changes in the concentration of regulatory hormones: a decrease in triiodothyronine (T3), insulin-like growth factor-1, leptin, and testosterone. M. Papageorgiou *et al.* (2017) recorded changes in the ovulatory cycles of women due to inhibition of gonadotropin-releasing hormone, reduction of pulsation of luteinising hormone and concentration of estrogens. The effect of a short-term low energy deficit at 15 kcal/kg on bone metabolism in women and men was studied. It has been established that with a lack of energy, the process of bone formation slows down and the resorption of bone tissue increases only in women. These metabolic changes are accompanied by a decrease in the response to insulin and leptin.

The results of studies by J. Kwon *et al.* (2023) expanded the understanding of the role and mechanism of action of leucine. It has been shown that dietary protein is necessary for the body's recovery process and its adaptation after training. Previous reports suggest that many athletes meet their total daily protein requirement at dinner. In contrast, an even distribution of protein doses ≥0.24 g/kg body weight consumed throughout the day provides rapid rates of anabolism in skeletal muscle compared to a skewed intake pattern. Leucine is considered the main driver of the anabolic reaction after eating, and its content determines the quality of the protein. Adaptation to speed-force load occurs by stimulating the activity of the protein synthetic mechanism. Increasing the biosynthesis of muscle proteins is an adaptive response to an increase in the concentration of leucine and exogenous amino acids. The above-mentioned amino acid is not synthesised in the human body. It participates in the process of building and restoring muscle mass. It is part of a complex of three amino acids (leucine, isoleucine, valine) for sports nutrition and is designated as BCAA (branched-chain amino acids). The most leucine is in meat, dairy products, fish, soybeans, beans and other legumes. With a lack of amino acids in the body, growth slows down, muscle tissue breaks down, and body weight decreases. The main part of food amino acids is metabolised in the liver. At the same time, according to the research of Y. Huang *et al.* (2011) amino acids with branched side chains undergo catabolic transformations in skeletal muscle, heart, neurons, adipose tissue, and kidney. They ensure the stability of the tertiary structure of proteins.

Leucine, isoleucine and valine are amino acids with hydrophobic properties. Hydrophobic repulsion during interaction with hydrogen and ionic bonds and Van der Waals forces ensure the processes of formation and stabilisation of the tertiary structure of the protein. Leucine is involved in the stabilisation of the tertiary structure of proteins, the binding of O_2 to myoglobin and haemoglobin, and the interaction of enzymes with substrates. It accelerates anabolic reactions, leads to increased muscle mass and strengthens bones, ligaments and tendons. The above-mentioned amino acid accelerates the process of wound healing and recovery after injuries. Leucine provides backup energy in the event of a lack of glucose and leads to an increase in endurance and strength. Before use, it is diluted in a bottle with drinking water in the amount of 2.5-5 g and used before, during or after training and washed down with water. Leucine regulates the rate of muscle protein synthesis by activating the mTOR complex (mammalian target of rapamycin) and possibly other proteins of this regulatory pathway.

The dose of leucine that is inside the protein (the leucine threshold hypothesis) and its subsequent variables in the plasma (the leucine trigger hypothesis; peak magnitude, rate of rise, and total availability) determine the magnitude of the response of muscle protein synthesis after exercise and eating. K. Wilkinson *et al.* (2023) found a pronounced correlation between the dose of leucine and muscle protein synthesis after exercise in the elderly. On the other hand, the correlation was less expressed in young people.

