Joanisse, Sophie and Lim, Changhyun and McKendry, James and Mcleod, Jonathan C and Stokes, Tanner and Phillips, Stuart M (2000) Recent advances in understanding resistance exercise training-induced skeletal muscle hypertrophy in humans. F1000Research, 9. p. 141.

Downloaded from: http://e-space.mmu.ac.uk/626494/

Version: Published Version

Publisher: F1000 Research Ltd

DOI: https://doi.org/10.12688/f1000research.21588.1

Usage rights: Creative Commons: Attribution 4.0

Please cite the published version
REVIEW

Recent advances in understanding resistance exercise training-induced skeletal muscle hypertrophy in humans

Sophie Joanisse, Changhyun Lim, James McKendry, Jonathan C. Mcleod, Tanner Stokes, Stuart M. Phillips

Exercise Metabolism Research Group, Department of Kinesiology, McMaster University, Hamilton, ON, Canada

Abstract

Skeletal muscle plays a pivotal role in the maintenance of physical and metabolic health and, critically, mobility. Accordingly, strategies focused on increasing the quality and quantity of skeletal muscle are relevant, and resistance exercise is foundational to the process of functional hypertrophy. Much of our current understanding of skeletal muscle hypertrophy can be attributed to the development and utilization of stable isotopically labeled tracers. We know that resistance exercise and sufficient protein intake act synergistically and provide the most effective stimuli to enhance skeletal muscle mass; however, the molecular intricacies that underpin the tremendous response variability to resistance exercise-induced hypertrophy are complex. The purpose of this review is to discuss recent studies with the aim of shedding light on key regulatory mechanisms that dictate hypertrophic gains in skeletal muscle mass. We also aim to provide a brief up-to-date summary of the recent advances in our understanding of skeletal muscle hypertrophy in response to resistance training in humans.

Keywords

resistance exercise, muscle, protein, hypertrophy
Corresponding author: Stuart M. Phillips (phillis@mcmaster.ca)

Author roles: Joanisse S: Conceptualization, Project Administration, Writing – Original Draft Preparation, Writing – Review & Editing; Lim C: Conceptualization, Project Administration, Writing – Original Draft Preparation, Writing – Review & Editing; McKendry J: Conceptualization, Project Administration, Writing – Original Draft Preparation, Writing – Review & Editing; Mcleod JC: Conceptualization, Project Administration, Writing – Original Draft Preparation, Writing – Review & Editing; Stokes T: Conceptualization, Project Administration, Writing – Original Draft Preparation, Writing – Review & Editing; Phillips SM: Conceptualization, Supervision, Writing – Original Draft Preparation, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

Grant information: SMP holds grants from the Canadian Institutes for Health Research and the National Science and Engineering Council of Canada and thanks the Canada Research Chairs Program for their support. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Copyright: © 2020 Joanisse S et al. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

How to cite this article: Joanisse S, Lim C, McKendry J et al. Recent advances in understanding resistance exercise training-induced skeletal muscle hypertrophy in humans [version 1; peer review: 2 approved] F1000Research 2020, 9(F1000 Faculty Rev):141 (https://doi.org/10.12688/f1000research.21588.1)

First published: 24 Feb 2020, 9(F1000 Faculty Rev):141 (https://doi.org/10.12688/f1000research.21588.1)
Introduction
Skeletal muscle is the organ of locomotion but is also a large contributor to resting energy expenditure\(^1\) and is the largest reservoir for post-prandial insulin-stimulated disposal of blood glucose\(^2\). Thus, beyond skeletal muscle’s obvious role in locomotion and mobility, its maintenance is critical for metabolic health. Indeed, lower-than-predicted norms of skeletal muscle mass and function are associated with a variety of negative health outcomes such as cardiovascular disease, cancer, and increased risk for disability\(^3\). Therefore, concerted efforts to maintain, increase, or regain lost skeletal muscle mass (for example, due to muscle disuse) are of relevance to human health\(^4\).

Skeletal muscle exhibits an extraordinary range of phenotypic plasticity in response to changing contractile stimuli. Skeletal muscle hypertrophy can be defined as an increase in muscle axial cross-sectional area (CSA), assessed via magnetic resonance imaging (MRI), computed tomography, ultrasound, and/or biopsies examining muscle fiber CSA (FCSA). Presently, chronic resistance exercise (RE) training (RET) and sufficient dietary protein feeding provide the most effective non-pharmacological strategies to promote skeletal muscle hypertrophy\(^5\). Significant attention has been directed towards deciphering the mechanistic underpinnings of what gives rise to skeletal muscle hypertrophy. The purpose of this review is to provide a brief up-to-date narrative on recent advances in our understanding of RET-induced skeletal muscle hypertrophy. It is notable that similar topical reviews have recently been published (see references\(^6\)–\(^8\)), and they should be consulted to obtain other viewpoints on this topic.

Exogenous versus endogenous variables in determining hypertrophy
Muscle hypertrophy is influenced by factors that can be broadly grouped into two categories: exogenous and endogenous variables. Exogenous factors include RE-related variables (load, reps, time under tension, volume, etc.), diet-related variables such as protein supplementation, energy intake, and consumption of anabolic supplements (i.e. creatine), and administration of anabolic hormones. The hypertrophic response to RET can be augmented marginally via greater-than-recommended protein ingestion, but the response is saturated around self-reported intakes of \(\sim 1.6 \text{ g protein/kg body mass/day}\); however, in resistance-trained individuals, protein intake may need to be greater (\(\sim 2.0–2.2 \text{ g protein/kg body mass/day}\)) to maximize whole-body anabolism\(^9\)–\(^10\). Specifically, leucine has been repeatedly shown to be the most potent, and possibly exclusively in human skeletal muscle\(^10\), amino acid agonist that induces muscle protein synthesis (MPS)\(^10\)–\(^12\).

