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NOVEL INVESTIGATIONAL DRUGS MIMICKING EXERCISE FOR THE TREATMENT OF CACHEXIA

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Abstract

Introduction: Cachexia is a syndrome characterized by body weight loss, muscle wasting, and metabolic abnormalities, that frequently complicates the management of people affected by chronic diseases. No effective therapy is actually available, although several drugs are under clinical evaluation. Altered energy metabolism markedly contributes to the pathogenesis of cachexia; it can be improved by exercise, able to both induce anabolism and inhibit catabolism.

Areas covered in this review: This review will focus on exercise mimetics and on their potential inclusion in combined protocols to treat cachexia, with particular reference to the cancer-associated one.

Expert opinion: Despite exercise improves muscle phenotype, most patients retain sedentary habits quite difficult to disrupt. Moreover, they frequently present with chronic fatigue and co-morbidities that reduce exercise tolerance. For these reasons drugs mimicking exercise could be beneficial also in the absence of patient compliance to the practice of physical activity. Different exercise mimetics are now available and, yet some of them may exert serious side-effects, investigating their effectiveness on muscle phenotype will provide a new tool for the management of cachexia.

1. Introduction

Cachexia is a multi-organ syndrome that frequently occurs in chronic pathologies such as autoimmune or neurodegenerative diseases, sepsis, diabetes and cancer. The etiopathogenesis of cachexia is multifactorial, including reduced caloric intake, altered energy and protein metabolism, enhanced lipolysis, systemic inflammation, perturbations of the hormonal homeostasis. Among the different features that characterize the onset and progression of cachexia, body weight loss and inflammation are the most relevant [(1)].

In cachexia, both skeletal muscle mass and function are markedly reduced. Muscle nitrogen balance is regulated by a complex network of factors, such as humoral mediators, nutrient and energy availability, or contractile activity, all of which concur to unbalance protein turnover rates, favoring degradation. Ca^{2+}-dependent, ATP-ubiquitin-dependent and autophagic lysosomal pathways have all been shown to contribute to muscle protein wasting [(2)]. Since the discovery and characterization of different muscle-specific ubiquitin ligases[(3)], attempts have been made to antagonize muscle depletion by targeting these enzymes. The results, however, do not allow to validate ubiquitin ligases as therapeutic targets for muscle wasting. Not only, specific systemic inhibition of the different proteolytic pathways does not appear as an effective strategy to contrast the progression of cachexia. This has been shown for proteasome inhibitors such as bortezomib[(4)(5)], while suppression of autophagy may even reveal detrimental[(6)(7)(8)]. The possibility that muscle-specific inhibition of proteolysis might prove effective remains an open question, however.

In various forns of cachexia muscle protein wasting is associated with an unbalance between pro- and anti-inflammatory cytokines, due to chronic inflammation. For example, circulating levels of Tumor Necrosis Factor (TNF)α and TNF soluble receptors are increased in patients affected by cancer, chronic renal or heart failure[(9)(10)(11)], while mice lacking the anti-inflammatory IL-10 display muscle depletion and weakness[(12)]. Pro-inflammatory cytokines, in particular, play a pivotal role in modulating muscle protein turnover. Systemic inflammation is associated with enhanced rates of protein degradation, that can be paralleled or not by decreased protein synthesis. Cytokine-induced activation of the transcription factors Nuclear Factor (NF)-κB and Signal Transducer and Activator of Transcription (STAT)3 results in muscle atrophy[(13)(14)]. Several studies report that anabolism can also be affected by pro-inflammatory cytokines. As an example, TNFα down-regulates anabolic signaling pathways such as those activated by insulin and Insulin-like Growth Factor (IGF)-1[(15)]. Particularly relevant to muscle mass homeostasis

are the insulin receptor substrate (IRS)-1/PI3K/Akt axis[16] and the signaling pathways dependent on myostatin[15][17] or on bone morphogenetic proteins[18], all of which are subject to cytokine-mediated modulations.

2. Therapeutic approaches to cancer cachexia

Cachexia develops in a large proportion of cancer patients, being estimated to account for about 20% of cancer-related deaths[1]. Its occurrence markedly complicates patient management, negatively impinging on the tolerance and response to antineoplastic treatments, and on the quality of life as well. Reverting overt cachexia, however, definitely is not an easy task, and the most likely scenario is to develop strategies aimed at delaying its onset. To achieve this goal, early stages of cachexia must be identified. Along this line, Fearon and collaborators[15] proposed a classification that defines three progressive stages (pre-cachexia, cachexia, and refractory cachexia), aimed to help clinicians to start the few available treatments as soon as possible.

Studies performed on experimental models have provided the basis for potential treatments of human cachexia, some of which are in phase I or II clinical trials. Provided the multifactorial nature of this syndrome, the rationale of such approaches ranges from nutritional support to the inhibition of specific molecular pathways.

The early institution of a personalized nutritional counseling is recommended to improve patient nutritional status, treatment tolerance and clinical outcome. Indeed, the maintenance of patient body weight by nutritional interventions allows the completion of chemotherapy, frequently interrupted because of severe side effects[19]. In this regard, several agents have been tested to treat anorexia and increase appetite, such as corticosteroids, progestins (megestrol acetate), cannabinoids, ghrelin and its analogues (anamorelin), branched-chain amino acids[20].

Since systemic inflammation frequently occurs in cancer patients (see above), several anti-inflammatory drugs have been tested in experimental cachexia: steroids and Non-Steroidal Anti-Inflammatory Drugs (NSAIDs), anti-cytokine agents (pentoxifylline, thalidomide), anti-cytokine (IL-6, TNF) antibodies, ω-3 polyunsaturated fatty acids. Most of these agents, however, have not been validated for the clinical use, although some of them (NSAIDs, ω-3 polyunsaturated fatty acids) reached the clinical evaluation[20].

The metabolic alterations underlying cancer cachexia point towards enhanced catabolism and reduced anabolism. Accordingly, anabolic and/or anticatabolic strategies have been adopted to modulate these processes and attempts have been made to modulate the Growth Hormone (GH)/insulin/IGF-1 axis, the androgen-dependent signaling (anabolic-androgenic steroids, selective androgen receptor modulators), or the availability of amino acids and their metabolites (branched-chain amino acids, β-hydroxy-β-methylbutyrate, glutamine)[20].