The manipulation of cellular signals is a powerful factor in the therapy of diseases and the implementation of a strategy of influence on the motor system. Amino acids with a branched chain enter the blood from the gastrointestinal tract faster than others. When the concentration of leucine and necessary hormones increases, the synthesis of proteins in the muscles is accelerated. Leucine activates a special anabolic receptor – mTOR, which signals a sufficient amount of building materials for the construction of new proteins. The function of the above protein receptor belongs to serine/threonine protein kinases (mechanistic target of rapamycin kinase). Due to the effects of phosphorylation, leucine stimulates the rate of synthesis of growth hormone, which strengthens the mechanostat of bone tissue, accelerates the formation of muscle mass, cartilage and other connective structures. The mTOR receptor is part of two multimeric protein complexes that differ in activators, targets, and sensitivity to rapamycin. The first complex – TORC1 (target of rapamycin complex 1) stimulates cell growth by accelerating ribosome biogenesis and translation, slowing protein degradation, and activating the uptake of nutrients from the environment. The rapamycin complex 1 (mTORC1) complex includes: anabolic receptor – mTOR and proteins raptor (regulatory-associated protein of TOR), mLST8 (mammalian lethal with Sec13 protein 8), or GβL and PRAS40 (proline-rich PKB (protein kinase B)/AKT substrate 40 kDa).

mTORC1 is considered to be the main regulator of muscle protein synthesis. The amino acid lysine activates the mTORC1 complex through the enzymes of the cyclic nucleotide system, in particular – GTPases (a type of G-proteins that belong to the Ras superfamily), which leads to the relocalisation of the complex. Upon activation of the mTORC1 complex, muscle protein biosynthesis is enhanced due to the phosphorylation of key regulators of mRNA translation p70 kinase of ribosomal protein S6 (S6K1). Instead, rapamycin inhibits mTORC1, blocks cell proliferation, and suppresses the immune response to transplanted organs. As a result of physical exertion, branched-chain amino acids or other anabolic stimuli, this complex moves to the periphery of the cell. Leucine has the greatest anabolic effect among amino acids due to its ability to independently modulate muscle protein synthesis. Phosphorylation of RPS6 Ser240/244 (phospho-S6 ribosomal protein) occurs in human muscle fibres, where mTOR is translocated after anabolic stimuli. Focal adhesion complexes (point contacts) are considered a potential site of mTORC1 regulation *in vivo*. There are two types of adhesive contacts: cell-to-cell and cell-to-matrix. Translocation and accumulation of receptors on the cell membrane leads to binding of the cell to the extracellular matrix through the actin cytoskeleton and anabolic effects (Hodson *et al*., 2022). Food components can cause epigenetic changes through DNA methylation, or through changes in the expression of some miRNAs. Carbohydrates, proteins, and fats are broken down during digestion into monomeric compositions, respectively, into monosaccharides, amino acids, and fatty acids (Sohel, 2020; Norouzi *et al*., 2023).

Pathways of regulation of metabolic processes may involve common signalling links, for example, mTORC1. S. Abou Sawan *et al*. (2020) stated that skeletal muscles are characterised by high plasticity. Muscle tissue is capable of remodelling in response to a number of physiological stimuli. Historically, the lysosome has been considered a catabolic organelle that facilitates autophagy, phagocytosis, and exo-/endocytosis. However, evidence has recently emerged for several anabolic roles of lysosomes in the maintenance of skeletal muscle mass. They have been implicated in the mechanistic target of mTORC1 intracellular signalling centre and EB/lysosomal biogenesis transcription factor signalling in the mTORC1-mediated regulation of protein synthesis. It has been suggested that the lysosome is an understudied organelle that has the potential to support the adaptive response of skeletal muscle to anabolic stimuli. Regulation of autophagy protein expression rate in response to chronic contractile activity in postmitotic cells depends on lysosomes. A. Parousis *et al*. (2018) found that chronic contractile activity enhances mitochondrial biogenesis, as well as the ability of lysosomes to improve mitochondrial turnover. The hypothetical mechanism of the effect of leucine on increasing muscle mass is presented in a graphic version (Fig. 1).