Endogenous variables, namely genomic, epigenetic, transcriptomic, and proteomic variables\(^13\), are determinants of muscle hypertrophy. Importantly, each of these variables can ultimately be affected by exogenous variables, such as nutrition and RET paradigms, to which they may show differential responses. Extant literature demonstrates that manipulation of some RET variables has, at best, statistically significant but relatively small effects that are for the most part related to greater mechanical work (although this too would have a ceiling) and are most easily outwardly manifested by higher degrees of effort\(^14\). What is abundantly clear is that transient post-exercise rises in systemic concentrations of various anabolic hormones (testosterone, growth hormone, and insulin-like growth factor 1 (IGF-1)) are unrelated to muscle hypertrophy\(^15\)–\(^16\).

Although exogenous variables are important, it is becoming more widely appreciated that the endogenous molecular responses to RE are paramount in determining the hypertrophic response. Intramuscular mechano-sensitive signaling pathways and extracellular supporting structures (i.e. extracellular matrix and capillaries) appear to play important roles in hypertrophy\(^17\). While evidence is equivocal\(^18\)–\(^19\), our laboratory has demonstrated that individuals exhibiting greater hypertrophy in response to RET appear to have greater androgen receptor content at rest\(^20\), and the change in androgen receptor content is positively correlated with increased fCSA following RET\(^20\). Moreover, an enhanced satellite cell (SC) proliferation in response to loading\(^21\) differentiates higher from lower hypertrophic “responders” to RET. Furthermore, the aforementioned endogenous variables—higher androgen receptor content and augmented SC proliferation—have been reported to be greater in “high” compared to “low” responders to RET\(^22\)–\(^24\). Stimulation of MPS can also occur owing to increased efficiency of translation, with more mRNA translated per ribosomal unit\(^25\), or to increased translational capacity, which occurs by adding more ribosomes to translate existing mRNA. Therefore, ribosomal biogenesis has also been purported as an endogenous variable related to muscle hypertrophy\(^25\)–\(^26\). This concept is discussed in more detail further in the review. A schematic of these relationships is summarized in Figure 1. A tenet illustrated in this figure is that in response to mechanical loading, there are degrees of hypertrophic response on which people can, but also cannot, improve. Thus, similar to variability in response to any external stimulus, there is a response variability in exercise-induced hypertrophy that is propelled by external variables but predominantly translated into muscle growth through endogenous variables. Clearly, we do not have a complete picture of the loading-induced hypertrophic process, and further research is needed to define the relationship between exogenous variables and their effect on endogenous variables that directly mediate pathways leading to muscle hypertrophy.

Protein turnover and its role in skeletal muscle hypertrophy
Skeletal muscle hypertrophy occurs as the result of recurrent periods of positive net protein balance (NPB), when the rate of MPS exceeds that of muscle protein breakdown (MPB). In the post-absorptive (i.e. fasted) state, rates of MPB exceed MPS, resulting in a negative NPB\(^27\). Importantly, nutrition and contractile activity are potent modulators of MPS and, to a lesser extent, MPB in both trained\(^28\)–\(^30\) and untrained individuals\(^31\). Specifically, in the post-absorptive state, RE stimulates increases in both MPS and MPB, and while MPS is stimulated to a greater extent, NPB remains negative\(^31\). Ingestion of dietary protein containing sufficient essential amino acids\(^32\), in close temporal proximity to RE, augments MPS and attenuates the
Exercise-induced increase in MPB. Therefore, only when RE is coupled with protein feeding does NPB become positive, facilitating small periods of muscle protein accrual with RET that sum to yield eventual hypertrophy.\textsuperscript{37}

Changes in post-absorptive MPS are modified with RET (for review, see \textsuperscript{32}). Elevated post-absorptive MPS has been proposed as a primary contributor to muscle hypertrophy with RET (>6 weeks).\textsuperscript{6} Indeed, early observations in humans show that post-absorptive MPS is elevated in the trained state.\textsuperscript{32,33,34} However, identical to what is seen in untrained individuals, NPB in the post-absorptive state is always negative because of a concomitant elevation of MPB in trained individuals.\textsuperscript{30,32}

Thus, the trained state is demarked by an enhanced overall rate of protein turnover—elevated rates of MPS and MPB—that favors only net protein accretion, as demonstrated multiple times in the fed state. The elevation in MPB in the trained state is also supported by molecular evidence.\textsuperscript{36} Acute intermittent elevations in MPS in response to, and with persistent practice of, RE in combination with sufficient protein feeding are undeniably the major drivers of muscle protein accretion and skeletal muscle hypertrophy.\textsuperscript{37} We speculate that the overall increased protein turnover (as a result of cumulative greater acute periods of positive NPB) observed with chronic RET is advantageous and is reflective of a general increase in turnover of muscle proteins (i.e. upregulation...
of MPS and MPB) that favors efficient remodeling of protein that leads to a gradual muscle protein accrual manifested as hypertrophy; these concepts are depicted schematically in Figure 2.

