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Pharmacological targeting of exercise adaptations in skeletal muscle: Benefits and pitfalls

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Abstract

Exercise exerts significant effects on the prevention and treatment of many diseases. However, even though some of the key regulators of training adaptation in skeletal muscle have been identified, this biological program is still poorly understood. Accordingly, exercise-based pharmacological interventions for many muscle wasting diseases and also for pathologies that are triggered by a sedentary lifestyle remain scarce. The most efficacious compounds that induce muscle hypertrophy or endurance are hampered by severe side effects and are classified as doping. In contrast, dietary supplements with a higher safety margin exert milder outcomes. In recent years, the design of pharmacological agents that activate the training program, so-called “exercise mimetics”, has been proposed, although the feasibility of such an approach is highly debated. In this review, the most recent insights into key regulatory factors and therapeutic approaches aimed at leveraging exercise adaptations are discussed.

Keywords

exercise; skeletal muscle; AMPK; mTOR; PPAR β / δ ; PGC-1 α ; dietary supplements; exercise mimetics

Introduction

Physical activity is one of the most efficient, (cost-)effective and accessible treatments for obesity and other diseases [1]. Endurance exercise (EE) and resistance exercise (RE) significantly ameliorate the symptoms of various chronic pathologies, e.g. by reducing inflammation and insulin resistance, bolstering mood and general well-being, lowering stress levels and even positively influencing cognitive function [1]. Many of these systemic effects are mediated by signaling molecules that are produced and secreted by skeletal muscle in response to exercise [2]. Skeletal muscle is a remarkably malleable tissue capable of responding to physical and metabolic demands, which makes skeletal muscle an attractive target for pharmacological interventions to mimic exercise in order to enhance performance and ameliorate diseases. Generally speaking, EE training leads to increased mitochondrial content and thus oxidative capacity, while RE training elicits changes in size and contractile

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Conflict of interest

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properties of muscle fibers [3]. Some studies indicate that a combination of both modes of exercise may confer synergistic benefits [4] while others suggest that concurrent EE and RE may interfere with each other. Depending on the amount and modality of concurrent EE with RE, EE could hamper the hypertrophic response of the body to RE via the so-called “exercise interference” effect [5].

Increased mitochondrial content, fatty acid oxidation and capillary density facilitating oxygen exchange and nutrient uptake of EE-trained muscle fibers collectively provide adequate oxygen and substrate supply as well as the enzymatic capacity needed for aerobic energy production in times of elevated demand. Traditionally, prolonged sessions of EE have mostly been used to induce beneficial adaptations, while alternative training modalities have emerged in recent years [6]. For example, in high intensity training (HIT) featuring exercise bouts of higher intensity but significantly shorter duration, similar or even better adaptations in mitochondrial content and capillary density compared to moderate-intensity continuous training (MICT) were achieved [7], presumably by inducing a stronger stimulus to recruit more muscle fibers. In contrast to the potent effect on exercise adaptations, some evidence suggests that HIT may be less effective than prolonged training in a disease-context [8], while other data indicate an enhanced effectiveness in pathological situations [9].

RE leads to an increased myofiber size and elicits changes in the abundance of contractile and metabolic proteins of the exercised muscles [10]. RE triggers the accretion of protein in the growing myofiber, a process thought to be largely governed by the mammalian target of rapamycin (mTOR) [3]. RE may also increase proliferation of muscle stem cells, called satellite cells, which fuse with and thus donate their myonuclei to the growing myofibers to maintain equal myonuclear domains.

Mechanistic aspects of exercise-induced skeletal muscle adaptation

Even though the mechanisms underlying the plastic changes of skeletal muscle after exercise are far from being fully understood, several important regulators were identified. In this section, some key aspects of these regulators are summarized, including their potential for pharmacological modulation in diseases (Fig. 1).