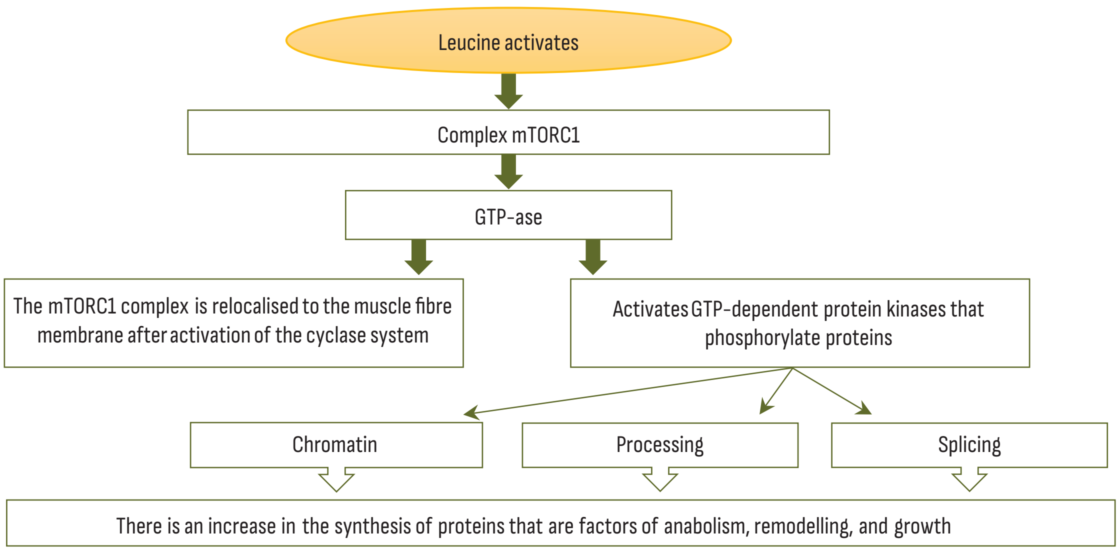

Figure 1. Hypothetical mechanism of action of leucine and anabolic processes of muscle tissue
Source: created by the authors

Leucine plays a plastic and signalling role in the body; it is considered the main driver of the anabolic reaction. The content of leucine and some amino acids with a branched chain and hydrophobic properties determine the nutritional value of the protein. The introduced hydrophobic repulsion of the amino acid complements the forming force of hydrogen bonds, ionic bonds and Van der Waals forces, which ensure the processes of formation and stabilisation of the tertiary structure of the protein. Lysine activates the central regulator of skeletal muscle anabolism – the mTORC1 complex, which consists of mTOR, Raptor, mLST8, PRAS40 and DEPTOR proteins. The regulatory signal is propagated with the participation of enzymes of the cyclic nucleotide system, in particular – GTPases. This complex is relocalised to the muscle fibre membrane. The protein receptor (mechanical target of rapamycin mTOR) belongs to serine/threonine protein kinases. As a result of the reaction of phosphorylation of chromatin proteins, participants in processing, splicing, etc., the amino acid stimulates the rate of synthesis of various regulators and factors of anabolism, remodelling, and growth. In particular, the production of growth hormone accelerates, which strengthens the mechanostat of bone tissue, accelerates the formation of muscle mass, cartilage and other connective structures.

In the course of the study, the following conclusions can be reached: the synthesis of muscle protein is maximally enhanced after the consumption of 10 g of essential amino acids in free form or in the form of a protein bolus in a dose of 20-40 g. The total daily rate of replenishment of the body with high-quality protein (~0.75-1.2 g/kg body weight) should be evenly divided into three portions (~ 0.25-0.40 g/kg body weight) and consumed every 3-4 hours during the day. The specified scheme of protein consumption is the most effective compared to other models of nutrition. Taking high-quality protein sources after exercise within 2 hours after exercise or consuming casein protein (~30-40 g) before bed stimulates a significant increase in muscle protein synthesis and does not affect lipolysis. If there is no physical exercise, then changing the frequency of meals has a rather limited effect on weight loss and body composition.