Finally, based on the mechanisms contributing to cancer-induced cachexia, some experimental drugs are currently under investigation, including anti-myostatin agents, β-adrenergic agonists, mitogen-activated protein kinase kinase (MEK), and extracellular signal regulated protein kinase (ERK) inhibitors, Interleukin (IL)-15. Although encouraging, in some cases at least, at the moment the results do not favor the possibility of their rapid translation into the clinical practice.

3. Novel aspects in the pathogenesis of cachexia: myogenesis and mitochondria

In the last few years two aspects have added complexity to the pathogenesis of cachexia: 1) the reduced myogenic potential; 2) the altered mitochondrial structure and function.

3.1 Impaired myogenesis

Myogenesis allows the development of skeletal muscle during the embryonal life. In the adulthood it drives the repair of injured muscles and the maintenance of physiological muscle cell renewal. After damage, myofiber degeneration is compensated by the regeneration of new fibers afforded by proliferation and subsequent fusion of resident myogenic precursors, mainly satellite cells (SCs)[21]. Impaired myogenesis has been associated with pathological muscle wasting. The levels of Pax7, a marker of SC proliferation, and those of myogenin, an indicator of ongoing differentiation, respectively increase and decrease, respectively, in the muscle of mice bearing the colon 26 carcinoma (C26) as well as in cancer patients, suggesting that myogenic precursors accumulate in the skeletal muscle of tumor hosts[22][23][24]. This process appears to depend on the maintenance of elevated Pax7 expression, apparently related to activation of the

transcription factor NF-κB[23]. Impaired myogenesis is associated with enhanced signaling through the stress kinase ERK, which is known to maintain cells in an undifferentiated state[22]. Indeed, treatment of C26-bearing mice with PD98059, a MEK inhibitor able to block ERK activation, restores the physiological levels of both Pax7 and myogenin expression[22]. In agreement with these results, muscle gain has been observed in cholangiocarcinoma patients assuming selumetinib, another MEK inhibitor[25]. Finally, the administration of the multikinase inhibitor sorafenib has been shown to improve both muscle wasting and physical activity in mice hosting the C26 or the Lewis Lung Carcinoma (LLC). Such effects are associated with modulation of both STAT3 and ERK activity in the muscle, leading to reduced expression of Pax7 and atrogin-1[26]. Further investigations in this direction are warranted to clarify if ERK can be a suitable therapeutic target in cancer cachexia.

3.2 Impaired mitochondrial function

Since mitochondria are the main source of the energy required for contraction, alterations in their homeostasis markedly affect muscle function. While the concept of energy wasting is not new in the literature on cancer cachexia[1], the underlying mechanisms became a hot topic just recently. Ultrastructural alterations have been reported in muscle mitochondria of animals bearing the C26[27], the LLC (Pin, Busquets et al., under revision) or the Yoshida Ascited Hepatoma (AH)-130 tumors[28]. Mitochondrial uncoupling occurs and oxidative capacity decreases in the muscle of tumor hosts[29][30], consistently with the occurrence of a shift from oxidative to glycolytic fibers (Pin, Busquets et al., under revision). Humoral mediators contribute to mitochondrial impairment. As an example, TNFα-induced activation of NF-κB reduces muscle oxidative capacity and negatively regulates the expression of molecules relevant to mitochondrial biogenesis[29]. C2C12 myotubes cultured in the presence of LLC cell conditioned medium show alterations of the electronic flow in the electron transport chain[31] and finally, muscle wasting in the ApcMin/+ mice, an experimental model of IL-6 driven cancer cachexia, is associated with altered expression of proteins regulating mitochondrial biogenesis and fusion[32].

A relation between mitochondria dynamics (balance between fission and fusion processes) and protein breakdown in the skeletal muscle has been previously

described[33][34]. Mitochondrial dysfunction, leading to oxidative and energy stress, would lead to hyperactivation of muscle proteolytic systems through adenosine monophosphate-activated protein kinase (AMPK)- and Forkhead transcription factor (Fox)O-dependent pathways[33], ultimately resulting in muscle wasting.

4. Importance of exercise in the management of cachexia

Most of the ongoing clinical trials are based on the use of nutritional and pharmacological interventions aimed at preventing the loss of body weight and muscle mass. However, muscle 'quality' is as important as muscle 'quantity'. Both aspects could be targeted by exercise training. Indeed, stimulating the increase of muscle mass and strength, it might improve cancer-induced wasting by both activating anabolic pathways and down-regulating the activity of pro-inflammatory cytokines[35].

Two different exercise modalities can be chosen: 1) resistance exercise, that causes a mechanical strain on the muscle resulting in different adaptations, among which hypertrophy, that occurs primarily through the accretion of contractile proteins, increases the capacity to generate force; and 2) endurance exercise, that generally leads to metabolic adaptations such as increased number of mitochondria, fiber-type shift to an oxidative profile and increased capillary density, with little changes in the skeletal muscle mass[36].

Endurance exercise increases the expression of the peroxisome proliferator-activated receptor (PPAR)-γ coactivator-1α (PGC-1α), a transcriptional co-regulator that binds to several transcription factors to promote mitochondrial biogenesis and oxidative metabolism[37]. Moreover, exercise also induces the expression of genes involved in mitophagy[38], a process that accounts for the clearance of damaged mitochondria. Consistently, PGC-1α has been recently shown to regulate mitophagy in the skeletal muscle[39], while the reduced mitochondrial content reported in ApcMin/+ cachectic mice is associated with down-regulation of PGC-1α and mitochondrial fusion proteins[32]. Finally, both resistance and endurance exercise appear to stimulate the proliferation of SCs, promoting myogenesis (see above)[40][41].