Peroxisome proliferator-activated receptor- γ (PPAR γ) coactivator 1 α (PGC-1 α)

PGC-1 α was originally identified in brown adipose tissue (BAT) as a coactivator of PPAR γ [11]. However, PGC-1 α is expressed in all tissues that are rich in mitochondria and show high oxidative capacity, including the heart, brain, kidney and skeletal muscle [11]. In all of these tissues, PGC-1 α is pivotal in regulating a plethora of biological responses related to glucose and fatty acid metabolism, most of which are functionally linked to the potent effect on mitochondrial biogenesis [11]. In addition, PGC-1 α controls tissue-specific programs, e.g. a fiber-type switching in skeletal muscle, thermogenesis in BAT or gluconeogenesis in the liver. Thus, for almost two decades since its discovery, PGC-1 α has been recognized as a powerful regulator of cellular energy metabolism and mitochondrial capacity in health and disease [12].

In skeletal muscle, PGC-1 α is highly inducible by acute and chronic EE training in rodents [13] and humans [14], where it orchestrates many of the adaptations to exercise like increased oxidative metabolism or vascularization [15]. PGC-1 α interacts with and activates a number of transcription factor partners and therefore serves as a pleiotropic modulator of a multitude of pathways, ultimately leading to an endurance-trained phenotype [15, 16]. Due to this effect, PGC-1 α constitutes an attractive drug target [17], however with several hurdles and pitfalls that have to be overcome. In most tissues, the physiological regulation of PGC-1 α results in transient bursts of expression, while constitutive elevation, in particular at supraphysiological levels, might lead to a detrimental outcome. For example, constitutive, postnatal cardiac overexpression of PGC-1 α leads to exacerbated mitochondrial biogenesis in cardiomyocytes and consequently to heart failure [18]. In a different study using an inducible mouse model where PGC-1 α expression was elevated for several weeks in the heart, the development of reversible cardiomyopathy with abnormal mitochondrial ultrastructure was observed [19]. Moreover, skeletal muscle-specific transgenic animals with supraphysiological PGC-1 α expression exhibited a myopathic phenotype [20, 21]. In contrast, sustained elevation of PGC-1 α at physiological levels in skeletal muscle leads to a variety of beneficial effects resembling the adaptation of EE [20]. Such transgenic overexpression of PGC-1 α in muscle entails a switch towards oxidative type IIa, and to a lesser extent, type I muscle fibers with elevated expression of mitochondrial markers, boosted mitochondrial number and size as well as improved fatigue resistance [20]. In addition, in several muscle wasting conditions such as denervation, hind limb unloading and even in Duchenne Muscular Dystrophy (DMD), elevation of muscle PGC-1 α reduces muscle atrophy and improves muscle function [22]. However, this mouse line also exhibits signs of pathology in specific contexts, e.g. when exposed to a high fat diet [23] and curiously, a reduced number of satellite cells, even though this reduction is not linked to an impaired regenerative capacity [17]. Tissue selectivity in pharmacological modulation of PGC-1 α might have to be aimed for, e.g. in the treatment of type 2 diabetes mellitus (T2DM). T2DM is a chronic metabolic disorder characterized by insulin resistance, hyperglycemia and insulin deficiency. Behavioral, environmental, as well as genetic factors play a role in the development of the disease. A combination of physical activity, dietary interventions and medication has been the weapon of choice in combatting T2DM [24]. In T2DM, PGC-1 α levels are heterogeneously dysregulated in various tissues, for example constitutively elevated in the liver [25] and pancreas [26], but at the same time decreased in skeletal muscle [27]. The increased activity of PGC-1 α in the liver stimulates gluconeogenesis [25] contributing to insulin resistance, while in the pancreas, this coactivator suppresses insulin secretion [26] and thus promotes insulin deficiency. The decreased levels of PGC-1 α in diabetic skeletal muscle lead to reduced oxidative phosphorylation, diminished glucose transporter 4 (GLUT4) expression and ultimately to glucose intolerance [28] as skeletal muscle normally accounts for the majority of insulin-stimulated glucose uptake, storage and utilization [29]. A therapeutic approach aimed at normalizing PGC-1 α levels would thus increase skeletal muscle PGC-1 α activity, while concomitantly inhibiting PGC-1 α in liver and pancreas. While the tissue-specific regulation and effects of PGC-1 α are still poorly understood, the selective coactivation of transcription factor binding partners in different cell types could potentially be leveraged for this purpose. For example, PGC-1 α coactivates the hepatic nuclear factor-4 α (HNF-4 α) in the control of