Conclusions

The need for protein increases when athletes are on a calorie-restricted or low-fat diet. For them, the need for protein can be 2.3-3.1 g/kg, taking into account the restriction of caloric content and leanness. Consuming high doses of protein during energy deficits combined with intense exercise results in effective muscle gain and body fat loss. Increased protein intake prevents muscle wasting and supports physical performance during periods of energy restriction. Pre- and/or post-exercise dietary interventions (carbohydrate + protein or protein alone) may act as an effective strategy to support strength gains and improve body composition. However, the size and timing of the pre-exercise meal can affect the degree of post-exercise protein requirement. Among commercial protein supplements for sports practice, some proven protein products of high biological value are selected: whey (milk), casein (milk), egg albumin, and soy protein. It is most effective to consume protein during the day by dividing it into 3 meals, which qualitatively affects the composition of the body compared to other types of food. Without exercise, changing the frequency of meals has little effect on body weight and composition. The use of protein after training or before casein at bedtime stimulates significant synthesis of muscle protein without affecting lipolysis.

Further research should examine the effect of combining an exercise programme with altered meal frequency on weight loss and body composition. It is worth making a comparative analysis between common regulatory blocks of the signalling mechanism of action of branched-chain amino acids and myokines, osteokines, hepatokines, adipokines, microRNAs during physical activity of different intensity, duration and during the recovery period. Useful information may be gained by the broader use of [^{14}C]-leucine and other labelled amino acids to investigate mTORC1-lysosomal repositioning and intracellular utilisation of dietary amino acids in response to anabolic stimuli. How lysosomal regulation may contribute to skeletal muscle anabolism needs to be elucidated.

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Conflict of Interest

None.

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Білки: механізм дії, фізичне навантаження та гіпокалорійний стан спортсмена за даними огляду літератури

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Анотація. Спортсмени та фізично активні люди приймають харчові добавки для покращення здоров'я, підтримки своїх фізичних якостей та оптимізації процесу відновлення. Водночас особливості їх використання та механізм дії при гострому дефіциті енергії і досі остаточно не досліджені, що робить вивчення впливу білків та амінокислот при гіпокалорійному стані людини актуальною проблемою. Метою дослідження було з'ясувати роль і механізм пластичного й регуляторного впливу білків та амінокислот із розгалуженим ланцюгом на організм спортсменів у гіпокалорійних умовах. Аналіз літератури здійснено за допомогою методів аналізу, синтезу, систематизації та узагальнення за методикою використання ключових слів в електронних базах даних «PubMed» та «SPORTDiscus». З'ясовано особливості гіпоенергетичного стану спортсмена та дієтичних білкових добавок для збереження м'язової маси спортсмена в умовах тривалих фізичних випробувань. Виявлено, що при обмеженому рівні калорій та низькому запасі жиру потреба в білку зростає майже вдвічі. Прийом протеїну впродовж 2 годин після тренування або перед сном стимулюють значне прискорення біосинтезу м'язового білка. Амінокислота лізин у складі протеїнів здійснює активацію центрального регулятора анаболізму скелетних м'язів – внутрішньоклітинного мультимолекулярного сигнального комплексу – мішені рапаміцину. Його регуляторні сигнали поширюються циклазною системою. Унаслідок дії відповідної протеїнкінази відбуваються реакції фосфорилювання білків хроматину, учасників процесингу, сплайсингу тощо. Зроблено припущення, що таким шляхом лізин стимулює швидкість синтезу різноманітних регуляторів та факторів анаболізму, ремодуляції та росту. Запропоновано гіпотетичний механізм впливу лейцину на анаболічні процеси м'язової тканини в графічному варіанті. Дослідження буде корисним фахівцям із біохімії, спортивної дієтології, спортивної та оздоровчої галузей

Ключові слова: протеїн; лізин; тренування; мішень рапаміцину; енергодефіцит