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Implications of Exercise Training and Distribution of Protein Intake on Molecular Processes Regulating Skeletal Muscle Plasticity

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Abstract

To optimize its function, skeletal muscle exhibits exceptional plasticity and possesses the fundamental capacity to adapt its metabolic and contractile properties in response to various external stimuli (e.g., external loading, nutrient availability, and humoral factors). The adaptability of skeletal muscle, along with its relatively large mass and high metabolic rate, makes this tissue an important contributor to whole body health and mobility. This adaptational process includes changes in the number, size, and structural/functional properties of the myofibers. The adaptations of skeletal muscle to exercise are highly interrelated with dietary intake, particularly dietary protein, which has been shown to further potentiate exercise training-induced adaptations. Understanding the molecular adaptation of skeletal muscle to exercise and protein consumption is vital to elicit maximum benefit from exercise training to improve human performance and health. In this review, we will provide an overview of the molecular pathways regulating skeletal muscle adaptation to exercise and protein, and discuss the role of subsequent timing of nutrient intake following exercise.

Keywords

Skeletal muscle; Exercise adaptation; Mitochondrial biogenesis; Muscle protein turnover; Dietary protein; Cellular signaling

Introduction

Skeletal muscle is a highly malleable tissue, capable of adapting to external stimuli to match structural, functional, and metabolic demands [1–3]. Exercise training, both endurance and resistance, have been well documented to stimulate alterations in muscle fiber type, size, and increase mitochondrial activity and content [2–4]. Training-induced adaptations in skeletal muscle are highly specific to endurance or resistance exercise, with further augmentation

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Human and Animal Rights and Informed Consent To the best of author's knowledge, all procedures performed in the cited studies involving human participants in this review were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards and all applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

based on the volume and frequency of training sessions [5]. Phenotypic adaptations to exercise arise from alterations in cellular mechanisms, which modify gene expression and intracellular signaling proteins [6].

Alterations in cellular mechanisms regulating training-induced adaptations are not only responsive to exercise bouts, but to nutrient intake as well [7, 8]. Specifically, dietary protein is a potent anabolic stimulator which when coupled with a bout of resistance exercise increases muscle protein synthesis to a greater extent than resistance exercise alone, over time resulting in enhanced muscle hypertrophy [9]. Furthermore, new evidence is now emerging that dietary protein may potentiate endurance exercise training adaptations through alteration in mitochondrial biogenesis [10, 11]. The interrelationship of exercise and dietary protein has led to much research focused on the optimization of nutrient timing following exercise to further potentiate long-term training adaptations. However, many of these investigations have focused solely on provision of dietary protein immediately post exercise (e.g., <5 h post exercise). As skeletal muscle has been reported to have an increased sensitivity to dietary protein for up to 24–48 h following a single bout of exercise, daily distribution of protein consumption may have an important function in enhancing long-term training adaptations [12, 13].

This review will provide an overview of the current understanding of molecular pathways regulating skeletal muscle adaptation to exercise training and dietary protein intake. Furthermore, this review will explore the role of optimized daily distribution of dietary protein intake and potential regulatory mechanisms.

Molecular Pathways Regulating Muscle Adaptation

Skeletal muscle is essentially made up of a mixture of relatively different multinucleated cells that are highly heterogeneous in fiber type (e.g., glycolytic vs. oxidative, highly fatigable vs. fatigue resistant, etc.) which contributes to its plasticity. These factors allow the ease of muscle's adaptation of its structure and function in response to different anabolic/catabolic stimuli such as, contractile activity (aerobic/resistance exercise, sedentary lifestyle, overload and unloading), growth factors and nutrient availability. For the purpose of this review, we will separate the discussion of skeletal muscle adaptation to molecular pathways regulating mitochondrial biogenesis, fiber type composition, and hypertrophy/atrophy (Fig. 1).