At the molecular level, RE and protein feeding increase MPS through mechanistic target of rapamycin complex 1 (mTORC1)-dependent and -independent mechanisms. Typically, mTORC1 phosphorylation activates several downstream kinases, augmenting translational efficiency (i.e. an increase in the rate of translation of mRNA by a constant pool of ribosomes) and, with RET, translational capacity (i.e. total number of available ribosomes). Recently, it has been suggested that increased translational capacity is central to changes in post-absorptive MPS with chronic RET. Several groups have demonstrated that chronic RET results in increased total RNA and ribosomal RNA (rRNA) content in addition to increases in regulators of rRNA synthesis. In contrast, other groups

---

**Figure 2.** Current understanding of changes in muscle protein turnover with chronic resistance exercise training. Skeletal muscle hypertrophy can occur only under periods of positive protein balance; that is, when relative rates of muscle protein synthesis (MPS) (blue line) exceed that of muscle protein breakdown (MPB) (red line). In the fasted state, rates of MPB exceed those of MPS, resulting in a negative net protein balance (NPB). Compared to untrained individuals (A), trained individuals (B) display higher fasted rates of MPS; however, protein balance remains negative because of the concomitant elevation of MPB in the trained state. Regardless of training status, nutritional and contractile stimuli are potent regulators of MPS and, to a lesser extent, MPB. Resistance exercise (RE) stimulates increases in both MPS and MPB, and NPB remains negative. Ingestion of dietary protein—in particular, essential amino acids—in close temporal proximity to RE augments MPS and attenuates the exercise-induced increase in MPB, resulting in a temporary state of positive protein balance. Chronic RE training (RET) modulates the time course of the increase in MPS following a bout of RE. Specifically, the initial increase in MPS following a bout of RE is less pronounced in the untrained state than in the trained state; however, it is longer lived and peaks later in the untrained than the trained state. MPS, MPB, and NPB during periods of (B) RE+Fasted and (C) RE+Fed in the untrained and trained state.
reported a reduction in biomarkers of ribosomal biogenesis\textsuperscript{43} or no change following 12 weeks of RET\textsuperscript{47}. Increases in RNA content—following 16 weeks\textsuperscript{45,48} and 6 weeks\textsuperscript{18} of RET—were similar between individuals showing either no change (i.e. “non/low responders”) or an extreme increase (i.e. “extreme/high responders”) in vastus lateralis muscle iCSA. In contrast, Stec and colleagues\textsuperscript{41} reported that only “extreme” responders to 4 weeks of RET had increases in total RNA and rRNA content. Conflicting results may be attributed to differences in participant characteristics, experimental design, and analytical techniques\textsuperscript{37}; however, current evidence does not demonstrate a clear connection between translational capacity and skeletal muscle hypertrophy in humans\textsuperscript{17}. We hypothesize that early on in a RET program, ribosomal capacity may increase as a general response to a need for greater rates of global protein synthesis\textsuperscript{46}. However, with persistent practice of RET once protein synthetic responses and transcriptional programs become “refined” and more specific to the stimulus of RET\textsuperscript{14}—as well as being shorter in duration—further increasing ribosomal capacity is not required and would either stabilize\textsuperscript{35,42} or possibly decline\textsuperscript{13,47}. This thesis would underpin why early during a RET bout a very short-term MPS response does not align well with eventual hypertrophy\textsuperscript{48}, but this is not the case with further RET where MPS shares common variance with hypertrophy\textsuperscript{46}. It should also be noted that the stabilization of ribosomal capacity following chronic RET\textsuperscript{40,42} does not indicate a loss of muscle ribosomes per se; instead, this likely reflects a dilution of the ribosomal capacity by larger, hypertrophied myofibers.

Understanding changes in translational capacity with RET is limited owing to a number of methodological constraints. Specifically, the study of ribosomal biogenesis relies heavily on static measures (i.e. immunoblotting and quantifying total RNA content and assuming rRNA content is responsible), and traditional stable isotope tracer investigations provide insight into only acute (i.e. hours) metabolic fluctuations\textsuperscript{39}. Recent advances in mass spectrometry techniques have led to the reintroduction of deuterium oxide (D\textsubscript{2}O)\textsuperscript{19-31}, which enables the assessment of metabolic flux in response to a variety of stimuli, such as skeletal muscle loading\textsuperscript{11,12,42,46}, unloading\textsuperscript{23,41}, and feeding\textsuperscript{35,42,46} under longer-term, “free-living” conditions (i.e. integrated over days to weeks). Brook and colleagues\textsuperscript{30} recently validated the use of D\textsubscript{2}O in monitoring the synthesis of ribonucleotides, providing the first dynamic measure of RNA synthesis in human skeletal muscle in response to RET. Of particular note in this study, RNA synthesis was increased above basal rates over the 0–6-week period with continuous RET\textsuperscript{10}. Importantly, myofibrillar MPS in these individuals was not significantly increased above basal levels during this period\textsuperscript{42}, showing a discordance between translational capacity and MPS with long-term muscle adaptations. Future studies incorporating dynamic measures of RNA synthesis and integrated rates of MPS in concert with omic-level measurements should provide a platform to elucidate the relative contribution, and time-course, of translational efficiency and capacity to changes in MPS and hypertrophy in response to chronic RET.

### Omic-based science and skeletal muscle hypertrophy

Our present mechanistic understanding of muscle hypertrophy has largely been informed by the use of “targeted” analytical approaches providing static snapshots (i.e. qPCR and immunoblotting). However, the increased usage of “omic” technologies can offer an unbiased and integrative understanding of the processes regulating muscle hypertrophy. Proteomic profiling has tremendous potential to advance our understanding of muscle growth; however, it is currently constrained by a relatively limited coverage of highly abundant proteins in the proteome versus a far larger coverage of RNA: <500 proteins reliably detected\textsuperscript{15,56} versus ~30,000 RNA species\textsuperscript{43}. This low protein:RNA ratio results in an incomplete understanding of downstream ontology/pathway analyses\textsuperscript{17} but could also mask the important role of less-abundant regulatory proteins in muscle hypertrophy (i.e. signaling molecules\textsuperscript{57} or integrin receptors\textsuperscript{89}). It is possible to circumvent these limitations by studying the expressed RNA complement of the cell (via transcriptomics) or translatome of the cell (via polysomal RNA and transcriptomics), given the close association between mRNA and protein abundance under most conditions\textsuperscript{99,60} and, in particular, the global translatome in skeletal muscle\textsuperscript{61,62}.

