Adams G

The Molecular Response of Skeletal Muscle to Resistance Training

Die molekulare Antwort der Skelettmuskulatur auf Krafttraining

Department of Physiology and Biophysics, University of California, Irvine, USA

SUMMARY

Skeletal muscle has a high degree of dynamic plasticity involving constant changes in the mix of metabolic, structural and contractile proteins which adapt this tissue to functional demands. Many mechanisms which regulate muscle adaptation are intrinsic to the muscles; i.e., relatively independent of central/ circulating regulatory factors. The aim of deliberate programs of training are the optimized activation of these intrinsic mechanisms. For example, training to improve muscle endurance targets adaptation in cellular sub-systems that regulate energy substrate selection and utilization. Training to improve muscle force targets subsystems that increase myofibrillar protein content. This latter case will be the focus of the current presentation. Cellular changes indicative of a nascent hypertrophy response can be detected within minutes to hours following a single bout resistance exercise. This includes changes in the production and/or accumulation of myogenic messenger RNA as well as increased flux in signaling pathways with known pro-anabolic effects such as the regulation of protein translation. Subsequent training sessions result in the summation of these acute responses leading to functionally relevant cellular adaptation. In addition to the regulation of myogenic mRNA production (transcription) there are regulatory elements that modulate steps between transcription and translation. These include mRNA binding proteins and non-coding RNA (e.g., microRNA) which regulate the abundance and translational activity of specific mRNAs. An additional area of interest in skeletal muscle adaptation has been the role of ancillary cell types such as satellite cells. In the specific case of skeletal muscle hypertrophy, it is clear that a number of the loading sensitive changes in myogenic gene expression are related to the mobilization of these cells. An understanding of the sensitivity and temporal responses of these anabolic regulatory mechanisms will provide practitioners with useful insights on the training stimuli necessary to optimize functional outcomes thereby improving performance.

Key words: Hypertrophy, Translation, mTOR, Satellite Cell

ZUSAMMENFASSUNG

Die dynamische Anpassungsfähigkeit der Skelettmuskulatur umfasst Veränderungen der metabolischen, strukturellen und kontraktilen Proteine, über die sich das Muskelgewebe an die jeweiligen funktionellen Erfordernisse anpasst.
Einige Mechanismen, welche die Muskeladaptation regulieren, sind intrinsischer Art und weitgehend unabhängig von zentralen/zirkulierenden Faktoren.
Das Ziel von spezifischen Trainingsprogrammen ist die optimale Aktivierung dieser intrinsischen Mechanismen. Das Training zur Verbesserung der muskulären Ausdauer zielt beispielsweise auf Anpassungen in den zellulären Subsystemen ab, welche die Verfügbarkeit und Utilisation von Energiesubstraten regulieren. Zelluläre Veränderungen, die auf eine beginnende Hypertrophie hinweisen, sind innerhalb von Minuten bis zu Stunden nach einer Krafttrainingseinheit nachweisbar. Davon betroffen sind sowohl Veränderungen in der Produktion und/oder Akkumulation der myogenen Messenger RNA´s als auch die erhöhte Aktivität in Signalwegen mit pro-anabolen Effekten wie beispielsweise die Regulation der Protein- Translation. Aufeinanderfolgende Trainingseinheiten bewirken die Summation dieser Akutantworten, die wiederum zu funktionell bedeutsamen zellulären Adaptationen führen. Neben der Regulation der myogenen mRNA-Produktion (Translation) gibt es weitere regulative Elemente, welche die Schritte zwischen Transkription und Translation modulieren. Diese umfassen proteinbindende mRNA und nicht kodierende RNA (e.g. microRNA) welche die Menge und Translations-Aktivität der spezifischen mRNA regulieren. Wichtig für die Adaptation des Skelettmuskels sind auch die Muskelvorläuferzellen, sogenannte Satellitenzellen. Im speziellen Fall der Skelettmuskelhypertrophie ist evident, dass bestimmte belastungssensitive Veränderungen in der myogenen Genexpression mit der Aktivierung dieser Zellen verbunden sind. Ein tieferes Verständnis für die Sensitivität und die zeitliche Antwort dieser anabolen regulativen Mechanismen liefert dem Fachmann hilfreiche Erkenntnisse über die erforderlichen Trainingsstimuli, die zur Optimierung funktioneller Outcomes und damit zu einer verbesserten Leistungsfähigkeit führen.