Energy-Sensing Pathways Modulating Mitochondrial Biogenesis and Fiber Type Distribution

The molecular events influencing the process of exercise and nutrient-induced adaptation of skeletal muscle likely begin with major disruptions to cellular homeostasis that occurs during exercise and changes in nutrient availability [14, 15]. Major modifications in energy homeostasis occur as a result of skeletal muscle contraction during exercise that contributes to its adaptation. Energy-sensing pathways that are regulated by the hydrolysis of ATP ([ATP/ADP], [Pi]), the redox of mitochondrial nicotinamide adenine dinucleotide (NAD⁺/NADH), and the release of calcium ion (Ca²⁺) from the sarcoplasmic reticulum (SR) activating the metabolic sensors, 5AMP-activated protein kinase (AMPK), NAD-dependent

deacetylase sirtuin-1 (SIRT1), and Ca^{2+} /calmodulin-dependent protein kinase II (CAMKII), respectively. Ultimately, these intracellular alterations result in the upregulation peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1 α), a master regulator of mitochondrial biogenesis, resulting in increased mitochondria size, content, and activity (Fig. 1).

Skeletal muscle displays remarkable changes in mitochondrial oxidative capacity as an adaptive response to contractile activity and nutrient availability [16–19]. Endurance exercise training induces increases in the size, number, and oxidative capacity of mitochondria [20, 21]. A single bout of exercise is a sufficient stimulus to induce mitochondrial biogenesis, with this mitochondrial proliferation being maintained by repeated bouts of exercise [20, 22]. The transcriptional coactivator PGC-1 α has been demonstrated to have a critical role in the induction of mitochondrial biogenesis, as overexpression of PGC-1 α in muscle cells and transgenic mice results in increased mitochondria content through increased expression of mitochondrial DNA by mitochondrial transcription factor (TFAM) [14]. The overexpression of PGC-1 α in myotubes induces mitochondrial biogenesis and increases the expression of genes involved in oxidative phosphorylation (e.g., COXI and IV), mirroring an exercise-like effect [23]. Furthermore, overexpressed PGC-1 α transgenic mice were observed to have a transition in skeletal muscle with their white muscle (e.g., glycolytic) having a more red muscle phenotype (e.g., oxidative) [23]. Both a single bout and chronic endurance exercise training are known to increase gene and protein expression of PGC-1 α in the skeletal muscle of both humans and rats [24–29]. Recently, studies have questioned if PGC-1 α is required for exercise-induced adaptive responses in skeletal muscle [29, 30]. Although there are some questions about the extent of PGC-1 α 's role in exercise training-induced adaptations of skeletal muscle, the importance of this protein for the determination of fiber type composition is well recognized [23, 29, 31, 32].

Upstream of PGC-1 α are several regulator proteins that respond to alterations in the intracellular milieu which upregulates their activity, resulting in stimulation of PGC-1 α . Muscle contraction and exercise are observed to change AMP/ATP and NAD^+/NADH levels thus activating AMPK and SIRT, respectively [33, 34]. The activation of AMPK can bring about a significant adaptation of skeletal muscle by regulating the expression of specific genes involved in energy metabolism [35] and the inhibition of energy consuming processes [36]. AMPK activation through daily AICAR (an AMP analog and potent activator of AMPK) injections (1 mg/g) for 4 weeks was associated with increased expression of GLUT4, hexokinase II (HKII), and mitochondrial proteins (i.e., citrate synthase, cytochrome C) in rodent skeletal muscle [37, 38]. Furthermore, AMPK recently has been described to directly regulate both expression and activity of PGC-1 α [34, 39]. In cultured myocytes, SIRT1 appears to be necessary for the cell-autonomous switch from glucose utilization to fatty acid oxidation; an important metabolic shift for endurance athletes to maintain blood glucose concentrations and energy availability for prolonged training sessions [40]. This altered metabolic regulation is associated with the activation of PGC-1 α , mediated through deacetylation by SIRT1 [40]. Following a single bout of endurance exercise, there is an upregulation in SIRT1 activity [41], with total SIRT1 protein content elevated with sustained endurance exercise training [42].

It is well established that with exercise there is an increased flux of Ca^{2+} into the intracellular space, which is essential for the contraction-relaxation cycle of skeletal muscle [43, 44]. However, more recently the importance of Ca^{2+} signaling in regulating activity-dependent muscle gene expression and muscle fiber type heterogeneity has been acknowledged [44, 45]. Wright et al. [45], reported calcium-induced mitochondrial biogenesis was dependent on the CAMK phosphorylation of *p38* MAPK which regulated the activation and expression of PGC-1 α . Furthermore, CAMK upregulates PGC-1 α expression through activation of the transcription factors cAMP response element-binding protein (CREB) and myocyte enhancer factor 2 (MEF2) [46, 47]. Activation of CREB and MEF2 allows for binding of these transcription factors to the promoter region of PGC-1 α to upregulate transcription and initiate mitochondrial biogenesis.