Early applications of transcriptomics have shown that older adults, and lower hypertrophic responders in general\textsuperscript{49}, express a pro-inflammatory gene profile at rest and respond to an acute bout of RE with an exaggerated inflammatory response\textsuperscript{44}, linking inflammation with an attenuated muscle growth response to RET. Elderly adults also have an elevated expression of p21\textsuperscript{15}, a cell cycle inhibitor that affects SC proliferation\textsuperscript{64} and may therefore impair muscle growth following RET\textsuperscript{47}. In contrast, higher hypertrophy responders to RET express higher levels of several well-known growth and remodeling genes prior to training compared to lower responders, which is suggestive of a “primed” basal state of protein turnover\textsuperscript{46}. Higher RET responders also express greater levels of oxidative, angiogenic, and extracellular matrix remodeling genes after RET\textsuperscript{36,67}. Two noteworthy yet ill-characterized genes that are also upregulated in high responders in the basal state include NAP1L1 and DGKz\textsuperscript{63}, which encode a nucleosome-associated protein and diacylglycerol kinase zeta (DGKz), respectively. The protein encoded by NAP1L1 controls chromatin compaction but has also been shown to bind to and regulate the nuclear-cytoplasmic shuttling of DGKz\textsuperscript{68}. Importantly, DGKz was shown recently to play a pivotal role in mechanical overload-induced muscle hypertrophy in rodents, but only if the nuclear localization signal of DGKz was intact\textsuperscript{69}. While the nature of this interaction in humans warrants further investigation, the example attests to the hypothesis-generating power of transcriptome profiling and its inherent potential for biological discovery.

An ongoing challenge in transcriptomics is the use of gene ontology (i.e. DAVID\textsuperscript{70}) and network analytical tools (ingenuity pathway analysis [IPA]\textsuperscript{71}), which are commonly used to uncover functional relationships from large lists of RET-regulated genes. These tools rely on the function(s) of a gene product...
being known\textsuperscript{36}. However, data-driven networks (DDNs) are networks constructed on the basis of experimentally derived gene co-expression similarities, without \textit{a priori} knowledge of gene function. Clarke and colleagues\textsuperscript{52} used a DDN approach to construct gene networks from pre- and post-muscle transcriptome samples obtained from the HERITAGE study\textsuperscript{19} (endurance-based training) and identified \textit{EIF6} as an exercise-responsive highly interconnected “hub” gene. EIF6 was therefore predicted, on the basis of being highly connected to other regulated genes, to play an important role in the adaptation to endurance training. Indeed, subsequent development of a mutant EIF6 murine model was shown to affect many of the same signaling pathways predicted by the HERITAGE study\textsuperscript{72,73} that affect phenotype. Greater use of DDNs and network modeling could be applied to the study of muscle hypertrophy with RET with, we propose, great potential.

\textbf{SCs and their role in RET-induced hypertrophy}

In humans, increases in muscle fiber size are commonly reported with a concomitant increase in the number of myonuclei\textsuperscript{74}, an observation that lends credence to the myonuclear domain theory of muscle growth\textsuperscript{75}. This theory suggests that each myonucleus governs a set volume within the muscle fiber and, when the ceiling of the muscle fiber volume is reached, the transcriptional capacity of an existing myonucleus is reached and new myonuclei must be added to maintain (or re-establish) transcriptional control over a defined myonuclear domain. Skeletal muscle is a post-mitotic tissue; therefore, the addition of new myonuclei must come from a new source, which occurs via donation from skeletal muscle stem cells, i.e. SCs.

Activation of SCs occurs following various stimuli such as injury, damage, and exercise. Once activated, SCs progress from proliferation to terminal differentiation, eventually fusing and donating their nuclei to existing myofibers, a process termed the myogenic program. Although common dogma had long associated SCs with skeletal muscle hypertrophy\textsuperscript{76,77}, this concept has recently been challenged. McCarthy and colleagues\textsuperscript{78} were the first to use the Pax7-DTA mouse strain that results in conditional SC ablation to demonstrate that significant overload-induced hypertrophy, via synergist ablation, can occur in SC-depleted rodent skeletal muscle. The same group reinforced these findings using hind-limb suspension, to induce atrophy, followed by reloading and regrowth of muscle which was not affected by SC depletion, in the Pax7-DTA mouse\textsuperscript{79}. Importantly, while interesting, these results highlight that SCs are not necessary for hypertrophy in short-term extreme models of hypertrophy but do not address the question of whether SCs are involved in a more physiologically relevant hypertrophic situation (i.e. following RET). This notion was further challenged by a study from Egner and colleagues\textsuperscript{80}, in which they describe impaired hypertrophy with 2 weeks of overload, via synergist ablation, using the same Pax7-DTA mouse strain\textsuperscript{80,81,82}. Further to this, work by Murach and colleagues\textsuperscript{31} demonstrated that myonuclear accretion via the SC is necessary to support overload-induced hypertrophy in younger growing mice, highlighting that the requirement of SCs to support hypertrophy is affected by age. Notably, the extent of hypertrophy is attenuated following 8 (versus 2) weeks of overload-induced hypertrophy in Pax7-DTA mice\textsuperscript{82}, suggesting that SCs are involved in muscle growth. Importantly, the researchers described an accumulation of the extracellular matrix in SC-depleted mice following 8 weeks of overload, which resulted in the impaired hypertrophic response\textsuperscript{83}. These data suggest that SCs are able to support muscle growth not only by fusing to existing fibers resulting in myonuclear accretion but also by their interaction with other cell types to regulate the extracellular matrix deposition\textsuperscript{84}. Although work in rodent models has been essential in providing insight into the basic cellular and molecular mechanisms that result in muscle hypertrophy, these results cannot always easily be translated to humans. For example, cerebral palsy, a developmental motor disorder characterized by a reduction in muscle fiber size, is also associated with a reduction in SC content\textsuperscript{44,45}, and it is postulated that the reduction in SC content may contribute to the impairment in muscle growth\textsuperscript{85}. For obvious reasons, it isn’t possible to study the effects of SC depletion in humans, and the observation of SCs in a human model with a reduced (although not ablated) SC content is often confounded by the presence of chronic disease, where factors other than SC content may contribute to the inability of muscle to hypertrophy.