Schlüsselwörter: Hypertrophie, Translation, mTOR, Satellitenzellen

INTRODUCTION

Skeletal muscle is largest single organ system in the human body. It functions in obvious ways, such as locomotion, breathing, and postural maintenance. More recently, less intuitive roles such as endocrine and possibly immune functions have been attributed to this tissue as well (23,30). As a result, the understanding of the totality of cellular and molecular processes within skeletal muscle has been recognized as being remarkably complex and well beyond the scope of a brief review. The literature pertaining to molecular and cellular aspects of muscle adaptation can be very difficult to decipher. In particular, human studies, due to low tissue yields, are generally focused on a very few outcome variables. Review articles can provide a broader perspective. However, the authors of reviews (including the current one) are limited by their own interests and perspectives. The topic of the current review is extensive, easily

sufficient for an entire text book. Accordingly, the current paper will be focused, narrowly, on a limited number of adaptive cellular and molecular regulatory mechanisms related to the adaptation of mature skeletal muscle in response to increased loading such as that encountered in a sports training setting. The mechanisms and processes were selected because, in the authors opinion, they are instructive representatives of how adaptation is regulated and are critical for hypertrophy to occur.

INTRINSIC REGULATION OF HYPERTROPHY

An organizational theme for this brief review derives from a key concept; that the primary mechanisms regulating the adaptation of mature skeletal muscle to increased loading reside within the affected muscle. To illustrate this point: Experimental hypophysectomy drastically reduces, either directly or indirectly, a number of critical circulating hormones and growth factors known to regulate skeletal muscle growth. Key examples of this are thyroid hormone, growth hormone and insulin-like growth factor-1 (IGF-I). When the hypophysectomy procedure is performed in young adult rats it arrests all further body growth. However, when individual muscles or muscle groups experience increased muscle loading in hypophysectomized rats the relative degree of hypertrophy is the same as that seen in control animals (3).

TRAINING CELLS

A second and complementary concept is that any and all exercise training is inherently targeted on the intra- and possibly inter-cellular mechanisms of cells which reside in the targeted muscle.
In the context of functional hypertrophy, effective training must activate the appropriate anabolic regulatory pathways within muscle to a sufficient magnitude and with a temporal pattern that summates to produce sustained responses leading to adaptation. Leaving aside the motor learning and neural components of strength which are outside the scope of this review. Although there are some changes in myofiber phenotype , e.g., glycolytic to oxidative glycolytic shifts, these adaptations represent fine tuning of metabolic parameters and have minimal impact of force generation. Therefore, the primary cellular adaptation leading to increased strength will be an increase in contractile components along with the structure necessary to support and transmit the increased force.

To this end, training parameters such as exercise frequency, intensity and duration are scaled to provide the cellular stimuli necessary to entrain anabolic regulatory mechanisms. Using training frequency as an example, a given bout of weight lifting may elicit a robust response from an anabolic intracellular signaling mechanism (Fig. 1). Repeating that exercise bout several days later may elicit the same level of response (Fig. 2). Repeating the exercise bout 24-36 hours after the first might result in a different response (Fig. 3). In this hypothetical setting, the bouts depicted in Figures 1 & 2 would be unlikely to stimulate adaptation, at least adaptation that would be dependent on the given cellular mechanism depicted here. They would essentially be two independent bouts of exercise. In contrast, the scenario described in Figure 3 suggests a summation of the regulatory response which, if repeated, would effect longer term alterations in the processes

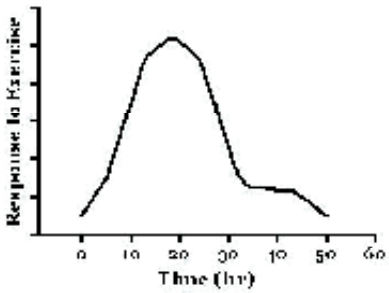

Figure 1: The effects of exercise frequency on hypothetical cellular or molecular response. This curve could describe a number of processes such as an increase in a given mRNA or increased signaling activity in a regulatory pathway. Following resistance exercise (RE), the response increases, reaches a peak then declines with some characteristic pattern.