Taking into account that at least some of the metabolic changes occurring in the skeletal muscle of cancer patients (alterations of mitochondrial structure, reduced ATP

synthesis, shift from oxidative to glycolytic metabolism, impaired myogenesis)[1][42] can be rescued by physical training, a combination of endurance and resistance exercise would probably be the right choice to improve cachexia.

5. Drugs mimicking exercise

At present three main classes of drugs mimicking the effects of aerobic/endurance exercise have been described: agonists of PPARδ or of AMPK and molecules able to activate silent-information-regulator-two-protein (SIRT)1. In addition, hyperexpression of PGC-1α could be an effective strategy, too.

PPARδ, the most abundant PPAR in the skeletal muscle, is involved in the regulation of lipid metabolism, mitochondrial function and fiber-type determination. Its expression in the skeletal muscle is induced by exercise. As a consequence of its activation the energy source is switched from glucose to fatty acids and myofiber phenotype converted from fast (glycolytic) to slow (oxidative) twitch. Muscles in mice overexpressing PPARδ are characterized by increased proportion of oxidative fibers, enhanced activity of enzymes involved in the oxidation/phosphorylation reactions, and increased mitochondrial biogenesis and uncoupling proteins[43]. Moreover, constitutive hyperactivation of PPARδ results in increased exercise capacity and in protection against diet-induced obesity or type 2 diabetes[44]. By contrast, lack of PPARδ leads to an opposite muscle phenotype, characterized by slow-to-fast myofiber conversion and down-regulation of gene products involved in fatty acid oxidation[45].

Among the various PPARδ agonists presently available, the most widely studied is GW501516, which has been shown to activate PPARδ in an exercise-like manner, although not reproducing the increased mitochondrial gene expression and function or the switch of fiber type[46][47]. Increased fat *versus* carbohydrate oxidation rate, up-regulation of mitochondrial biogenesis and increased expression of genes encoding contractile proteins characteristic of type I myofibers can be observed in mice after few days of synthetic PPARδ agonist administration to mice[44]. Moreover, both angiogenesis and biogenesis of mitochondria are induced in mice 24 h after GW0742 administration, while a fiber type switch is evident after 48h[48]. Finally, myocyte cultures exposed to PPARδ agonists show increased glucose and fatty acid metabolism, with enhanced expression and activation of AMPK and p38[48].

The potential of small molecule PPARδ agonists to treat various pathological conditions is highlighted by several studies. As an example, GW501516 improves experimental atherosclerosis in apoE$^{-/-}$ mice[49], while its administration to obese mice reduces weight gain and increases both resting energy expenditure and peripheral insulin sensitivity[50]. Just few studies investigate PPARδ agonist effects in humans. The results suggest that these drugs could reduce obesity, insulin resistance and hyperlipidemia. Indeed, treatment with GW501516 results in improved circulating apolipoproteins, LDL and HDL cholesterol and hepatosteatosis[51]. Finally, administration of MBX-8025 to moderately obese subjects improves circulating lipid and cholesterol profiles[52]. However, very few data are available to define if PPARδ agonists can also affect muscle phenotype. GW501516 treatment improves muscle dystrophy by down-regulating inflammation-dependent pathways[53]. The same drug attenuates metabolic abnormalities and improves muscle phenotype in experimental models of cardiovascular disease[48]. A specific class of PPAR agonists, namely glitazones, known for their insulin sensitizing action in the skeletal muscle, appear promising to treat muscle atrophy, but their clinical use is controversial. Data obtained on a genetic model of diabetes, the *db/db* mice, show that rosiglitazone improves insulin resistance and Akt-dependent signaling pathway, while it inhibits both muscle proteasome and caspase 3 activities[54]. In addition, a transient muscle-sparing effect associated with normalization of PPARδ expression has been reported in C26-bearing mice treated with rosiglitazone[55][56]. Despite the above described positive effects, however, elderly obese patients practicing resistance training and assuming pioglitazone show both visceral fat and muscle depletion[57], while contradictory results have been reported also for other PPARδ agonists. Treatment of L6 myotubes with the GW0742 increases protein degradation and reduces myotube size, and rats administered the PPARδ inhibitor GSK0660 are resistant to glucocorticoid or sepsis-induced muscle protein breakdown[58]. These findings are in line with previous data showing that PPARδ expression is induced in the skeletal muscle of tumor-bearing rats[59]. A final consideration is that glitazones show several severe side effects, including cardiovascular and hepatic adverse events and increased risk of cancer development[60]. For these reasons, rosiglitazone has been withdrawn from the european market by EMA, while strict criteria for prescriptions and additional warning labeling are required by FDA[61].

AMPK activation results in modulations of the metabolic balance aimed to spare energy, mainly achieved by stimulating catabolism and inhibiting anabolism. For example, AMPK activation induces the expression of muscle-specific ubiquitin ligases and

stimulates autophagy[62][63]. Genetic AMPK inactivation in the muscle results in reduced mitochondrial mass, loss of type I fibers and reduced lipid metabolism. However, besides the pro-catabolic action of AMPK, the enhancement of its activity leads to increased oxidative metabolism and exercise efficiency[64]. In this regard, AMPK activation should be pursued in pathological states characterized by energy deficit, despite the concomitant occurrence of hypercatabolism.

The activation of AMPK can be achieved by drugs such as 5-aminoimidazole- 4-carboxyamide ribonucleoside (AICAR), metformin, β-guanidinoproprionic acid (β-GPA), resveratrol, quercetin, and salicylate[48]. Increased fatty acid oxidation and oxygen consumption, IIb-to-IIx myosin isoform shift, enhanced aerobic capacity and running endurance have been reported after AICAR administration to experimental animals[46]. AICAR-induced AMPK activation improves muscle phenotype in dystrophic *mdx* mice by activating autophagy[53]. The same drug reverts angiotensin II-induced muscle wasting, down-regulating the induction of the muscle-specific ubiquitin ligases and improving PGC-1α expression[65]. Chronic administration of β-GPA to experimental animals induces a fast-to-slow myofiber shift, increases PGC-1α expression and reduces muscle content of Receptor-interacting protein (RIP)140, a repressor of the oxidative phenotype (reviewed in[48].