Importantly, the majority of evidence stemming from human studies has implicated a role for SCs in contributing to increases in muscle fiber size. Several studies have described a positive relationship between muscle fiber size and number of myonuclei in human muscle\textsuperscript{86,87,88,89}. In addition, studies have also described an increase in myonuclear number with training-induced fiber hypertrophy concomitant with an increase in SC content\textsuperscript{88,89,90}. It is, however, important to note that several groups have reported an increase in fCSA without an increase in SC/myonuclear content\textsuperscript{92-94}. This may be due to several factors, one of which is the ability of existing myonuclei to increase their transcriptional capacity to support the increase in muscle fiber size\textsuperscript{91}.

Interestingly, individuals classified as “extreme” (hypertrophy) responders to RET had greater basal SC content compared to “lower” and “moderate” responders, which translated to a greater expansion of the SC pool with training and was accompanied by an increase in myonuclear content; however, the myonuclear domain also increased\textsuperscript{91}. Thus, similar to transcriptional observations, the basal characteristics of skeletal muscle (i.e. SC content) may play a role in response plasticity to hypertrophic stimulus. Congruent with previous work\textsuperscript{21}, we demonstrated that the acute SC response to a bout of unaccustomed RE is related to the increase in quadriceps volume observed following training\textsuperscript{97}. Although SCs likely contribute to hypertrophic adaptation via myonuclear accretion, it is important to recognize the ability of resident myonuclei to respond to varying stimuli such as RET and their inherent ability to support growth. The concept of muscle “memory”, manifested through possible epigenetic changes, is also likely an important contributor to the ability of skeletal muscle to hypertrophy. Seaborn and colleagues\textsuperscript{98} demonstrated
that prior RE-induced hypertrophy enhanced the subsequent response to a bout of resistance training, following a period of detraining, which may be a consequence of the widespread hypomethylation incurred during the first adaptive response. Together, the evidence in humans reporting an increase in muscle fiber size with a concomitant increase in myonuclei[19,21,47,57–92 highlights that SCs likely play a role in mediating skeletal muscle hypertrophy. However, as shown by Kirby and colleagues[65, using a time-course experiment following synergist ablation in the Pax7-DTA mouse model, the ability of existing resident myonuclei to support periods of fiber growth cannot be disregarded.

### Conclusion and future directions

Skeletal muscle plays an indispensable role in an array of mechanical and metabolic functions[66]. Typically, as we age, the quantity and quality of skeletal muscle deteriorates owing to the infiltration of non-muscle tissue including adipose and connective tissue[67]. Therefore, concerted efforts to increase and maintain skeletal muscle mass should be made by a range of individuals spanning from those striving to improve athletic performance to those focused on extending the healthspan. RE and dietary protein act synergistically and, at present, provide the most effective strategy to augment skeletal muscle mass[3]. Skeletal muscle hypertrophy is a complex process with multiple regulatory gene/protein hubs that have recently received significant attention in helping to decipher the mechanistic underpinnings that dictate the skeletal muscle adaptive response. As a result, a number of exogenous factors that influence endogenous pathways have been identified to play an important role in skeletal muscle hypertrophy.

MPS is the principal locus of control that influences muscle protein accretion in response to anabolic stimuli, as opposed to MPB[38–31]. However, the relative contribution of increased translational efficiency and translational capacity in affecting hypertrophy remains unclear. Intermittent elevations in rates of MPS in response to exogenous stimuli (i.e. RE and protein nutrition) drive muscle hypertrophy[32–33]. Nevertheless, research focused on translational capacity is in its infancy, and the proposed importance of ribosomal biogenesis has yet to be confirmed.

What is clearly evident is that muscle hypertrophy is a multifaceted process. However, targeted approaches that probe specific genes and proteins will provide only an incomplete picture of muscle growth. Unbiased, global “omic” technologies have the potential to provide a more comprehensive understanding of the underlying prerequisites for muscle growth but have inherent limitations that need to be considered.

Myonuclear accretion, due to a loading stimulus, is a means by which the transcriptional capacity of the skeletal muscle may be increased. The addition of new myonuclei is due to the activation and subsequent fusion of SCs to muscle fibers, and substantial evidence shows a role for SCs in muscle hypertrophy in humans. Although this is speculative, we hypothesize that resident myonuclei likely possess the ability, possibly through epigenetic modification, to increase transcriptional capacity to a certain extent, ultimately supporting muscle growth.

Although significant progress has been made, considerable work remains to be done in order to deepen our understanding of the processes that govern RET-induced muscle hypertrophy. Future studies incorporating dynamic measures of RNA synthesis, integrated rates of MPS, and SC/myonuclei assessments in concert with “omic” technologies and DDNs will provide a platform to elucidate the relative contribution, and time-course, of translational efficiency and capacity to changes in MPS and hypertrophy in response to chronic RET.