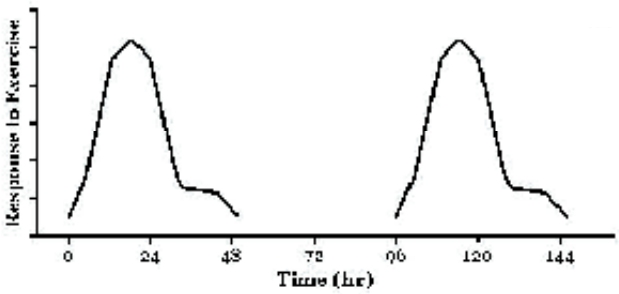

Figure 2: The effects of exercise frequency on hypothetical cellular or molecular response to repeated bouts of resistance exercise. A second bout of training four days after the first results in a similar pattern. The area under these two curves would be the same.

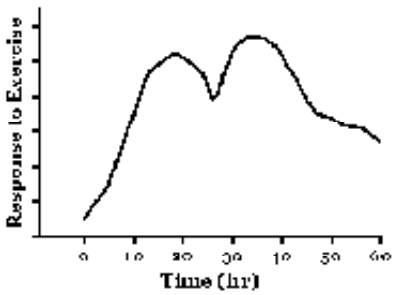

Figure 3: The pattern produced when the second bout of exercise follows more closely to the first. The response may show an increase in magnitude and/or duration. The area under the curve described in Fig. 3 would be much greater than that in Fig. 2. In both humans and animals, changes in mRNA and signaling processes such as phosphorylation of components of signaling cascades have been demonstrated to follow the patterns described in this figure (15, 7).

modulated by that pathway. This would be expected to lead to adaptation. Such processes are implicit in the intent of training programs. In practice, acute cellular and molecular changes indicative of a nascent hypertrophy response can be detected within very short time frames (e.g., minutes – hours) following a single

bout resistance exercise. These responses include rapid changes in the production and/or accumulation of myogenic messenger RNA as well as increased flux in signaling pathways with known pro-anabolic effects highly concentrated in the area of regulation of protein translation. Subsequent training sessions result in the temporal summation of these acute responses such that functionally relevant cellular adaptation will occur leading to increases in muscle size and strength. In a research setting, the temporal responses of anabolic signaling to resistance exercise and their summation have been demonstrated in both animals and humans (15,7).

CRITICAL PROCESSES

This review will focus on two processes considered to be critical for a sustained hypertrophic response to increased loading. The first, protein translation and, the second, activation and incorporation of satellite cells. There is currently a strong consensus for the first, some controversy regarding the second.

REGULATION OF PROTEIN TRANSLATION

There are a myriad of regulatory signaling pathways that have been identified as being relevant to the development of loading induced muscle hypertrophy (24,28,34,37). In the simplest case, loading induced increases in the contractile protein content of skeletal muscle occur via increased production of protein rather than a decrease in protein degradation (25). This is an inherently logical approach since the routine degradation of proteins serves the important purpose of removing less functional proteins, the retention of which would be expected to negatively impact function. Accordingly, pathways that regulate messenger ribonucleic acid (mRNA) translation, i.e., the process of protein synthesis, have received a tremendous amount of attention.

There are three primary components of protein translation that are regulated during the adaptation induced by increased loading; 1. Translation initiation, 2. The availability of substrate, 3. The levels of translational capacity. A comprehensive treatment of the regulation of translation is beyond the scope of this review. However, the salient points follow.

Regulation of Translation Initiation

Several of the key regulatory steps for initiation of translation involve the removal of inhibition. The initiation complex consists of a rather large number of proteins called initiation factors (IF). These initiation factors in many ways fill niches similar to those seen for the regulation of gene expression. Some dock with specific sites on the mRNA and serve to recruit additional initiation factors. Others serve to promote processes such as removing structural impediments in the mRNA which prevent translation. The final result being the recruitment of the ribosomal machinery.

When two of these critical initiation factors, IF3 and IF4E are bound by inhibitory proteins the process of initiation is prevented (Fig. 4). Selective phosphorylation of these inhibitory proteins causes them to dissociate from their target initiation factor and allows for initiation to proceed (Fig. 5) (19). In both cases, a kinase

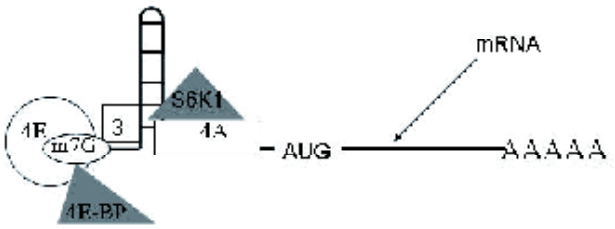

Figure 4: mTOR mediated phosphorylation of inhibitory proteins contributes to the regulation of translation initiation. The initiation complex from on the 5`end of the mRNA. When inhibitory proteins (grey) are bound to initiation factor proteins (IF) translation initiation is arrested.