As for metformin, few studies investigate its effects on muscle atrophy, both in humans and in experimental animals. Severely burned patients treated with metformin show reduced endogenous glucose production and increased glucose clearance and oxidation, with decreased glycemia. In parallel muscle wasting is improved, due to increased protein synthesis rather than to reduced breakdown[66]. Metformin appears to improve aging-related sarcopenia[67] and has been proposed to treat cancer-induced muscle wasting[68].

AMPK can also be activated by resveratrol, since many effects exerted on the muscle by this drug are attenuated by AMPK inhibition. Consistently, the increased mitochondrial biogenesis and endurance performance induced by resveratrol in wild type mice cannot be observed when the drug is administered to AMPKα_1 or -α_2 deficient mice[69]. Similarly, muscle wasting and reduced AMPK activation in IL-10 deficient mice can be improved by grape seed extract[70]. Finally, resveratrol administration to obese men activates AMPK and increases PGC-1α and SIRT1 protein levels, improving circulating lipid profile and inflammation[71].

Mammalian sirtuins (SIRT1–7) are a class of deacetylases involved in several processes such as metabolic regulation, cell death, autophagy, DNA repair and circadian rhythm. In addition, SIRTs appear crucial in lifespan regulation in lower eukaryotes, while not in healthy mice[72]. SIRT deregulation is involved in aging as well as in chronic pathologies such as obesity and cancer[73].

SIRT1 is the most extensively studied. It is involved in the modulation of various epigenetic mechanisms and also behaves as a critical regulator of energy homeostasis. SIRT1 positively modulates several processes such as cell survival, autophagic protein degradation, insulin production, regulation of mitochondrial mass, lipid metabolism and glucose homeostasis[73]. Consistent with its role as energy regulator, SIRT1 expression is induced in response to caloric restriction[73], and can be activated in the skeletal muscle by AMPK, linking a critical energy sensor to a regulator of aerobic capacity[74].

Mice genetically deficient of SIRT1 show inflammation and metabolic defects[75], while SIRT1 hyperexpression improves high fat diet-induced metabolic alterations[76]. Recent data show that muscle-specific SIRT1 overexpression causes a fast-to-slow fiber type transition. Such modification is paralleled by reduced gastrocnemius weight, which is consistent with the lower cross sectional area of oxidative compared to glycolytic fibers. However, transgenic animals are protected, partially at least, from fasting or denervation-induced atrogene hyperexpression[77]. Finally, muscle phenotype in *mdx*/SIRT1 double transgenic mice is improved[78].

Both natural and synthetic SIRT1 activators are available. Among the former, polyphenols, such as resveratrol, are the most studied. Resveratrol administration to mice increases increased muscle mitochondrial biogenesis and oxidative metabolism, resulting in enhanced exercise capacity and in protection against obesity and type 2 diabetes[79]. Resveratrol improves the lifespan of mice maintained on high fat diet[80] and is also beneficial in obese humans (see above). Part of the effects exerted by resveratrol derive from SIRT1-dependent modulations of PGC-1α acetylation state, and are recapitulated by selective SIRT1 activators[81].

Synthetic selective SIRT1 activators such as SRT2104 appear well tolerated in healthy adults and in elderly people[82][83]. SRT2104 administration to healthy volunteers improves plasma lipid profile and insulin sensitivity[82][84]. At present no studies address the effectiveness of synthetic SIRT1 activators on muscle mass and function in humans. Such

aspect has been recently investigated in an experimental study where short-term treatment with SRT2104 has been shown to attenuate muscle mass depletion induced by inactivity or fasting[85]. The protection exerted by SRT2104 on inactivity-induced muscle atrophy is associated with increased PGC-1α levels[85], consistent with previous observations showing that PGC-1α overexpression is able to protect against muscle atrophy induced by denervation or fasting[86].

PGC-1α is currently considered one of the major regulators of exercise-induced phenotypic adaptation in the skeletal muscle. It modulates mitochondrial biogenesis and its hyperexpression leads to increased mitochondrial mass and enhanced oxidative metabolism[87].

Both transcriptional and post-translational mechanisms contribute to regulate PGC-1α activity. As an example, endurance exercise leads to PGC-1α deacetylation, up-regulating the expression of several target genes. Exercise-induced activation of PGC-1α may also be influenced by different signaling pathways such as those dependent on Mitogen Activated Protein Kinases (MAPKs), oxidative stress, AMPK, SIRT1[87].

In the skeletal muscle PGC-1α participates in fiber-type specification, promoting a fast-to-slow transition. When specifically overexpressed in the muscle, it leads to a markedly oxidative phenotype characterized by high respiratory capacity and increased resistance to fatigue that result in improved exercise performance, without affecting muscle mass. Such phenotype closely resembles the one induced by aerobic training. By contrast, lack of PGC-1α results in decreased exercise performance and oxidative metabolism, paralleled by a slow-to-fast fiber type transition[87]. Evidence has been provided of a link between PGC-1α and the TNF-like weak inducer of apoptosis (TWEAK)-Fibroblast growth factor inducible (Fn)14 signaling pathway[88], known to be involved in muscle atrophy[89]. Indeed, muscle-specific PGC-1α overexpression has been shown to inhibit TWEAK-induced expression of atrogenes and to prevent the inducible expression of TWEAK receptor Fn14 in denervated muscle[88]. Finally, recent results obtained in our laboratory show that PGC-1α overexpression improves muscle wasting in tumor-bearing mice (Pin, Busquets et al., under revision). These results are consistent with those reported by Ruas and coworkers[90], showing that transgenic mice overexpressing the PGC-1α4 isoform, characterized by muscle hypertrophy, are more resistant than wild-type animals to hindlimb-associated muscle atrophy as well as to cancer-induced cachexia.

6. Conclusion

In the last decades several mechanisms underlying muscle wasting in cachexia have been unraveled, however further research is required, with particular reference to the need to confirm in humans data arising from animal models. The reduction of physical activity that generally occurs in cancer patients likely significantly contributes to the pathogenesis of muscle wasting in cachexia. For this reason, exercise training is regarded as a promising intervention that can attenuate cancer-induced muscle wasting. However, since exercise capability can be impaired in cancer patients, drugs able to mimick exercise effects would be welcome. In this regard, PPARδ or AMPK agonists as well as SIRT1 activators have been proposed as potentially useful drugs (Table 1). The results actually available are encouraging but still at the pre-clinical level. Further studies are thus warranted to extend the experimental observations and to translate them to the clinical practice.