hepatic gluconeogenesis [25]. Since this interaction is preferentially observed in the liver and not skeletal muscle or other tissues that lack substantial HNF-4 α expression, selective inhibitors of the PGC-1 α -HNF-4 α interaction could constitute a feasible strategy to counteract excessive glucose production in T2DM [25]. Unfortunately, compounds that block or enhance the coactivation of specific transcription factors by PGC-1 α are still rare. Proof-of-concept for a therapeutical approach to reduce PGC-1 α activity in the liver of T2DM mice was demonstrated in a recent screening study, in which a compound, SR-18292, was identified to increase acetylation of PGC-1 α , thereby reducing the coactivation of HNF-4 α and ultimately the expression of the gluconeogenic genes phosphoenolpyruvate carboxykinase 1 (*PCK1*) and glucose-6-phosphatase catalytic (*G6PC*) in the liver (Fig. 2) [30]. Treatment with SR-18292 resulted in diminished hepatic glucose production and thereby ameliorated the diabetic phenotype of these animals. However, since acetylation as a mechanism of regulation of PGC-1 α activity is observed in different tissues including skeletal muscle [31], treatment with a non-tissue selective deacetylation inhibitor could potentially lead to unwanted side effects. Thus, pharmacological modulation of PGC-1 α would optimally have to be achieved in a temporally and spatially controlled manner within a therapeutic window to avoid detrimental outcomes of insufficient and exacerbated PGC-1 α levels [22].

Peroxisome proliferator-activated receptor β/δ (PPAR β/δ)

PPAR β/δ is a nuclear receptor encoded by the *PPARD* gene and is the PPAR isoform with the highest expression in skeletal muscle [32]. Upon binding of fatty acids and other ligands, PPARs become transcriptionally active, heterodimerize with retinoid X receptors (RXR), and recruit coactivators and histone acetyltransferase (HAT) complexes to stimulate the expression of target genes. Inversely, PPARs can also inhibit gene expression by assembling corepressors and histone deacetylases (HDAC) [33]. PPAR β/δ activity plays a key role in the regulation of a wide variety of metabolic processes, but has also been implicated in the development of chronic diseases such as obesity, atherosclerosis, diabetes and cancer [34]. The switch towards oxidative muscle fibers triggered by PPAR β/δ and the concomitant switch of fuel source from glucose to fatty acids [35] can at least in part be explained by the epistasis with PGC-1 α . PPAR β/δ not only is an upstream regulator of PGC-1 α , but also a transcription factor binding partner [36], even though PGC-1 α function is not dependent on PPAR β/δ [37].

Based on its effect on skeletal muscle and the presence of a well-characterized ligand-binding domain, PPAR β/δ is an interesting pharmacological target for muscular and metabolic diseases. GW501516, better known as Cardarine ¹ or Endurobol ² amongst performance-seeking athletes ³, is a well-characterized PPAR β/δ agonist with promising effects on skeletal muscle, yet it is an example of a failed “exercise mimetic”. In mice, GW501516, either when combined with exercise or at higher doses by itself, induces some hallmarks of EE adaptation such as mitochondrial biogenesis, fatty acid oxidation, an

¹<https://www.evolutionary.org/cardarine-gw-501516-explained> (Accessed 07.07.2017)

²<https://runswimthrowheat.blogspot.ch/2014/02/gw501516-endurobol-and-doping-whats-all.html> (Accessed 14.08.2017)

³<https://www.steroid.com/Cardarine.php> (Accessed 07.07.2017)

oxidative fiber-type switch and improved insulin sensitivity via AMP-activated protein kinase (AMPK) [35]. GW501516 has also been proposed as a treatment for DMD by stimulating the expression of utrophin A and restoring sarcolemmal integrity in mature *mdx* mice (an X chromosome-linked mouse mutant that exhibits histological lesions characteristic of muscular dystrophy) [38]. To its detriment however, tumorigenic effects of GW501516 have been reported and development was discontinued by Glaxo in Phase II clinical trials [39]. Despite these important issues, GW501516/Cardarine/Endurobol is openly available on the black market and its side effects are deceptively downplayed^{1,2,3}. Accordingly, the World Anti-Doping Agency (WADA) has issued an unusual warning about its toxicity to athletes looking for potential performance enhancements⁴.