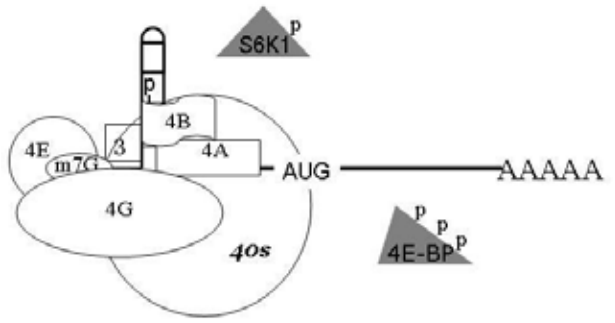

Figure 5: mTOR mediates the phosphorylation of these inhibitory proteins causing them to dissociate from the IF and allowing the remainder of the initiation complex to form. Key; eukaryotic initiation factors : 3, 4A, 4B, 4E, 4G . S6K1: Ribosomal S6 protein kinase. 4E-BP: eukaryotic initiation factor binding protein. AUG: Protein translation start codon. m7G: mRNA cap consisting of a series of methylguanosines added during mRNA processing. Facilitates formation of the initiation complex. 40s: Small Ribosomal subunit

complex which includes the protein mTOR (mammalian target of rapamycin) is responsible for the release of this inhibition (19).

Substrate for Translation

Two classes of substrate are required for translation to proceed; 1 Amino Acids. 2 mRNA. In healthy individuals, availability of amino acids is largely a function of nutrition. The availability of specific mRNA is regulated at two levels; 1 Transcription (and processing). 2 Degradation. There are a number of non- and muscle-specific transcription factors that regulate the production of myogenic mRNAs. The family of myogenic regulatory factors such as MyoD and myogenin are some of the most commonly cited muscle specific transcription factors and have powerful effects on muscle gene expression (6).

The regulation of mRNA stability can be accomplished via the binding of proteins to internal AU rich regions of mRNA (12). The expression and activity of these regulatory proteins has recently begun to be explored in skeletal muscle (13, 40, 42). In pilot studies we have found the levels of mRNA for AU binding proteins such as HuR (Human antigen R) and Tis11B (tristetraprolin family protein) be very sensitive to increased or decreased loading in rodent muscles (unpublished).

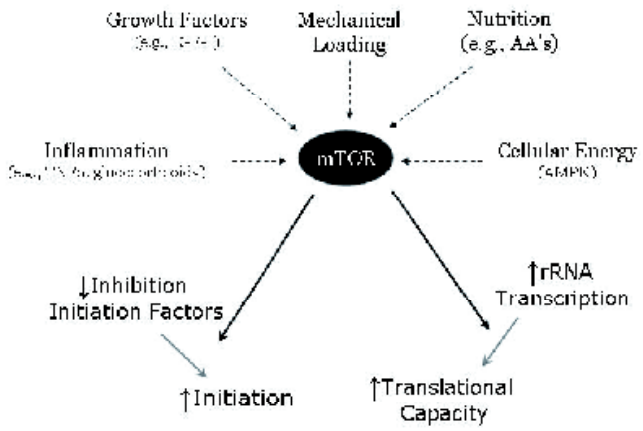

Figure 6: Regulation of mTOR. mTOR activity integrates input from many cell regulatory mechanisms. mTOR is activated by signaling cascades initiated by the ligation of growth factor receptors such as those serving Insulin and IGF-I. mTOR receives direct input from cell membrane mediated processes that are sensitive to mechanical deformation. mTOR activity increases in the presence of certain amino acids. mTOR activity can be inhibited by AMPK when cellular energy systems are being taxed as during endurance mode exercise. mTOR is also targeted by signaling associated by inflammatory mediators leading to decreased protein synthesis in skeletal muscle.

Recent discoveries have shown that, in addition to proteins, there is a class of small non-protein coding RNAs called microRNA (miRNA) can alter the stability of mRNA via binding to complementary sequences in the mRNA (41). In addition to altering stability, miRNA binding can prevent mRNA from completing the translation process.