7. Expert opinion

Cancer cachexia originates from a complex network of profound metabolic alterations that reflects the action of inflammatory mediators, of energy deficit and of hypercatabolic stimuli. At the molecular level at least, such perturbations may occur at very early disease stages, well before any evidence of general wasting.

Tumor eradication obviously is the only radical approach to deal with cachexia. However, not only this goal is totally beyond reach, but cachexia compromises its feasibility. Therefore, reliable therapeutic strategies aimed at interfering with the onset and/or progression of cachexia need to be pursued and timely implemented in patients. On the basis of results obtained in experimental studies, several drugs are now under evaluation in clinical trials to test their effectiveness in preventing/delaying cachexia in cancer patients. The rationale underlying such trials is quite variable. In general, however, it is important to realize that monotherapies are hardly adequate to deal with cachexia and this view seems to gain a growing consensus. Rather, the most suitable approach should likely be a multidirectional one, selectively tailored, whenever possible, on a panel of the manifold and variable (e.g., inflammatory cytokines) pathogenic factors involved.

In the last years the knowledge about the mechanisms underlying cachexia has expanded considerably, also taking into consideration impaired myogenesis and the occurrence of energy deficit, paving the way to new potential therapeutic targets. Few attempts to improve cachexia by impinging, directly or indirectly, on myogenesis have been reported so far (see above). While the results appear encouraging, further studies are needed to clarify this issue.

The occurrence of energy dysmetabolism in cachexia is long recognized, however the research on the underlying mechanisms and the definition of useful therapeutic strategies is at the very beginning. In the last years, alterations in mitochondrial structure and function have been reported in the skeletal muscle of cancer hosts. Being mitochondria the main source of energy used by the muscle for contractile activity, their dysfunction would result in oxidative and energy stress, ultimately contributing to muscle wasting. Exercise has proven to be very effective to improve muscle phenotype by impinging on both anabolic and catabolic (pro-inflammatory) pathways.

Despite the potentially favorable perspectives reported above, to disrupt the sedentary habits shared by most patients is not an easy task. In addition, chronic fatigue and co-morbidities frequently occurring in cancer patients, such as anemia and cardiac dysfunctions, are limiting factors for practicing physical activity, and can eventually lead to exercise intolerance. Consistently, muscle wasting is not prevented by exercise in mice bearing the C26 tumor (Pin, Busquets et al., under revision), in which cachexia is associated with anemia and cardiac alterations[91], while excessive endurance exercise is associated with increased mitochondrial fission in the absence of mitophagy induction[92]. For these reasons, the availability of drugs that mimic the effects of exercise action would be desirable in order to maintain the beneficial effects of exercise avoiding the negative aspects represented by potential muscle injury and by the need of a 'permissive' energy metabolism, frequently lacking in cancer patients, especially in advanced disease stages (Figure 1).

The strength of the research on drugs mimicking exercise is to clarify if they can effectively become part of a tailored therapeutic protocol aimed at preventing/delaying cachexia. This would allow the maintenance of cancer patients in the pre-cachexia stage as long as possible, markedly improving the possibility to complete the scheduled antineoplastic treatment, achieving the most possible effectiveness. However, since in most cases the data available on drugs mimicking exercise are at the pre-clinical level,

studies aimed at validating the results also at the clinical level are warranted. This is not an easy task, however, since such studies would be more complicated, in terms of both invasiveness of the approach and ethical issues, than prospective clinical investigations based on anthropometric and/or biological fluid-derived data.

Finally, a note of care must be posed, since many exercise mimetics, in particular those able to induce PPARδ (glitazones, metformin), may produce dangerous side-effects such as worsening the cardiovascular function or increasing the oncogenic risk. In this regard, studies should concentrate on those drugs able to improve energy metabolism, acting on mitochondrial turnover and oxidative metabolism, being well tolerated at the same time.

Table 1. Drug mimicking exercise: the state of the art

	CURRENT STATUS	REFERENCES
PPARδ AGONISTS		
GW501516	pre-clinical	44, 46-48, 53
GW0742	pre-clinical	48
glitazones	pre-clinical	54-56
AMPK ACTIVATORS		
AICAR	pre-clinical	46, 53, 65
β-GPA	pre-clinical	48
metformin	clinical	66-68
resveratrol	pre-clinical	69, 70
SIRT1 ACTIVATORS		
resveratrol	pre-clinical	79, 81
SRT2104	pre-clinical	85, 86

Drugs currently under investigation for their use as exercise mimetics are listed. References reporting effects on target tissues different from the skeletal muscle are not included.