### References

1. Zurlo F, Larson K, Bogardus C, et al.: Skeletal muscle metabolism is a major determinant of resting energy expenditure. J Clin Invest. 1990; 86(5): 1423–7. PubMed Abstract | Publisher Full Text | Free Full Text

2. Thibaud D, Jacob E, DeFronzo RA, et al.: The effect of graded doses of insulin on total glucose uptake, glucose oxidation, and glucose storage in man. Diabetes. 1982; 31(11): 957–63. PubMed Abstract | Publisher Full Text

3. mallor JC, Stokes T, Phillips SM: Resistance Exercise Training as a Primary Countermeasure to Age-Related Chronic Disease. Front Physiol. 2019; 10: 645. PubMed Abstract | Publisher Full Text | Free Full Text

4. Phillips SM, McGorry C: CrossTalk proposal: The dominant mechanism causing muscle atrophy is decreased protein synthesis. J Physiol. 2014; 592(24): 5341–3. PubMed Abstract | Publisher Full Text | Free Full Text

5. Morton RW, Murphy KT, McAllister SR, et al.: A systematic review, meta-analysis and meta-regression of the effect of protein supplementation on resistance training-induced gains in muscle mass and strength in healthy adults. Br J Sports Med. 2018; 52(6): 376–84. PubMed Abstract | Publisher Full Text | Free Full Text

6. Figueiredo VC: Revisiting the roles of protein synthesis during skeletal muscle hypertrophy induced by exercise. Am J Physiol Regul Integr Comp Physiol. 2019; 317(5): R709–R718. PubMed Abstract | Publisher Full Text | F1000 Recommendation

7. Lavin KM, Roberts BM, Fry CS, et al.: The Importance of Resistance Exercise Training to Combat Neuromuscular Aging. Physiology (Bethesda). 2019; 34(2): 112–22. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

8. Roberts MD, Hain CT, Mobley CB, et al.: Physiological Differences Between Low Versus High Skeletal Muscle Hypertrophic Responders to Resistance Exercise Training: Current Perspectives and Future Research Directions. Front Physiol. 2018; 9: 183. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

9. Mazziolla M, Sawan SW, Williamson E, et al.: Protein Intake to Maximize Whole-Body Anabolism during Postexercise Recovery in Resistance-Trained Men with High Habitual Intakes is Severalfold Greater than the Current Recommended Dietary Allowance. J Nutr. 2019. pii: mxz249. PubMed Abstract | Publisher Full Text | F1000 Recommendation

10. Albertron PJ, Smith K, Etheridge T, et al.: Distinct anabolic signalling responses to amino acids in C2C12 skeletal muscle cells. Amino Acids. 2010; 38(5): 1533–9. PubMed Abstract | Publisher Full Text

11. Devries MC, McGlory C, Bolster DR, et al.: Protein leucine content is a determinant of shorter- and longer-term muscle protein synthetic responses at rest and following resistance exercise in healthy older women: A randomized, controlled trial. Am J Clin Nutr. 2018; 107(2): 217–26. PubMed Abstract | Publisher Full Text
Changes in Skeletal Muscle Protein Synthesis and Their Contribution to Hypertrophy. Sports Med. 2020; 50(4): 801–7.

Pubmed Abstract | Publisher Full Text

33. Kim PL, Staron RS, Phillips SM: Fasted-state Skeletal Muscle Protein Synthesis After Resistance Exercise Is Altered With Training. J. Physiol. 2005; 568(Pt 1): 283–90.

Pubmed Abstract | Publisher Full Text | Free Full Text

34. Wilkinson SB, Phillips SM, Atherton PJ, et al.: Differential Effects of Resistance and Endurance Exercise in the Fed State on Signalling Molecule Phosphorylation and Protein Synthesis in Human Muscle. J Physiol. 2008; 585(15): 3701–17.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

35. Kumar V, Atherton P, Smith K, et al.: Human Muscle Protein Synthesis and Breakdown During and After Exercise. J Appl Physiol. 1985; 59(6): 2026–39.

Pubmed Abstract | Publisher Full Text | Free Full Text

36. Seabrook RA, Hughes DC, Turner DC, et al.: UBRs is a novel E3 ubiquitin ligase involved in skeletal muscle hypertrophy and recovery from atrophy. J Physiol. 2019; 597(14): 3727–49.

Pubmed Abstract | Publisher Full Text | F1000 Recommendation

37. Brook MS, Wilkinson DJ, Smith K, et al.: It’s not just about protein turnover: the role of ribosomal biogenesis and satellite cells in the regulation of skeletal muscle hypertrophy. Eur J Sport Sci. 2019; 19(7): 492–63.

Pubmed Abstract | Publisher Full Text | F1000 Recommendation

38. Hodson N, West DWD, Philip A, et al.: Molecular regulation of human skeletal muscle protein synthesis in response to exercise and nutrients: a compass for overcoming age-related anabolic resistance. Am J Physiol Cell Physiol. 2019; 317(6): C1061–C1078.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

39. You JS, McNally RM, Jacobs BL, et al.: The role of raptor in the mechanical load-induced regulation of mTOR signaling, protein synthesis, and skeletal muscle hypertrophy. FASEB J. 2019; 33(3): 4021–34.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

40. Hammarström D, Ofsteng S, Koll L, et al.: Benefits of higher resistance-training volume are related to ribosomal biogenesis. J Physiol. 2020; 598(3): 543–65.

Pubmed Abstract | Publisher Full Text | F1000 Recommendation

41. Shec MJ, Kelly NA, Many GM, et al.: Ribosome biogenesis may augment resistance training-induced myofiber hypertrophy and is required for myotube growth in vitro. Am J Physiol Endocrinol Metab. 2019; 319(6): E652–E661.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

42. Brook MS, Wilkinson DJ, Mitchell WK, et al.: Synchronous deficits in cumulative muscle protein synthesis and ribosomal biogenesis underlie age-related anabolic resistance to exercise in humans. J Physiol. 2016; 594(24): 7399–417.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

43. Fyle JJ, Bishop DJ, Bartlett JD, et al.: Enhanced skeletal muscle ribosome biogenesis, yet attenuated mTORC1 and ribosome biogenesis-related signalling, following short-term concurrent versus single-mode resistance training. Sci Rep. 2018; 8(1): 560.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

44. Bazman MN, Petrella JK, Kim JS, et al.: Cluster analysis tests the importance of myogenic gene expression during myofiber hypertrophy in humans. J Physiol. 2007; 102(6): 2232–9.