Regulation of Translational Capacity

Translational capacity is determined by the number of functional ribosomes present in cells. Ribosomes consist primarily of ribosomal RNA (rRNA) and accessory proteins. The most critical step required to increase ribosomal capacity involves up-regulation of the activity of RNA Polymerases (POL) leading to the transcription of ribosomal DNA (rDNA) and ribosomal proteins (27). In this context, mTOR, a key regulator of translation initiation, is also known to play an important role in significantly up regulating RNA Polymerase activity leading to increased production of ribosomal RNA and proteins (27).

While translational efficiency, i.e., protein produced per unit of ribosome, can increase to some extent (8), sustained increased in ribosomal capacity are necessary for a successful, sustained, hypertrophic responses to increased loading (2). Ribosomal RNA (rRNA) makes up the preponderance of total RNA. As a result, changes in total RNA and, presumably, translational capacity can be inferred via the relatively simple process of measuring total RNA content in muscles or muscle samples. Such measurements demonstrate that this response is very sensitive to increased loading. In both rodents and humans we have found that total RNA increases significantly following just two consecutive bouts of resistance exercise (7,16).

mTOR a Critical Regulator of Hypertrophy

These foregoing vignettes have touched upon the critical role of mTOR in the regulation of translation. More globally, mTOR is recognized as a powerful regulator of cell size in many cell types (27). One of the primary roles of mTOR is thought to be sensing whether conditions favor growth (18). In this role, mTOR receives input from a number of signaling cascades that respond to growth factors and hormones, most notably, Insulin and IGF-I (38). Recent data suggests that mechanisms responsive to mechanical loading of muscle cells can also regulate mTOR activity independent growth factor input (17).

In addition to growth factors and mechanical loading, mTOR activity is sensitive to nutritional status, in particular, amino acid availability (21). This role, as a sensor of amino acid availability, may be of particular interest in the context of sport. Published attempts at regulating the availability and timing of nutritional support relative to training bouts may be directly manipulating mTOR activity (21).

Clearly, common sense would suggest that the presence of inflammation would be expected to be detrimental to anabolic processes. Components of the inflammatory response are known to negatively impact mTOR activity (26). This response has had adaptive value in that it conserves both energy and amino acid pools to promote an effective response to injury and infection at time when an organism would generally experience a decrease in its ability to gather or hunt food. In modern humans, assuming the availability of adequate nutrition, this response has lost its adaptive value.

Of particular interest in the setting of sports training are recent reports that endurance mode exercise may also directly, if transiently, regulate mTOR activity. There are reports that the protein AMPK (5' adenosine monophosphate-activated protein kinase) when activated, can down regulate mTOR activity (36). In this context it is important to appreciate that the process of translation is costly in terms of energy. For example, for each amino acid added to the nascent polypeptide approximately four high energy bonds are consumed (9). AMPK activity is sensitive to the energy charge in cells and is activated at times of high energy usage (22). Acutely, the primary role of AMPK appears to be the up regulation of processes that increase energy supply. In light of the high energy cost of protein synthesis, AMPK signaling that leads to decreased mTOR activity appears to be quite logical, deferring the less immediate need for protein production in favor of the need for energy conservation to support contractile activity.

In sum, the regulation of mTOR is critical to processes that contribute to the hypertrophic response and it in turn integrates stimuli from many sources to modulate this response (38) (Fig. 6).

Muscle Satellite Cells and Hypertrophy

In addition to the more obvious need to increase muscle protein, a robust and sustained hypertrophic response appears to require the activation, proliferation, differentiation and fusion of satellite cells. The context for this theoretical framework is rooted in the myonuclear domain- (4) or DNA unit-hypothesis (10). This hypothesis holds that there is a finite relationship between the number of myonuclei and the size of myofibers and that, above some threshold of expansion, the addition of myonuclei is necessary to maintain ongoing hypertrophic processes (4,32). Some investigators have speculated that, under some circumstances, the incorporation of myonuclei into myofibers may precede and drive subsequent hypertrophy (5).

However, some experimental results have been interpreted to indicate that satellite cells are not required to support the hypertrophic response. For example, Kadi et al. observed, in human studies, that moderate levels of muscle hypertrophy can occur in the absence of significant levels of myonuclear incorporation (20). However, the question which must be asked is what degree of muscle hypertrophy is attained across the myofibers in a given study. It is to be expected that the relationship between myofiber size and myonuclear number would have a fairly wide range. Such a design seems logical in that there would be an appreciable metabolic and resource expense associated with the constant activation of satellite cell proliferation in response to moderate fluctuations in muscle loading.