AMP-activated protein kinase (AMPK)

AMPK is a known upstream regulator of skeletal muscle PGC-1 α and is involved in the initiation of mitochondrial biogenesis [40]. AMPK serves as a sensor of cellular energy levels as it is activated by a low ATP to AMP ratio, low glucose or glycogen [41]. Once activated, AMPK promotes processes to rectify this dysbalance, e.g. by enhancing glucose transport and fatty acid oxidation through GLUT4 transport to the sarcolemma [42] and inhibition of acetyl-CoA carboxylase [43], respectively. Importantly, AMPK was shown to be indispensable for the activation of PGC-1 α and calcium/calmodulin-dependent protein kinase type IV (CaMKIV) in response to chronic energy deprivation [40]. Accordingly, pharmacological activation of AMPK by 5-aminoimidazole-4-carboxamide ribonucleotide (AICAR) improved endurance capacity in mice by over 40% [44]. However, as part of the catabolic program controlled by AMPK, AICAR also increases the expression of E3 ligases and thereby promotes protein degradation [45]. Therefore, it is unclear whether a beneficial outcome can be achieved in humans with acceptable side effects caused by a signaling pathway that primarily activates catabolic processes [46]. Like GW501516, AICAR has been banned by WADA and added to the list of prohibited substances in sports⁵. Of note, the pan-AMPK activator MK-8722 has recently been tested in different animal models and even though an improvement in glucose homeostasis was observed, the concomitant cardiac hypertrophy would be highly troubling, particularly for athletes [47].

Mammalian target of rapamycin (mTOR)

The mTOR signaling pathway plays a central regulatory role in many cellular processes and is implicated in a large number of pathological conditions and diseases [48]. mTOR is a highly conserved serine/threonine protein kinase that forms two structurally and functionally distinct complexes: mTOR Complex 1 (mTORC1), which governs anabolic processes such as protein synthesis in response to nutrients or RE and mTOR Complex 2 (mTORC2), which controls many other cellular processes and has recently been shown to regulate muscle glucose uptake during EE [49]. Not surprising based on the activation by RE, mTORC1 signaling plays a fundamental role in the hypertrophic response of skeletal muscle [3]. mTORC1 also influences mitochondrial function by regulating mitochondrial oxygen

⁴<https://www.wada-ama.org/en/media/news/2013-03/wada-issues-alert-on-gw501516> (Accessed 07.07.2017)

⁵<https://www.wada-ama.org/> (Accessed 07.07.2017)

consumption and oxidative capacity through the cooperative modulation of the activity of the transcription factor yin-yang 1 (YY1) together with PGC-1 α [50]. At the moment, rapamycin and rapalogs are the only clinically approved pharmacological mTOR modulators used for immunosuppressive and cancer treatment [51]. Experimentally, rapamycin-induced inhibition of mTORC1 enhances longevity in mice [52]. Interestingly, inhibition of the rapamycin-sensitive mTOR component only partially blunted the activation of muscle protein synthesis and the hypertrophic response following chronic RE [53], suggesting additional rapamycin-insensitive mechanisms to contribute to this exercise response. In genetic models for sustained, skeletal muscle-specific inhibition or activation of mTORC1, pathological consequences were reported [54, 55]. It thus is unclear if and how therapeutic effects could be achieved with a pharmacological activator of mTOR.

Dietary supplementation in health and disease

As outlined above, pharmacological targeting of known regulators of exercise adaptation in skeletal muscle is still in its infancy. Other compounds with a known effect on muscle size and function include many drugs that are banned as doping and in most cases exhibit unacceptable side effects in non-substitution therapy, such as anabolic steroids or β 2-adrenergic receptor agonists. Dietary supplements constitute a third category of substances that are used to boost muscle performance, even though the true efficacy of dietary supplements to improve exercise performance is often highly disputed. Since these types of compounds are not regulated by the FDA, purity and dose often are difficult to estimate in over-the-counter formulations. Nevertheless, we will discuss some of the most sought-after dietary supplements that are prescribed to patients and used by athletes to increase strength, skeletal muscle mass, or overall well-being. The effects of macronutrients, micronutrients (such as vitamins and antioxidants), and nutraceuticals on skeletal muscle mass, metabolism and exercise adaptations are not a focus of this review and will therefore not be covered in detail (see [56] for more details on these aspects).