Pubmed Abstract | Publisher Full Text | Free Full Text

45. Kim JS, Petrella JK, Cross JM, et al.: Load-mediated downregulation of myostatin mRNA is not sufficient to promote myofiber hypertrophy in humans: a cluster analysis. J Appl Physiol (1985). 2007; 103(5): 1488–95.

Pubmed Abstract | Publisher Full Text | Free Full Text

46. Dumas F, Phillips SM, Libardi CA, et al.: Resistance training-induced changes in integrated myofibrillar protein synthesis are related to hypertrophy only after attenuation of muscle damage. J Physiol. 2016; 594(18): 5209–22.

Pubmed Abstract | Publisher Full Text | Free Full Text

47. Kadi F, Schjerling P, Andresen LL, et al.: The effects of heavy resistance training and detraining on satellite cells in human skeletal muscles. J Physiol. 2004; 558(Pt 3): 1025–10.

Pubmed Abstract | Publisher Full Text | Free Full Text

48. Mitchell CJ, Churchward-Venne TA, Parise G, et al.: Acute Post-Exercise Myofibrillar Protein Synthesis Is Not Correlated With Resistance Training-Induced Muscle Hypertrophy in Young Men. J Appl Physiol. 2014; 92(4): e89431.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

49. Mitchell CJ, Churchward-Venne TA, Cameron-Smith D, et al.: What is the relationship between the acute muscle protein synthesis response and changes in muscle mass? J Physiol. 2015; 118(4): 495–7.

Pubmed Abstract | Publisher Full Text | Free Full Text

50. Brook MS, Wilkinson DJ, Mitchell WK, et al.: A novel D_0 tracer method to quantify RNA turnover as a biomarker of de novo ribosomal biogenesis, in vitro, in animal models, and in human skeletal muscle. Am J Physiol Endocrinol Metab. 2017; 313(6): E681–E689.

Pubmed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

51. Wilkinson DJ, Franch MR, Brook MS, et al.: A validation of the application of D_0 stable isotope tracer techniques for monitoring day-to-day changes in muscle growth.
Integrin signaling: linking mechanical Potts GK, McNally RM, Blanco R, Murach KA, White SH, Wen Y, Satellite cell depletion prevents fiber recruitment. J Appl Physiol (1985). 2019; 126(6): 1861–70.

Timmons JA, Atherton PJ, Larsson O, et al.: A coding and non-coding transcriptomic perspective on the genomics of human metabolic disease. Nucleic Acids Res. 2018; 46(10): 7772–92.

Timmons JA, Szpak KJ, Gallagher U: Multiple sources of bias confound functional enrichment analysis of global -omics data. Genome Biol. 2015; 16: 186.

Potts GK, McNally RM, Blanco R, et al.: A map of the phosphoproteome alterations that occur after a bout of maximal-intensity contractions. J Physiol. 2017; 598(15): 3209–3226.

Boppard MD, Mahmassani ZS: Integrin signaling: linking mechanical stimulation to skeletal muscle hypertrophy. Am J Physiol Cell Physiol. 2019; 317(4): C629–C641.

Li JJ, Bickel PJ, Biggin MD: System wide data analyses have underestimated protein abundances and the importance of transcription in mammals. PeerJ. 2014; 2: e270.

Chen YW, Nader GA, Baar KR, et al.: Response of rat muscle to acute resistance exercise defined by transcriptional and translational profiling. J Physiol. 2002; 546(1): 27–41.

Roberts MD, Childs TE, Brown JD, et al.: Age-specific functional epigenetic changes in p21 and p16 in injury-activated satellite cells. Stem Cells. 2015; 33(9): 951–61.

Thalacker-Mercer A, Stoe M, Cui X, et al.: Cluster analysis reveals differential transcript profiles associated with resistance training-induced human skeletal muscle hypertrophy. Physiol Genomics. 2013; 45(12): 499–507.

Thalacker-Mercer AE, Dhillon AJ, Cui X, et al.: Differential genomic responses in old vs. young humans despite similar levels of modest muscle damage after resistance loading. Physiol Genomics. 2010; 40(3): 141–9.

Roux U, Trape T, Carpentier ET, et al.: Transcriptional signature of resistance exercise adaptations: mixed muscle and fiber type specific profiles in young and old adults. J Appl Physiol (1985). 2012; 112(10): 1625–36.

Okada M, Hozumi Y, Ichimura T, et al.: Interaction of nucleosome assembly proteins abolishes nuclear localization of DGKβ, by attenuating its association with importins. Exp Cell Res. 2011; 317(20): 2853–63.

Jo YS, Dooley MS, Kim CR, et al.: A DGK-FOXO ubiquitinyl proteolytic axis controls fiber size during skeletal muscle remodeling. Sci Signal. 2018; 11(530): eaao847.

Guo J, Wang R, Zhang Y, et al.: Integrating biological meaning from large gene lists with DAVID. Curr Protoc Bioinformatics. 2009: Chapter 13: Unit 13.11.

Krämmer A, Green J, Pollard JR, et al.: Causual analysis approaches in Ingenuity Pathway Analysis. Bioinformatics. 2014; 30(5): 523–30.

Clarke K, Ricciardi S, Pearson T, et al.: The Role of E36 in Skeletal Muscle

Homeostasis Revealed by Endurance Training Co-expression Networks. Cell Rep. 2017; 21(6): 1507–1520.

Timmons JA, Krudsen S, Rankinen T, et al.: Using molecular classification to predict gains in maximal aerobic capacity following endurance exercise training in humans. J Appl Physiol (1985). 2010; 108(6): 1487–96.