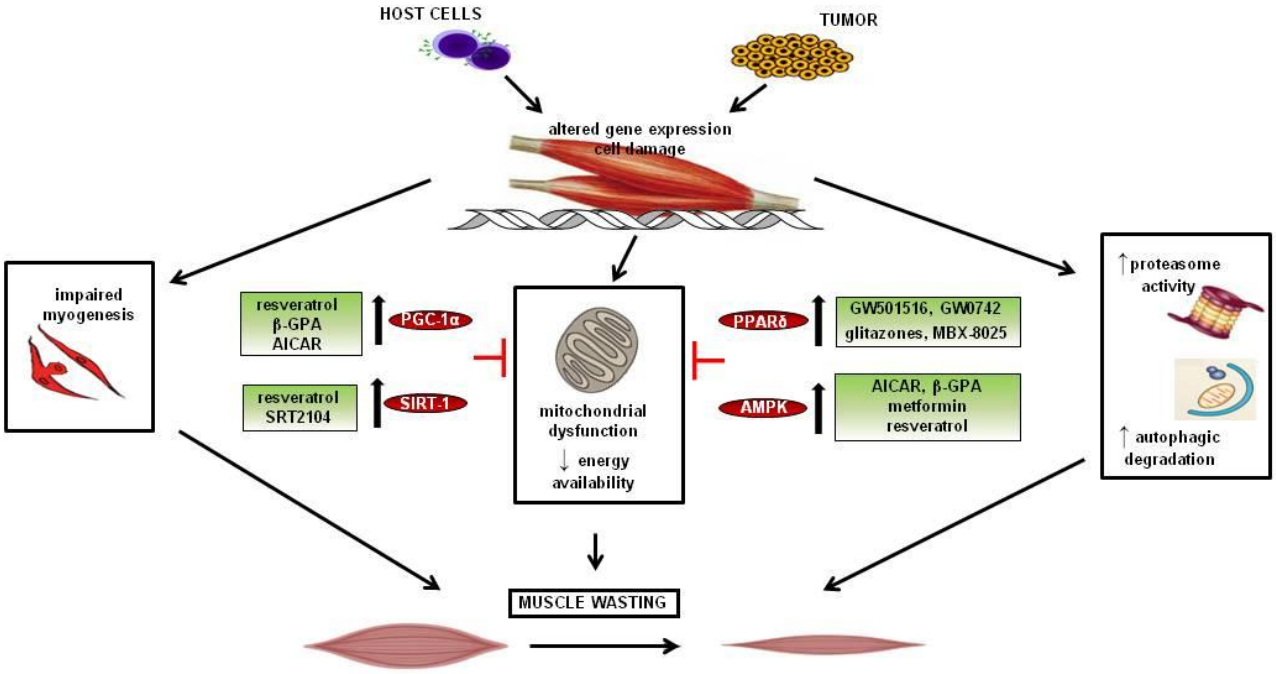

Figure 1. Different classes of molecules are able to mimic the effects of aerobic/endurance exercise. Both host cells and the tumor secrete humoral factors and produce metabolic changes which modify skeletal muscle gene expression and cause cell damage. These alterations lead to the hyperactivation of protein degradation, affect myogenesis and impair mitochondrial function, ultimately leading to muscle wasting. Exercise mimetics (green squares) act on exercise-responsive factors (AMPK, PPARδ, PGC-1α, SIRT1), improving the energy deficit by restoring mitochondrial biogenesis, oxidative capacity and ATP synthesis rates.

Disclosure

The authors declare that there is no conflict of interest.

6. References

Papers of special note have been highlighted as either of interest (●) or of considerable interest (●●) to readers.

1. Argilés JM, Busquets S, Stemmler B et al. Cancer cachexia: understanding the molecular basis. Nat Rev Cancer 2014;14:754–62

● One of the most complete reviews on cancer cachexia and energy metabolism alterations

2. Penna F, Baccino FM, Costelli P. Coming back: autophagy in cachexia. Curr Opin Clin Nutr Metab Care 2014;17:241–6
3. Tessitore L, Costelli P, Baccino FM. Pharmacological interference with tissue hypercatabolism in tumour-bearing rats. Biochem J 1994;299:71–8
4. Chacon-Cabrera A, Fermoselle C, Urtreger AJ et al. Pharmacological strategies in lung cancer-induced cachexia: effects on muscle proteolysis, autophagy, structure, and weakness. J Cell Physiol 2014;229:1660–72
5. Penna F, Bonetto A, Aversa Z et al. Effect of the specific proteasome inhibitor bortezomib on cancer-related muscle wasting. J Cachexia Sarcopenia Muscle 2015. Available from: http://doi.wiley.com/10.1002/jcsm.12050
6. Masiero E, Agatea L, Mammucari C et al. Autophagy Is Required to Maintain Muscle Mass. Cell Metab 2009;10:507–15
7. Penna F, Costamagna D, Pin F et al. Autophagic degradation contributes to muscle wasting in cancer cachexia. Am J Pathol 2013;182:1367–78
8. Carnio S, LoVerso F, Baraibar MA et al. Autophagy impairment in muscle induces neuromuscular junction degeneration and precocious aging. Cell Rep 2014;8:1509–21.
9. Li JP, Lu L, Wang LJ et al. Increased serum levels of S100B are related to the severity of cardiac dysfunction, renal insufficiency and major cardiac events in patients with chronic heart failure. Clin Biochem 2011;44:984–8
10. Niewczas MA, Gohda T, Skupien J et al. Circulating TNF receptors 1 and 2 predict ESRD in type 2 diabetes. J Am Soc Nephrol 2012;23:507–15
11. Seruga B, Zhang H, Bernstein LJ et al. Cytokines and their relationship to the symptoms and outcome of cancer. Nat Rev Cancer 2008;8:887–99
12. Walston J, Fedarko N, Yang H et al. The physical and biological characterization of a frail mouse model. J Gerontol A Biol Sci Med Sci 2008;63:391–8
13. Cai D, Frantz JD, Tawa NE et al. IKKbeta/NF-kappaB activation causes severe muscle wasting in mice. Cell 2004;119:285–98
14. Bonetto A, Aydogdu T, Kunzevitzky N et al. STAT3 activation in skeletal muscle links muscle wasting and the acute phase response in cancer cachexia. PLoS One 2011;6:e22538
15. Fearon K, Strasser F, Anker SD et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011;12:489–95
16. Biolo G, Cederholm T, Muscaritoli M. Muscle contractile and metabolic dysfunction is a common feature of sarcopenia of aging and chronic diseases: from sarcopenic obesity to cachexia. Clin Nutr 2014;33:737–48
17. Harrington D, Anker SD, Chua TP et al. Skeletal muscle function and its relation to exercise tolerance in chronic heart failure. J Am Coll Cardiol 1997;30:1758–64