Creatine (Cr)

With hundreds of studies over the last decades, Cr is one of the most studied dietary supplements that is readily available on the market, inexpensive and routinely used by recreational and professional athletes. Cr is a naturally occurring amine that is synthesized endogenously by the liver, pancreas and kidney from the amino acids glycine, methionine and arginine at a rate of roughly 2 g per day [57], or taken up in the diet, mostly found in meat and wild fish (Cr uptake of < 5 g/day). Cr enters muscle cells against a concentration gradient via the Cr transporter (CreaT) [58]. The majority of Cr content in the body is found in skeletal muscle (>90 %) and lower amounts are found in the testicles, brain and bones [57].

Oral Cr supplementation leads to an increase in the mean phosphocreatine (PCr)/inorganic phosphate ratio in the skeletal muscle [59]. PCr anaerobically donates a phosphate group to ADP to form ATP during the first 10 seconds of an intense muscular effort. ATP can be resynthesized a dozen times faster from PCr compared to oxidative phosphorylation and almost a hundred times faster compared to *de novo* pathways [60]. Increased muscular PCr

levels upon oral Cr supplementation thus may increase exercise performance. Cr supplementation could also improve recovery by reducing muscle cell damage and inflammation following exhaustive exercise, as shown in finishers of a 30 km race [61].

Oral Cr supplementation also improved heavy RE performance in various studies, presumably by enabling higher quality training sessions (enhanced average lifting volume per session) [62]. Overall increases in fat-free mass and measures of strength, such as squat and bench press performance, were observed [62]. The observed boost in fat-free mass and strength was linked to an elevated intracellular water content and cell swelling, which in itself could serve as an anabolic stimulus [63, 64]. Short-term Cr supplementation modulated transcript and protein levels of genes and kinases involved in osmosensing, protein and glycogen synthesis, satellite cell proliferation and other signal transduction pathways [63]. Accordingly, it is thought that at least some of the weight gain associated with Cr supplementation is due to increased total body water retention and not necessarily muscle mass [64]. The average boost in muscle strength when combining RE with Cr ingestion compared to a placebo was reported to be around 8% [65]. However, the response to Cr supplementation is highly variable in different individuals [65], which may explain the contradictory results found in some studies where Cr supplementation showed no effect on RE performance or the anabolic response [66].

Creatine in Duchenne Muscular Dystrophy

DMD is a genetic disorder characterized by progressive muscle degeneration and weakness caused by the absence of *Dystrophin*, a structural protein that contributes to protection from mechanical stress [67]. Oral Cr supplementation of 5 g/day for 8 weeks significantly increased the mean PCr/inorganic phosphate ratio in young boys suffering from DMD [59]. Patients reported a subjective improvement by Cr supplementation compared to placebo and their muscle strength was preserved, at least in the short term. However, the study provided no evidence for a long-term effect of the treatment [59]. In another study, four months of Cr supplementation led to an increase in fat-free mass and grip strength, as well as to a reduction in a marker of bone breakdown in boys affected by DMD [68]. Several other studies provide further evidence for a positive effect of Cr supplementation on bone mineral density, especially when combined with exercise or increased locomotion [69–71]. Furthermore, it has been reported that Cr improves mitochondrial function and prevents calcium buildup in a mouse model for DMD (*mdx* mice) [72]. As oral Cr is generally well-tolerated, supplementation strategies may provide symptomatic relief for patients affected by muscular dystrophies such as Duchenne and Becker Muscular Dystrophy, a milder form of DMD.