Exercise induces the conversion of muscle fibers from the more glycolytic type 2x (humans/rodents) and/or 2b (rodents) (white, glycolytic, fast twitch) to the more oxidative type 2a (white, oxidative, medium twitch) that has a more type 1 phenotype (red, oxidative, slow twitch) [48–51]. Type 1 fibers are characterized by an increased mitochondrial number and oxidative state as well as a specific set of contractile proteins such as troponin I slow and myoglobin [50, 52]. The ability to “shift” skeletal muscle fibers from a more glycolytic fiber to a more oxidative fiber is critical for energy availability to support skeletal muscle during endurance exercise.

Intracellular Pathways Modulating of Muscle Protein Synthesis and Breakdown

The ability for resistance exercise to stimulate hypertrophy (e.g., increased muscle fiber size) is a result of daily balance between muscle protein synthesis (MPS) and muscle protein breakdown (MPB) [53]. When individuals are in a net positive balance, with MPS exceeding MPB, hypertrophic gains can be achieved. Regulation of skeletal muscle protein turnover is a highly integrated network involving gene transcription, translation, post-translational modifications, and proteolysis [54, 55]. The most well-described mechanism by which dietary protein and exercise modulates skeletal MPS is the mechanistic target of rapamycin complex 1 (mTORC1) signaling pathway [55, 56]. This intracellular signaling cascade influenced by energy status, growth factors (e.g., insulin, growth hormone), contractile activity, and nutrient availability regulates MPS by modifying mRNA translation initiation and elongation. Activation of mTORC1 triggers downstream signaling through p70 ribosomal S6 kinase (p70 S6K1), ribosomal protein S6 (rpS6), eukaryotic elongation factor 2 kinase (eEF2), and eukaryotic initiation factor 4E-binding protein (4E-BP1), which increases mRNA translational efficiency and ultimately MPS (Fig. 1).

Recently, it has been reported that the RhoA GTPase is associated with the adaptation of skeletal muscle via multiple signal transduction pathways in skeletal muscle through its downstream targets, Rho-Associated Kinase (ROCK), Striated Muscle Activator of Rho Signaling (STARS), and Serum Response Factor (SRF) (Fig. 1). RhoA plays key roles in the regulation of diverse myocellular functions, including cell motility, cell growth, and gene transcription. Recent work has shown increased contractile activity (functional overload, exercise) and inactivity (disuse) of skeletal muscle alters RhoA expression [57–59].

Furthermore, RhoA targets ROCK, STARS, and SRF are shown to be required for crucial events such as myocyte fusion and differentiation, hypertrophy, and atrophy [60].

The ubiquitin proteasome system is one of the major regulators of MPB [61] (Fig. 1). The resulting myofibrillar proteolysis from ubiquitination is through the muscle-specific E3 class of ubiquitin ligases, atrogin-1/muscle atrophy F-box (MAFbx), and muscle RING finger-1 (MuRF1) [62]. Activity of atrogin-1/MAFbx and MuRF1 are regulated by the forkhead box O (FOXO) family of transcription factors, which when dephosphorylated translocate to the nucleus to mediate increased expression of these ubiquitin ligases. The ubiquitinated proteins are transferred to the 26S proteasome for subsequent degradation [63]. Anabolic and energy sensitive processes have been reported to alter the ubiquitin proteasome pathway [64]. Specifically, upregulation of anabolic stimulators, such as Akt, an upstream mediator of mTORC1 activity, have been reported to inactivate FOXO through phosphorylation and thus inhibits proteolysis [65]. Contrarily, stimulation of energy-sensing pathways, such as AMPK may potentiate the ubiquitin proteasome pathway through inhibition of anabolism and stimulation of catabolism to liberate energy stores [66, 67].