18. Sartori R, Schirwis E, Blaauw B et al. BMP signaling controls muscle mass. Nat Genet 2013;45:1309–18
19. Muscaritoli M, Molfino A, Lucia S et al. Cachexia: a preventable comorbidity of cancer. A T.A.R.G.E.T. approach. Crit Rev Oncol Hematol 2015;94:251–9
20. Aoyagi T, Terracina KP, Raza A et al. Cancer cachexia, mechanism and treatment. World J Gastrointest Oncol 2015;7:17–29
21. Mauro A. Satellite cell of skeletal muscle fibers. J Biophys Biochem Cytol 1961;9:493–5
22. Penna F, Costamagna D, Fanzani A et al. Muscle wasting and impaired Myogenesis in tumor bearing mice are prevented by ERK inhibition. PLoS One 2010;5.
23. He WA, Berardi E, Cardillo VM et al. NF-κB-mediated Pax7 dysregulation in the muscle microenvironment promotes cancer cachexia. J Clin Invest 2013;123:4821–35
24. Ramamoorthy S, Donohue M, Buck M. Decreased Jun-D and myogenin expression in muscle wasting of human cachexia. Am J Physiol Endocrinol Metab 2009;297:E392–401
25. Prado CMM, Bekaii-Saab T, Doyle LA et al. Skeletal muscle anabolism is a side effect of therapy with the MEK inhibitor: selumetinib in patients with cholangiocarcinoma. Br J Cancer 2012;106:1583–6
26. Toledo M, Penna F, Busquets S et al. Distinct behaviour of sorafenib in experimental cachexia-inducing tumours: the role of STAT3. PLoS One 2014;9:e113931
27. Shum AMY, Mahendradatta T, Taylor RJ et al. Disruption of MEF2C signaling and loss of sarcomeric and mitochondrial integrity in cancer-induced skeletal muscle wasting. Aging 2012;4:133–43
28. Fontes-Oliveira CC, Busquets S, Toledo M et al. Mitochondrial and sarcoplasmic reticulum abnormalities in cancer cachexia: altered energetic efficiency? Biochim Biophys Acta 2013;1830:2770–8
29. Julienne CM, Dumas J-F, Goupille C et al. Cancer cachexia is associated with a decrease in skeletal muscle mitochondrial oxidative capacities without alteration of ATP production efficiency. J Cachexia Sarcopenia Muscle 2012;3:265–75
30. Tzika AA, Fontes-Oliveira CC, Shestov AA et al. Skeletal muscle mitochondrial uncoupling in a murine cancer cachexia model. Int J Oncol 2013;43:886–94
31. McLean JB, Moylan JS, Andrade FH. Mitochondria dysfunction in lung cancer-induced muscle wasting in C2C12 myotubes. Front Physiol 2014;5:503
32. White JP, Puppa MJ, Sato S et al. IL-6 regulation on skeletal muscle mitochondrial remodeling during cancer cachexia in the ApcMin/+ mouse. Skelet Muscle 2012;2:14
33. Romanello V, Sandri M. Mitochondrial biogenesis and fragmentation as regulators of muscle protein degradation. Curr Hypertens Rep 2010;12:433–9
● A very good survey of the relation between altered mitochondria and muscle protein breakdown
34. Varanita T, Soriano ME, Romanello V et al. The OPA1-dependent mitochondrial cristae remodeling pathway controls atrophic, apoptotic, and ischemic tissue damage. Cell Metab 2015;21:834–44
35. Zinna EM, Yarasheski KE. Exercise treatment to counteract protein wasting of chronic diseases. Curr Opin Clin Nutr Metab Care 2003;6:87–93
●This review clearly shows the effectiveness of exercise in correcting muscle wasting
36. Kirby TJ, McCarthy JJ. MicroRNAs in skeletal muscle biology and exercise adaptation. Free Radic Biol Med 2013;64:95–105

37. Puigserver P, Spiegelman BM. Peroxisome proliferator-activated receptor-gamma coactivator 1 alpha (PGC-1 alpha): transcriptional coactivator and metabolic regulator. Endocr Rev 2003;24:78–90
38. Drake JC, Wilson RJ, Yan Z. Molecular mechanisms for mitochondrial adaptation to exercise training in skeletal muscle. FASEB J 2015; Available from: http://www.ncbi.nlm.nih.gov/pubmed/26370848
39. Vainshtein A, Desjardins EM, Armani A et al. PGC-1α modulates denervation-induced mitophagy in skeletal muscle. Skelet Muscle 2015;5:9
● This paper demonstrates the effect of PGC-1α modulations on mitophagy
40. Itoh Y, Hayakawa K, Mori T et al. Stand-up exercise training facilitates muscle recovery from disuse atrophy by stimulating myogenic satellite cell proliferation in mice. Physiol Rep 2014;2:e12185–e12185
41. Mangan G, Bombardier E, Mitchell AS et al. Oestrogen-dependent satellite cell activation and proliferation following a running exercise occurs via the PI3K signalling pathway and not IGF-1. Acta Physiol 2014;212:75–85
42. Constantinou C, Fontes de Oliveira CC, Mintzopoulos D et al. Nuclear magnetic resonance in conjunction with functional genomics suggests mitochondrial dysfunction in a murine model of cancer cachexia. Int J Mol Med 2011;27:15–24
43. Luquet S, Lopez-Soriano J, Holst D et al. Peroxisome proliferator-activated receptor delta controls muscle development and oxidative capability. FASEB J 2003;17:2299–301
44. Wang Y-X, Zhang C-L, Yu RT et al. Regulation of muscle fiber type and running endurance by PPARdelta. PLoS Biol 2004;2:e294
45. Schuler M, Ali F, Chambon C et al. PGC1alpha expression is controlled in skeletal muscles by PPARbeta, whose ablation results in fiber-type switching, obesity, and type 2 diabetes. Cell Metab 2006;4:407–14
46. Narkar VA, Downes M, Yu RT et al. AMPK and PPARdelta agonists are exercise mimetics. Cell 2008;134:405–15
●● The first demonstration that exercise can be mimicked by drugs
47. Kleiner S, Nguyen-Tran V, Baré O et al. PPAR{delta} agonism activates fatty acid oxidation via PGC-1{alpha} but does not increase mitochondrial gene expression and function. J Biol Chem 2009;284:18624–33
48. Ljubicic V, Burt M, Jasmin BJ. The therapeutic potential of skeletal muscle plasticity in Duchenne muscular dystrophy: phenotypic modifiers as pharmacologic targets. FASEB J 2014;28:548–68
●● An elegant demonstration that PPARδ agonists improve muscle distrophy
49. Barish GD, Atkins AR, Downes M et al. PPARdelta regulates multiple proinflammatory pathways to suppress atherosclerosis. Proc Natl Acad Sci U S A 2008;105:4271–6
50. Tanaka T, Yamamoto J, Iwasaki S et al. Activation of peroxisome proliferator-activated receptor delta induces fatty acid beta-oxidation in skeletal muscle and attenuates metabolic syndrome. Proc Natl Acad Sci U S A 2003;100:15924–9
51. Olson EJ, Pearce GL, Jones NP et al. Lipid effects of peroxisome proliferator-activated receptor-δ agonist GW501516 in subjects with low high-density lipoprotein cholesterol: characteristics of metabolic syndrome. Arterioscler Thromb Vasc Biol 2012;32:2289–94
52. Bays HE, Schwartz S, Littlejohn T et al. MBX-8025, a novel peroxisome proliferator receptor-delta agonist: lipid and other metabolic effects in dyslipidemic overweight patients treated with and without atorvastatin. J Clin Endocrinol Metab 2011;96:2889–97