It also seems reasonable to expect that, after a period of rapid satellite cell or myoblast activity (i.e., proliferation, differentiation and fusion) there would be a period during which new myonuclei become operational and contribute to the process of protein synthesis leading to a reestablishment of the myonuclear number to myofiber size ratio and that this would take place in the absence of further cell replication events (29,33).

Are Satellite Cells Required for Hypertrophy?

In rodent muscles we found that incapacitation of satellite cell proliferative activity severely limits the hypertrophic response to a powerful loading stimulus (2). More recently, Petrella et al. reported that, in a large cohort of subjects who participated in a 16 week resistance training study, a sizable proportion of subjects experienced a negligible amount of hypertrophy (non-responders) while responders had increases in myofiber size of ~40% (31). One of the primary characteristics which distinguished these two groups was the ability to add myonuclei to myofibers. This same research group went on to demonstrate that the relative ability to mobilize satellite cells and add myonuclei corresponded to the degree of hypertrophy seen in human subjects (32). Taken together, these results indicate that, above some threshold of myofiber to myonucleus ratio, the ability to add myonuclei via the mobilization of satellite cells is an important contributor to the hypertrophic response.

Activation of Muscle Satellite Cells

Satellite cell participation in the hypertrophic process has been the focus of intense study for a number of years. In that time much has been learned about the regulatory mechanisms within satellite cells (39). Recently, results have been published which suggest a, much sought after, direct link between the mechanical loading of myofibers and the initiating events leading to the activation of satellite cells. For example, Kosek and Bamman reported that resistance training results in changes in the dystrophin-associated protein complex which may provide a regulatory link (24). It is suggested that loading induced changes in nitric oxide synthase (NOS) activity, associated with the dystrophin-associated protein complex, could result in the release of hepatocyte growth factor (HGF) from the extracellular matrix of myofibers allowing it to interact with receptors on satellite cells. HGF has been shown to be critical for the transition from quiescence to activation in satellite cells (35).

SYNTHESIS

This brief review has touched on but a small percentage of the information available regarding cellular and molecular regulation of muscle adaptation. However, it is hoped that the approach used in this review can be used to synthesize new, testable hypotheses. For example, in the study by Petrella at al., discussed above, the non-responders who failed to incorporate new myonuclei did experience a robust level of satellite cell proliferation (31). This suggests that the activation steps such as the production of NO and release of HGF via alterations in the dystrophin-associated protein complex were probably intact in these subjects. The defect would seem to be related to either the differentiation of satellite cell progeny or their incorporation into myofibers. In the cited work by Kosek and Bamman, older subjects experienced similar changes in dystrophin-associated protein complex following resistance training but also failed to demonstrate similar levels of hypertrophy relative to young subjects (24). One of the features that distinguished the old from the young in that study was enhanced activation of stress related kinase p38. This kinase has been associated with muscle atrophy, in part, via the activation of muscle specific ubiquitin-ligases such as MuRF1 (11, 14). One of the key processes down stream of IGF-I signaling is known to be the suppression of muscle specific ubiquitin-ligase expression. This suggests a point of potential interaction between IGF-I and p38 related signaling. More generally, IGF-I has been shown to stimulate the differentiation of satellite cells and promote their subsequent incorporation into existing myofibers (1). A synthesis of the results from these studies suggests that increased p38 signaling in older subjects may have antagonized IGF-I related signaling pathways critical to these secondary and tertiary steps in the process of satellite cell responses to increased loading thereby blunting the hypertrophy response overall. Obviously, this hypothesis requires testing.

SUMMARY

Skeletal muscle hypertrophy is often quantified by as an increase in myofiber cross sectional area. Functionally significant increases in myofiber cross sectional area are a result, primarily, of an increase the amount of contractile protein present in myofibers. This occurs via the process of protein synthesis, that is, mRNA translation.

In this context it is important to remember that the mechanisms which sense changes in loading state and those which generate adaptive responses reside within the cells of the targeted muscles. From this awareness proceeds the understanding that training programs which seek to increase muscle size are manipulating cellular and molecular mechanisms. With regard to understanding specific regulatory mechanisms, an appreciation of the various stimuli and signaling pathways that alter the activity of mTOR can be a very fruitful approach to learning about regulatory mechanisms in general and a useful starting point for understanding muscle specific regulation. The second critical process presented in this review is a subject of ongoing debate. When following this debate, the various models used to derive results should be carefully and critically evaluated. As an intellectual approach to understanding muscle hypertrophy, the literature that contributes to the debate regarding satellite cell participation will

provide many useful insights regardless of bias or the eventual outcome of the debate.