Murak KA, Fry CS, Kirby TJ, et al.: Starring or Supporting Role? Satellite Cells and Skeletal Muscle Fiber Size Regulation. Physiology (Bethesda). 2018; 33(1): 26–38.

Allen DL, RO RR, Edgerton VR: Myonuclear domains in muscle adaptation and disease. Muscle Nerve. 1999; 22(10): 1350–60.

Adams GR, Caiccoo VJ, Haddad F, et al.: Cellular and molecular responses to increased skeletal muscle loading after irradiation. Am J Physiol Cell Physiol. 2002; 283(4): C1185–92.

Rosenblat JD, Parry DJ: Gamma irradiation prevents compensatory hypertrophy of overloaded mouse extensor digitorum longus muscle. J Appl Physiol (1985). 1992; 73(9): 2548–43.

McCarthy JJ, Mula J, Miyazaki E, et al.: Effective fiber hypertrophy in satellite cell-depleted skeletal muscle. Development. 2011; 138(7): 3657–66.

Jackson JR, Mula J, Kirby TJ, et al.: Satellite cell depletion does not inhibit adult skeletal muscle regrowth following unloading-induced atrophy. Am J Physiol Cell Physiol. 2012; 303(9): C864–C861.

Egner IM, Bruusgaard JC, Gundersen K: Satellite cell depletion prevents fiber hypertrophy in skeletal muscle. Development. 2016; 143(16): 2898–906.

Murak KA, White SH, Wen Y, et al.: Differential requirement for satellite cells during overload-induced muscle hypertrophy in growing versus mature mice. Skelet Muscle. 2017; 7(1): 14.

Fry CS, Lee JD, Jackson JR, et al.: Regulation of the muscle fiber microenvironment by activated satellite cells during hypertrophy. FASEB J. 2014; 28(4): 1654–65.

Fry CS, Kirby TJ, Kosman K, et al.: Myogenic Progenitor Cells Control Extracellular Matrix Production by Fibroblasts during Skeletal Muscle Hypertrophy. Cell Stem Cell. 2017; 20(1): 56–69.

Smith LR, Chambers HG, Lieber RL: Reduced satellite cell population may lead to contractures in children with cerebral palsy. Dev Med Child Neurol. 2013; 55(3): 264–70.

Dayanidhi S, Dykstra PB, Lubysuyv V, et al.: Reduced satellite cell number in situ in muscular contractures in children with cerebral palsy. J Orthop Res. 2015; 33(7): 1039–45.

Dayanidhi S, Lieber RL: Skeletal muscle satellite cells: mediators of muscle growth during development and implications for developmental disorders. Muscle Nerve. 2014; 50(5): 723–32.

Bellamy LM, Joannis S, Grubb A, et al.: The acute satellite cell response and skeletal muscle hypertrophy following resistance training. PLoS One. 2014; 9(10): e109799.

Kadi F, Thomell LE: Concomitant increases in myonuclear and satellite cell content in female trapezius muscle following strength training. Histochim Cell Biol. 2000; 113(2): 99–103.

Leenders M, Verjil LB, van der Hoeven L, et al.: Elderly men and women benefit equally from prolonged resistance-type exercise training. J Gerontol A Biol Sci Med Sci. 2013; 68(7): 769–79.

Farup J, Raabek SK, Ris S, et al.: Influence of exercise contraction mode and protein supplementation on human skeletal muscle satellite cell content and muscle fiber growth. J Appl Physiol (1985). 2014; 117(8): 898–909.

Petrella JK, Kim JS, Cross JM, et al.: Efficacy of myonuclear addition may explain differential myofiber growth among resistance-trained young and older men and women. Am J Physiol Endocrinol Metab. 2006; 291(5): E937–46.
93. Fry CS, Noehren B, Mula J, et al.: Fibre type-specific satellite cell response to aerobic training in sedentary adults. J Physiol. 2014; 598(12): 2625–35. PubMed Abstract | Publisher Full Text | Free Full Text

94. Murach KA, Walton RG, Fry CS, et al.: Cycle training modulates satellite cell and transcriptional responses to a bout of resistance exercise. Physiol Rep. 2016; 4(18): pii: e12973. PubMed Abstract | Publisher Full Text | Free Full Text

95. Kirby TJ, Patel RM, McClintock TS, et al.: Myonuclear transcription is responsive to mechanical load and DNA content but uncoupled from cell size during hypertrophy. Mol Biol Cell. 2016; 27(5): 788–98. PubMed Abstract | Publisher Full Text | Free Full Text

96. Seaborn RA, Strauss J, Cocks M, et al.: Human Skeletal Muscle Possesses an Epigenetic Memory of Hypertrophy. Sci Rep. 2018; 8(1): 1898. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

97. Frontera WR, Ochala J: Skeletal muscle: a brief review of structure and function. Calcif Tissue Int. 2015; 96(3): 183–95. PubMed Abstract | Publisher Full Text

98. Nilwik R, Snijders T, Leenders M, et al.: The decline in skeletal muscle mass with aging is mainly attributed to a reduction in type II muscle fiber size. Exp Gerontol. 2013; 48(5): 492–8. PubMed Abstract | Publisher Full Text | F1000 Recommendation
Open Peer Review

Current Peer Review Status: ✔️ ✔️

Editorial Note on the Review Process
F1000 Faculty Reviews are written by members of the prestigious F1000 Faculty. They are commissioned and are peer reviewed before publication to ensure that the final, published version is comprehensive and accessible. The reviewers who approved the final version are listed with their names and affiliations.

The reviewers who approved this article are:

Version 1
1 John J McCarthy
   Department of Physiology, College of Medicine, University of Kentucky, Lexington, KY, USA
   Competing Interests: No competing interests were disclosed.
2 Michael Roberts
   School of Kinesiology, Auburn University, Auburn, AL, USA
   Competing Interests: No competing interests were disclosed.

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com