Creatine in type 2 diabetes mellitus

When combined with an exercise program, Cr supplementation (5 g/day) improves glycemic control in T2DM patients, most likely due to an increase in recruitment of GLUT4 to the sarcolemma [73]. Notably, Cr supplementation in combination with exercise also enhanced glucose metabolism by the same GLUT4-dependent mechanism in an immobilization/rehabilitation study [74]. It is important to stress that participants of studies that failed to combine Cr supplementation with exercise showed no improvements in insulin sensitivity

and glucose tolerance, despite an elevated muscle Cr content [75]. Interestingly, increased muscle glycogen storage was observed, yet GLUT4 mRNA and protein content remained unchanged [75]. Collectively, the current research suggests that Cr supplementation, normally well-tolerated, could be a tool for ameliorating T2DM, but only when combined with exercise. This poses a limitation on its use for the treatment of obese and elderly individuals, where sufficient levels of physical exertion might not be achieved, thus rendering Cr supplementation potentially ineffective.

Citrulline malate and L-citrulline

The amino acid ornithine merges with carbamoyl phosphate, forming the amino acid citrulline in the urea cycle [76]. Citrulline, in particular when supplemented orally, accelerates ammonia clearance, which accumulates in working muscles during exercise, and thereby ameliorates fatigue [77]. Malate is an organic salt often used as a food preservative and as a supplement, the effect of malate aims at improving the stability of citrulline by a direct interaction. Citrulline malate (CM) mitigates the build-up of excess lactate, acidosis and hyperammonaemia in the blood [78]. Even though accumulation of lactate and the acidosis resulting from the associated protons might not be involved in causing muscle fatigue as previously proposed [79], accumulation of ammonia is a less ambiguous fatigue factor due to the inhibitory effect on cellular energy processes. A single dose of CM (8 g) has been shown to increase levels of arginine and ornithine in the body, improve exercise performance and reduce muscle soreness 24-48h post RE [80], indicating that CM could elicit ergogenic effects. Moreover, CM allowed for significantly more repetitions in the later sets (4 or more sets) of intense RE, where muscular fatigue, and not strength itself becomes the limiting factor. This increase in total repetitions leads to an enhanced effective training volume and may thus contribute to a better training stimulus. In a recent study, however, acute CM supplementation (12 g) did not improve the repeated high-intensity cycling performance or time-to-exhaustion of well-trained males [81]. Interestingly, oral L-citrulline supplementation reduced completion time of a cycling trial by 1.5% in healthy trained men compared to placebo controls and increased plasma L-arginine levels [82]. L-citrulline significantly improved the subjective perception of concentration and muscle fatigue immediately after exercise [82]. Therefore, whether acute CM (or L-citrulline alone) supplementation increases work capacity in high-intensity anaerobic exercise with short rest times remains debatable. Chronic CM supplementation over 15 days (6 g/day) enhanced skeletal muscle power output and lead to a lower pH-to-power ratio, eliciting improved oxidative energy turnover [83]. Unfortunately, only a few long-term studies investigating the ergogenic effects of continuous CM supplementation on exercise adaptations have been conducted so far and further research in this area is required. However, despite research literature being inconclusive and sparse, CM supplementation enjoys a huge popularity amongst strength and endurance athletes and has been coined the “Fatigue Fighter” in popular online media⁶.

⁶<https://www.bodybuilding.com/fun/citrulline-malate-the-fatigue-fighter.html> (Accessed 07.07.2017)

Citrulline and mitochondrial diseases

Citrulline and the amino acid arginine are nitric oxide (NO) precursors. Of note, oral L-citrulline supplementation leads to a greater boost in arginine levels than arginine supplementation itself [84]. A deficiency in NO plays a major role in the maternally inherited mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes syndrome (MELAS) [85]. This mitochondrial disorder is believed to result from several interacting mechanisms, including microvasculature angiopathy and impaired mitochondrial energy production [85]. Children with MELAS have lower arginine and citrulline flux, plasma arginine, and NO production [86]. L-Citrulline supplementation increases all of these parameters [86]. Thus, long-term studies investigating the role of NO precursors like L-citrulline in the treatment of mitochondrial diseases should be conducted to explore their therapeutic potential.