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Competing interests: None

LITERATUR

1. **Adams GR:** Invited Review: Autocrine/paracrine IGF-I and skeletal muscle adaptation. J Appl Physiol 93 (2002) 1159-1167.
2. **Adams GR, Caiozzo VJ, Haddad F, Baldwin KM:** Cellular and molecular responses to increased skeletal muscle loading after irradiation. Am J Physiol 283 (2002) C1182-C1195.
3. **Adams GR, Haddad F:** The relationships between IGF-1, DNA content, and protein accumulation during skeletal muscle hypertrophy. J Appl Physiol 81 (1996) 2509-2516.
4. **Allen DL, Roy RR, Edgerton VR:** Myonuclear domains in muscle adaptation and disease. Muscle & Nerve 22 (1999) 1350-1360.
5. **Barton-Davis ER, hoturma DI, Sweeney HL:** Contribution of satellite cells to IGF-I induced hypertrophy of skeletal muscle. Acta Physiol Scand 167 (1999) 301-305.
6. **Berkes CA, Tapscott SJ:** MyoD and the transcriptional control of myogenesis. Semin Cell Dev Biol 16 (2005) 585-595.
7. **Bickel CS, Slade J, Mahoney E, Haddad F, Dudley GA, Adams GR:** Time course of molecular responses of human skeletal muscle to acute bouts of resistance exercise. J Appl Physiol 98 (2005) 482-488.
8. **Booth FW, Tseng BS, Fluck M, Carson JA:** Molecular and cellular adaptation of muscle in response to physical training. Acta Physiol Scand 162 (1998) 343-350.
9. **Browne GJ, Proud CG:** Regulation of peptide-chain elongation in mammalian cells. Eur J Biochem 269 (2002) 5360-5368.
10. **Cheek DB, Holt AB, Hill DE, Talbert JL:** Skeletal muscle mass and growth: the concept of the deoxyribonucleic acid unit. Pediatr Res 5 (1971) 312-328.
11. **Childs TE, Spangenburg EE, Vyas DR, Booth FW:** Temporal alterations in protein signaling cascades during recovery from muscle atrophy. Am J Physiol 285 (2003) C391-C398.
12. **Clark A, Dean J, Tudor C, Saklatvala J:** Post-transcriptional gene regulation by MAP kinases via AU-rich elements. Front Biosci 14 (2009) 847-871.
13. **Deschênes-Furry J, Bélanger G, Mwanjewe J, Lunde JA, Parks RJ, Perrone-Bizzozero N, Jasmin BJ:** The RNA-binding protein HuR binds to acetylcholinesterase transcripts and regulates their expression in differentiating skeletal muscle cells. J Biol Chem 280 (2005) 25361-25368.
14. **Glass DJ:** Skeletal muscle hypertrophy and atrophy signaling pathways. Int J Biochem Cell Biol 37 (2005) 1974-1984.
15. **Haddad F, Adams GR:** Acute cellular and molecular responses to resistance exercise. J Appl Physiol 93 (2002) 394-403.
16. **Haddad F, Adams GR:** Aging Sensitive Cellular and Molecular Mechanisms Associated with Skeletal Muscle Hypertrophy. J Appl Physiol 100 (2006) 1188-1203.
17. **Hornberger TA, Chu WK, Ma, YW, Hsiung JW, Huang SA, Chien S:** The role of phospholipase D and phosphatidic acid in the mechanical activation of mTOR signaling in skeletal muscle. Proc Natl Acad Sci 103 (2006) 4741-4746.
18. **Huang J, Manning BD:** The TSC1-TSC2 complex: a molecular switchboard controlling cell growth. Biochem J 12 (2008) 179-190.
19. **Jastrzebski K, Hannan KM, Tchoubrieva EB, Hannan RD, Pearson RB:** Coordinate regulation of ribosome biogenesis and function by the ribosomal protein S6 kinase, a key mediator of mTOR function. Growth Factors 25 (2007) 209-226.
20. **Kadi F, Schjerling P, Andersen LL, Charifi N, Madsen JL, Christensen LR, Andersen JL:** The effects of heavy resistance training and detraining on satellite cells in human skeletal muscles. J Physiol 558 (2004) 1005-1012.
21. **Kimball SR:** The role of nutrition in stimulating muscle protein accretion at the molecular level. Biochem Soc Trans 35 (2007) 1298-1301.