Influence of Dietary Protein on Muscle Protein Turnover

Dietary protein is a potent anabolic stimulus which when consumed results in transient increases in MPS [68]. When dietary protein is consumed following resistance exercise, MPS is elevated to a greater extent than with resistance exercise alone [69]. Over time, the rise in MPS may enhance skeletal muscle accretion [70, 71]. The stimulation of MPS to dietary protein is primarily driven by exogenous essential amino acids (EAA) [72, 73], especially the branched-chain amino acid (BCAA) leucine [74], with little contribution from non-essential amino acids (NEAA) [75]. Elevations in MPS following dietary protein intake is a dose-dependent and saturable process, with maximal stimulation being achieved with ingestion of 10 g of EAA, 20 g of an isolated high-quality protein source, or 30 g of protein as part of a mixed meal [76–80]. Consumption of exogenous EAA increases extracellular and intracellular concentration of EAAs [80], with increased availability of EAA in the muscle upregulating the mTORC1 intracellular signaling cascade [55]. In contrast, it does not appear that EAA stimulate activation of the RhoA/STARS pathway to enhance muscle hypertrophy. Vissing et al. [59] reported significant changes to Rho signaling immediately after resistance exercise, with no further effect observed with whey protein supplementation. These data suggest that EAA modulate MPS through mTORC1, while RhoA/STARS regulation is contraction dependent.

It is believed that the response in MPS to exercise and feeding is the primary factor dictating long-term resistance exercise training adaptations in skeletal muscle [81]. However, due to the difficult nature and potential unreliability of measurements of MPB utilizing stable isotope methodologies in response to feeding and exercise [82], the role of MPB is not well defined. Investigations assessing acute alterations in molecular regulation of MPB in response to exercise and protein intake have produced conflicting results [83–87]. Borgevnik et al. [83] reported that supplementation of BCAA by young, normal weight men and women, reduced expression of atrogin-1 and MuRF1 in response to resistance exercise, while others [84–86] have found no difference in markers of proteolysis with consumption of protein following

resistance exercise. When assessing change in muscle hypertrophy following a 12-week resistance training program with or without protein supplementation, Stefanetti et al. [86] reported that while habitual protein supplementation increased quadriceps muscle cross-sectional area to a greater extent than carbohydrate supplementation in young, normal weight men, there was no negative association with a reduction in intracellular markers of proteolysis. While these findings are consistent with the theory that alterations in MPB play a minor role in skeletal muscle adaptations to exercise and protein supplementation, the investigators did not report any markers of MPS. The absence of data on MPS makes it difficult to determine if difference in hypertrophy was driven by difference in MPS between the supplement groups.

Influence of Dietary Protein on Mitochondrial Biogenesis

While dietary protein has primarily been linked to muscle hypertrophy, recent publications have demonstrated that protein intake may have a role in augmenting mitochondrial biogenesis. In cell culture and animal models, provision of leucine has been reported to stimulate SIRT1 expression in skeletal muscle [88–90]. Furthermore, investigations in young, health men, assessing dietary protein intake in combination with endurance exercise have reported elevations in mixed MPS [91, 92]. As endurance exercise does not result in muscle hypertrophy, the rise in mixed MPS may be a result of elevations in mitochondria synthesis, potentially mediated by the increased SIRT1 activity induced by increased intracellular leucine concentrations. To date, the limited number of human trials assessing mitochondrial synthesis with stable isotope methodologies has resulted in conflicting findings when volunteers consumed dietary protein in combination with endurance exercise [93–98]. These discordant results may be due to discrepancies in analytical methodologies, as well as timing of analysis following exercise. The majority of these interventions have assessed mitochondrial synthesis within the first 3 h following exercise. However, in one investigation, which assessed PGC-1 α mRNA expression 6 h post endurance exercise, concluded that protein supplementation with endurance exercise enhanced markers of mitochondrial biogenesis to a greater extent than carbohydrate supplementation [98]. While there is insufficient evidence at this time to conclude the effect protein supplementation on mitochondrial biogenesis, this finding indicates that investigators should extend their assessment window, potentially measuring mitochondrial synthesis and markers of mitochondrial biogenesis up to 24 h following a bout of endurance exercise.

Distribution of Dietary Protein Intake

It has recently been reported that acute elevations in MPS with exercise and protein intake do not correlate with long-term adaptations to training [99, 100]. This finding may be due to a suboptimal distribution of dietary protein intake throughout the day following a bout of exercise. The elevation in MPS after resistance exercise with EAA results from increased sensitivity to EAA-induced MPS, which lasts at least 24 h post exercise [13]. As dietary protein intake by adults in the United States is skewed, with low intakes (<30 g) in the morning and excessively high intakes (>30 g) in the evening [101], consuming optimal amounts (30 g) of protein throughout the day may confer additional benefit to exercise-induced skeletal muscle adaptations. However, studies investigating the influence of dietary protein on MPS following exercise focus primarily on the initial recovery phase (4–6 h) [79,