β -hydroxy- β -methylbutyric acid (HMB)

HMB is a metabolite derived from leucine and its ketoacid α -ketoisocaproate. Much like Cr, HMB is naturally biosynthesized in humans and is a popular dietary supplement with the potential to increase lean body mass, muscular strength and size, reduce skeletal muscle damage, and enhance speed of recovery post-exercise [87]. A daily dose of 3 g of HMB helps to maintain or even gain muscle mass and was deemed relatively safe in the long-term, even though safety in a disease context has been questioned [88]. HMB (3 g/day over 6 weeks of daily training and supplementation) has been used successfully to reduce muscular damage in young endurance athletes measured by a reduction in circulating creatine kinase (CK) and lactate dehydrogenase (LDH) after a 20 km run [89]. Furthermore, acute pre-exercise HMB free acid-supplementation (3 g) improved markers of exercise-induced damage in RE-trained men [87]. In RE, acute HMB supplementation also improved the perceived recovery status of participants, implying that pre-exercise HMB supplementation might be a good strategy to improve training frequency due to quicker recovery from exercise bouts. However, 9 weeks of 3 g/day HMB coupled with RE training had a negligible effect on overall body composition of RE-trained men, but substantially increased lower-body strength measures (9.1% leg extension 1 RM increase) [90]. Thus, despite results suggesting small benefits, the overall effect of HMB on RE is inconclusive.

HMB in fiber atrophy pathologies

In the context of disease, HMB most prominently ameliorates proteolysis and leads to increased lean body mass in patients, e.g. in cancer/AIDS-related cachexia, immobilization, sepsis and steroid medication (reviewed in [91]). Furthermore, 12 weeks [92] or even 1 year [93] of daily HMB supplementation (1.5 to 5 g per day) counteracted sarcopenia in elderly people by increasing muscle mass, at least in combination with Arginine (Arg) and/or Lysine (Lys). However, strength increases were only observed when the HMB/Arg/Lys-cocktail was supplemented with Vitamin D, indicating a synergistic effect [93].

In conclusion, HMB supplementation shows beneficial effects in healthy, exercising subjects, in certain diseases where proteolysis occurs, and in the elderly, especially when combined with other nutrients such as Arg, Lys, and Vitamin D.

”Exercise mimetics” as a novel approach

Since the efficacy of existing drugs, supplements and nutraceuticals in muscle diseases remains small, novel therapeutic avenues are considered to remedy the situation of skeletal muscle to constitute one of the most undermedicated organs. In recent years, the concept of designing “exercise mimetics”, pharmacological compounds that recapitulate the many beneficial effects of EE and/or RE, has been proposed [44]. However, this concept is highly controversial and the “exercise mimetics” that have been tested so far predominantly remain in the experimental stage in rodents, with little evidence of feasibility and efficacy in humans [46]. Indeed, the pleiotropic effects of exercise in muscle and other organs are difficult to reconcile with the consequences that can be elicited by a single compound, thoroughly discussed in an excellent review by Booth and Laye [94]. Importantly, in contrast to the high safety of *bona fide* exercise, even drugs with limited beneficial effects on physical performance entail considerable adverse effects, such as in the case of erythropoietin [95]. The use and abuse of anabolic steroids in an effort to increase muscle mass, aesthetics, and muscular strength, while being extremely effective [96], triggers serious side effects, which in the worst case can be lethal [97]. Furthermore, many of the effects of “exercise mimetics” observed in rodents could not be recapitulated in humans, at least to the same extent. For example, the polyphenol resveratrol induces an endurance-trained state with the corresponding metabolic adaptations in mice, but has little or no effect in humans (summarized in [46]). It is currently unclear whether inherent differences between mice and men are responsible for these discrepancies e.g. in absorption, biotransformation and elimination of xenobiotics, the significantly higher metabolic rate of mice, or the extreme difference in life span and hence the aging process. Alternatively, as outlined by Laye and Booth [94], the experimental setting used to test these compounds in rodents might also confound the findings vis-à-vis translation into the human context. In most home cages, mice are in a state of sedentariness, and training interventions might not reflect true exercise, but merely reconstitute the normal activity levels of these rodents in the wild. In other words, the “control” group in rodent studies may be inadequate to capture the normal, basal state of human physiology and therefore result in a divergent outcome in regard to so-called “exercise mimetics”.