22. **Kimball SR:** Interaction between the AMP-activated protein kinase and mTOR signaling pathways. Med Sci Sports Exerc 38 (2006) 1958-1964.
23. **Koh TJ, Pizza FX:** Do inflammatory cells influence skeletal muscle hypertrophy? Front Biosci E 1 (2009) 60-71.
24. **Kosek DJ, Bamman MM:** Modulation of the dystrophin-associated protein complex in response to resistance training in young and older men. J Appl Physiol 104 (2008) 1476-1484.
25. **Kumar V, Atherton P, Smith K, Rennie MJ:** Human muscle protein synthesis and breakdown during and after exercise. J Appl Physiol 106 (2009) 2026-2039.
26. **Lang CH, Frost RA, Vary TC:** Regulation of muscle protein synthesis during sepsis and inflammation. Am J Physiol 293 (2007) E453-E459.
27. **Mayer C, Grummt I:** Ribosome biogenesis and cell growth: mTOR coordinates transcription by all three classes of nuclear RNA polymerases. Oncogene 25 (2006) 6384-6391.
28. **Miyazaki M, Esser KA:** Cellular mechanisms regulating protein synthesis and skeletal muscle hypertrophy in animals. J Appl Physiol 106 (2009) 1367-1373.
29. **Nader GA, McLoughlin TJ, Esser KA:** mTOR function in skeletal muscle hypertrophy: Increased ribosomal RNA via cell cycle regulators. Am J Physiol 289 (2005) C1457-C1465
30. **Pedersen BK, Febbraio MA:** Muscle as an endocrine organ: focus on muscle-derived interleukin-6. Physiol Rev 88 (2008) 1379-1406.
31. **Petrella JK, Kim JS, Cross JM, Kosek DJ, Bamman MM:** Efficacy of myonuclear addition may explain differential myofiber growth among resistance-trained young and older men and women. Am J Physiol 291 (2006) E937-E946.
32. **Petrella JK, Kim JS, Mayhew DL, Cross JM Bamman MM:** Potent myofiber hypertrophy during resistance training in humans is associated with satellite cell-mediated myonuclear addition: a cluster analysis. J Appl Physiol 104 (2008) 1736-1742.
33. **Rommel C, Bodine SC, Clarke BA, Rossman R, Nunez L, Stitt TN, Yancopoulos GD, Glass DJ:** Mediation of IGF-1-induced skeletal myotube hypertrophy by PI(3)K/Akt/mTOR and PI(3)K/Akt/GSK3 pathways. Nature Cell Biol 3 (2001) 1009-1013.
34. **Sandri M:** Signaling in muscle atrophy and hypertrophy. Physiology (Bethesda) 23 (2008) 160-170.
35. **Sheehan SM, Tatsumi R, Temm-Grove CJ, Allen RE:** HGF is an autocrine growth factor for skeletal muscle satellite cells in vitro. Muscle and Nerve 23 (2000) 239-245.
36. **Thomson DM, Fick CA, Gordon SE:** AMPK Activation Attenuates S6K1, 4E-BP1, and eEF2 Signaling Responses to High-frequency Electrically Stimulated Skeletal Muscle Contractions. J Appl Physiol 104 (2008) 625-632.
37. **Velloso CP:** Regulation of muscle mass by growth hormone and IGF-I Br J Pharmacol 154 (2008) 557-568.
38. **Wackerhage H, Ratkevicius A:** Signal transduction pathways that regulate muscle growth. Essays Biochem 44 (2008) 99-108.
39. **Yablonka-Reuveni Z, Day K, Vine A, Shefer G:** Defining the transcriptional signature of skeletal muscle stem cells. J Anim Sci 86 (2008) E207-E216.
40. **Yamaguchi Y, Oohinata R, Naiki T, Irie K:** Stau1 negatively regulates myogenic differentiation in C2C12 cells. Genes Cells 13 (2008) 583-592
41. **van-Rooij E, Liu N, Olson EN:** MicroRNAs flex their muscles. Trends Genet 24 (2008) 159-166.

42. VAN-DER-GIESSEN K, GALLOUZI IE: Involvement of transportin 2-mediated HuR import in muscle cell differentiation. Mol Biol Cell 18 (2007) 2619-2629.

Correspondence:
Gregory Adams
University of California, Irvine
Department of Physiology and Biophysics
Medical Sciences 1
Irvine, CA 92697-4560
USA
E-Mail: GRAdams@uci.edu