92, 102, 103]. While these studies offer valuable insight into the relationship between exercise and dietary protein intake, they fail to account for the remaining, larger portion, of an individual's day. As such, to investigate the influence of protein amount and timing following a bout of resistance exercise, Areta et al. [104] fed normal weight, young men 80 g (~40 g EAA) of a whey protein isolate over a 12-h period (Fig. 2A). Protein consumption regimens were 10 g every 1.5 h (8 per 12 h), 20 g every 3 h (4 per 12 h), or 40 g every 6 h (2 per 12 h). Results from this intervention were that over the 12-h feeding period, consuming 20 g protein every 3 h elicited a 31 and 48 % greater response in MPS compared to 10 g every 1.5 h and 40 g every 6 h, respectively. Furthermore, Mamerow et al. [105] measured MPS for 24 h, providing normal weight, middle age men, and women isocaloric/isonitrogenous diets, where protein intake was distributed evenly (breakfast: ~30 g, lunch: ~30 g, dinner: ~30 g) or skewed (breakfast: ~11 g, lunch: ~16 g, dinner: ~63 g; Fig. 2B) throughout the day for 7 days. The major finding of this investigation was that MPS was 25 % greater when protein intake was evenly distributed, compared to skewed. These novel findings indicate a potential-optimized interrelationship between amount and timing of protein consumption to maximally stimulate MPS.

The elevations in MPS seen with evenly distributed protein intake can likely be attributed to the transient nature of the response of MPS to dietary protein. The rise in MPS in response to exogenous EAAs is rapid, reaching maximal stimulation between 60 and 90 min and returning to postabsorptive values within 180 min [106, 107]. The return to basal MPS may occur despite extracellular and intracellular EAA concentrations remaining persistently elevated above basal concentrations [79, 106, 107]. This phenomenon is known as the “muscle full” hypothesis [108]. This hypothesis suggests that there is an upper limit to EAA delivery into the muscle, where once this point is reached there is a reduction in uptake, and EAA in the muscle are no longer utilized for MPS, but rather diverted to oxidation [107]. The “muscle full” hypothesis may partially be explained by amino acid transporter expression in response to EAA availability. Amino acid transporters are classified as solute-linked carrier (SLC) family members, are ubiquitously expressed in the plasma membrane of many cells, and typically coupled with counter-transporters, such as Na^+/K^+ ATPase, to maintain the ion concentration gradient [109–111] (Fig. 1). Investigators have focus specifically on L-type amino acid transport (LAT1), which couples with a glycoprotein (CD98), and sodium-coupled neutral amino acid transporter (SNAT2). These transporters work cooperatively, with SNAT2 mediating the uptake of small neutral amino acids, in particular glutamine, which enables the bitransporter LAT1/CD98 to export glutamine to increase the influx of leucine [112]. Increased expression of LAT1/CD98 and SNAT2 has been positively correlated with mTORC1 activation [113, 114]. Drummond et al. [115] demonstrated that 60 min following the consumption of 10 g of EAA, LAT1/CD98 and SNAT2 expression were significantly elevated compared to fasting conditions in young, normal weight men and women. Along with the increased transport of leucine into skeletal muscle, there was a 2.4-fold increase in MPS [115]. However, by 120 min leucine influx declined significantly, and transporter expression and MPS returned to basal levels, despite intracellular leucine concentrations remaining significantly elevated above fasted values for 180 min [115]. These data support the “muscle full” hypothesis, indicating a potential negative feedback mechanism where the mTORC1 signaling cascade is saturated, leading to

a downregulation in transport expression to reduce influx of EAA and a shift in nutrient partitioning where EAA are preferentially oxidized rather than utilized for MPS.