However, even the concept of a hypothetical “exercise mimetic” for human use could be questioned for many reasons [94]. For example, the human body has evolved to handle periods of strenuous physical activity, yet inadvertently has to compensate with rest and regeneration. This poses a huge challenge, as accurate pharmacological activation of exercise pathways to an adequate amplitude and time is difficult to achieve. Initiating and maintaining a metabolic overdrive for prolonged periods of time could potentially result in dangerous consequences as constant activation of exercise pathways likely induce a chronic catabolic state due to the inhibition of protein synthesis and activation of autophagy, e.g. by activation of AMPK and the ensuing inhibition of mTOR. Experimentally, long-term activation of mTORC1 in a transgenic mouse model leads to the development of a severe late-onset myopathy related to impaired autophagy [54]. Inversely, chronic inhibition of mTORC1 activity likewise triggers a myopathic phenotype [55], underlining the importance of adequate exercise and rest/recovery cycles.

Future directions

For the time being, changes in life style that entail physical activity and a balanced diet remain the most effective, affordable, and accessible form of treatment and prevention of chronic diseases [1], however with the caveat of compliance and exercise intolerance in some patients. “Exercise mimetics” remain an intriguing concept, even though a more limited scope might have to be aimed for. Thus, a partial activation of specific exercise pathways could be a promising approach to prevent and treat individual diseases, in particular in patient populations with a compromised exercise tolerance or a reduced ability to train. Optimally, these types of interventions, either by drugs or dietary supplements, should however be combined with *bona fide* physical activity whenever possible. For example, the paradoxical acceleration of the development of insulin resistance in high fat diet-fed PGC-1 α skeletal muscle-specific transgenic animals [23] was reversed by concomitant training [98]. Whatever pharmacological strategy intended for disease treatment will emerge, the potential of misuse as performance-enhancing aids by athletes (or even non-athletes!) exists, paving the road for difficult-to-control doping strategies under the veil of treatment. Nevertheless, these considerations should not negatively influence the development of novel treatment strategies that are needed in muscular dystrophies, sarcopenia, cachexia and other severe pathological contexts for which no therapy exists at the moment.

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Non-standard abbreviations

1 RM	one repetition maximum
CM	citrulline malate
Cr	creatine
EE	endurance exercise
HIT	high intensity training
HMB	β -hydroxy- β -methylbutyric acid
MELAS	maternally inherited mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes syndrome
PCr	phosphocreatine
RE	resistance exercise
T2DM	type 2 diabetes mellitus

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Fig. 1. Different exercise modalities and pharmaceutical compounds modulate mitochondrial function.

Endurance exercise leads to the activation of AMPK and consequently to increased coactivation of PPARβ/δ by PGC-1α, resulting in improved mitochondrial oxidative function. Different pharmacological compounds may activate this pathway even in absence of endurance exercise and at least partially confer endurance performance benefits. Resistance exercise activates mTORC1 and increases coactivation of YY1 by PGC-1α, presumably via phosphorylation of S6K (ribosomal protein S6 kinase). This also affects mitochondrial function. Rapamycin and other pharmacologic inhibitors of mTORC1, but also exercise interference can blunt this response. Exercise interference is a controversially discussed mechanism by which concurrent endurance exercise may negatively impact adaptations to resistance exercise by decreasing mTORC1 activity. AICAR, R419, Cpd14, Mk-8722, and Metformin are (potential) pharmacologic AMPK activators. GW501516 is a known pharmacologic PPARβ/δ activator. Arrows indicate activation, dashed lines indicate inhibition.

Fig. 2. Pharmacological modulation of PGC-1α acetylation affects the interaction with transcription factor bindings partners.

PGC-1α expression is increased in the diabetic liver, where it interacts with and coactivates HNF-4α to boost expression levels of the gluconeogenic genes PCK1 and G6PC. The result is increased hepatic glucose production that negatively effects the diabetic phenotype. The compound SR-18292 reduces the interaction between PGC-1α and HNF-4α by increasing PGC-1α acetylation, resulting in lower PCK1 and G6PC levels, reduced hepatic glucose production, and thus amelioration of the diabetic phenotype. SR-18292, as well as similar

compounds modifying hepatic PGC-1 α -HNF-4 α interactions, could therefore prove to be effective treatment options in T2DM.