The emerging complex mechanisms involved in the cellular regulation of MPS may also contribute to the need for appropriate amount and timing of protein intake. Recently, EAA have been shown to act on several signaling proteins upstream of mTORC1 to induce elevations in MPS [116] (Fig. 1). Rag GTPases are a family of four Ras-related small guanosine triphosphatases (RagA, RagB, RagC, RagD), which have been suggested to mediate EAA-induced mTORC1 activation, through formation of a heterodimer which binds with the mTORC1-associated protein Raptor and triggers relocation of mTORC1 to the lysosome or late endosomal membrane [117–119]. It is suggested that EAA facilitate activation of this process through promoting the active configuration of the Rag GTPase heterodimer by stimulating vacuolar H⁺-ATPase (v-ATPase) which controls binding of Regulator to the Rag GTPase complex [116]. Regulator tethers the Rag GTPase heterodimer to the lysosome, as well as activates the complex enabling mTORC1 binding [120]. Relocation of mTORC1 increases its proximity with its activator Rheb to promote signal transduction [118]. Given the complexity of these intracellular mechanisms that regulate MPS and the requirement for translocation and binding of mTORC1, there may be a rate-limiting step necessitating the need for appropriate dosing and distribution of dietary protein throughout the day to maximally stimulate MPS. Furthermore, the translocation of mTORC1 may act as a potential negative feedback mechanism, where once it relocates to the lysosome it initiates a downregulation in EAA uptake and diverts EAA to oxidative processes.

Conclusion

Cellular processes regulating skeletal muscle adaptation to exercise training are complex, resulting in transcriptional, translational, and post-translational modifications. Provision of dietary protein can potentiate these intracellular mechanisms, resulting in enhanced adaptations to exercise training. While the role of acute dietary protein intake on MPS is well defined, the role of dietary protein on MPB and mitochondrial biogenesis remains largely undefined. Furthermore, limited information is available on the influence of equivalent distribution of optimal protein amounts on modulation of exercise training adaptations. It is important to note that the majority of the human trials discussed in this review paper were performed in untrained or recreationally active normal weight, young, healthy individuals consuming eucaloric diets. As molecular processes regulating skeletal muscle plasticity may be impacted by training status, age, weight, and energy balance generalizability of this information may be limited. Future investigations examining the interrelationship between exercise and protein amount and distribution should focus on long-term adaptations to MPS, MPB, and mitochondrial biogenesis in various study populations. Elucidation molecular adaptation of skeletal muscle to exercise and protein consumption is vital to provoke maximum benefit from exercise training to improve human performance and health.

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Fig. 1.

Schematic overview of molecular pathways regulating skeletal muscle adaptation. Anabolic stimuli such as, exercise (aerobic and resistance) and nutrient availability (carbohydrate [CHO], fat, and protein) can induce changes in energy status and calcium (Ca^{2+}) release, hormonal status (insulin-like growth factor [IGF-1], mechanogrowth factor [MGF], and growth hormone [GH]), and the transport and utilization of energy substrates (amino acids [AA], glucose [Glc], and fatty acids [FA]). AA, Glc, and FA are transported into the cell through their transporters, solute carrier (SLCs), glucose transporter 4, and fatty acid transporter/CD36 (FAT/CD36). The GTPases Rag/Rheb localize mTOR to the lysosome in an AA-dependent interaction with Ragulator. Contraction-induced energy depletion and Ca^{2+} release lead to the activation of molecular pathways such as, AMP-activated protein kinase (AMPK), NAD-dependent deacetylase sirtuin-1 (SIRT1), and Ca^{2+} /calmodulin-dependent protein kinases (CAMK). Activation of these regulators of cellular energy homeostasis culminates in changes to transcriptional regulators of mitochondrial biogenesis and fiber type transformation, peroxisome proliferator-activated receptor (PPAR)-c coactivator 1 (PGC1 α), cAMP response element-binding protein (CREB), PPAR, myocyte enhancer factor (MEF), mitochondrial transcription factor A (TFAM), and nuclear respiratory factor (NRF). Contraction, hormonal status, and nutrients highly affect growth regulators, Akt, mechanistic target of rapamycin (mTOR), and p70 ribosomal S6 kinase 1 (S6K1) resulting in the disassociation of eukaryotic translation initiation factor 4E (EIF4E)-binding protein 1 (4EBP1) from EIF4E initiating translation and inhibiting of protein degradation-associated proteins Forkhead box (FOXO), muscle ring-finger (MURF1), and

muscle atrophy F-box (MAFbx). Recent data have revealed the contraction-induced regulation of RhoA and its downstream substrates ROCK (Rho-Associated Kinase), Striated Muscle Activator of Rho Signaling (STARS), and Serum Response Factor (SRF) leading to skeletal muscle hypertrophy

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Fig. 2.

Distribution of protein intake. **a** Amount and distribution of dietary protein intake post exercise adapted from Areta et al. [104]. **b** Amount and distribution of the daily dietary protein intake adapted from Mamerow et al. [105]