53. Jahnke VE, Van Der Meulen JH, Johnston HK et al. Metabolic remodeling agents show beneficial effects in the dystrophin-deficient mdx mouse model. Skelet Muscle 2012;2:16
54. Wang X, Hu Z, Hu J, Du J et al. Insulin resistance accelerates muscle protein degradation: Activation of the ubiquitin-proteasome pathway by defects in muscle cell signaling. Endocrinology 2006;147:4160–8
55. Asp ML, Tian M, Wendel AA et al. Evidence for the contribution of insulin resistance to the development of cachexia in tumor-bearing mice. Int J Cancer 2010;126:756–63
56. Asp ML, Tian M, Kliewer KL et al. Rosiglitazone delayed weight loss and anorexia while attenuating adipose depletion in mice with cancer cachexia. Cancer Biol Ther 2011;12:957–65
57. Shea MK, Nicklas BJ, Marsh AP et al. The effect of pioglitazone and resistance training on body composition in older men and women undergoing hypocaloric weight loss. Obesity 2011;19:1636–46
58. Castillero E, Alamdari N, Aversa Z et al. PPARβ/δ Regulates Glucocorticoid- and Sepsis-Induced FOXO1 Activation and Muscle Wasting. PLoS One 2013;8:e59726
59. Fuster G, Busquets S, Ametller E et al. Are peroxisome proliferator-activated receptors involved in skeletal muscle wasting during experimental cancer cachexia? Role of beta2-adrenergic agonists. Cancer Res 2007;67:6512–9
60. Starner CI, Schafer JA, Heaton AH et al. Rosiglitazone and pioglitazone utilization from January 2007 through May 2008 associated with five risk-warning events. J Manag Care Pharm 2015;14:523–31
61. Govindan J, Evans M. Pioglitazone in Clinical Practice: Where Are We Now? Diabetes Ther 2012;3:1
62. Krawiec BJ, Nystrom GJ, Frost RA et al. AMP-activated protein kinase agonists increase mRNA content of the muscle-specific ubiquitin ligases MAFbx and MuRF1 in C2C12 cells. Am J Physiol Endocrinol Metab 2007;292:E1555–67
63. Sanchez AMJ, Candau RB, Csibi A et al. The role of AMP-activated protein kinase in the coordination of skeletal muscle turnover and energy homeostasis. Am J Physiol Cell Physiol 2012;303:C475–85
64. Mounier R, Théret M, Lantier L et al. Expanding roles for AMPK in skeletal muscle plasticity. Trends Endocrinol Metab 2015;26:275–86
65. Tabony AM, Yoshida T, Galvez S et al. Angiotensin II upregulates protein phosphatase 2Cα and inhibits AMP-activated protein kinase signaling and energy balance leading to skeletal muscle wasting. Hypertension 2011;58:643–9
66. Diaz EC, Herndon DN, Porter C et al. Effects of pharmacological interventions on muscle protein synthesis and breakdown in recovery from burns. Burns 2015;41:649–57
67. Cetrone M, Mele A, Tricarico D. Effects of the antidiabetic drugs on the age-related atrophy and sarcopenia associated with diabetes type II. Curr Diabetes Rev 2014;10:231–7
68. Chevalier S, Farsijani S. Cancer cachexia and diabetes: similarities in metabolic alterations and possible treatment. Appl Physiol Nutr Metab 2014;39:643–53
69. Um J-H, Park S-J, Kang H et al. AMP-activated protein kinase-deficient mice are resistant to the metabolic effects of resveratrol. Diabetes 2010;59:554–63
70. Wang B, Yang G, Liang X et al. Grape seed extract prevents skeletal muscle wasting in interleukin 10 knockout mice. BMC Complement Altern Med 2014;14:162
71. Timmers S, Konings E, Bilet L et al. Calorie restriction-like effects of 30 days of resveratrol supplementation on energy metabolism and metabolic profile in obese humans. Cell Metab 2011;14:612–22

72. Naia L, Rego AC. Sirtuins: double players in Huntington's disease. Biochim Biophys Acta 2015;1852:2183–94
73. Haigis MC, Sinclair DA. Mammalian sirtuins: biological insights and disease relevance. Annu Rev Pathol 2010;5:253–95
74. Cantó C, Auwerx J. PGC-1alpha, SIRT1 and AMPK, an energy sensing network that controls energy expenditure. Curr Opin Lipidol 2009;20:98–105
75. Price NL, Gomes AP, Ling AJ et al. SIRT1 is required for AMPK activation and the beneficial effects of resveratrol on mitochondrial function. Cell Metab 2012;15:675–90
●● An elegant demonstration that SIRT1 is crucial for resveratrol-induced AMPK stimulation and mitochondiral function improvement
76. Bordone L, Cohen D, Robinson A et al. SIRT1 transgenic mice show phenotypes resembling calorie restriction. Aging Cell 2007;6:759–67
77. Lee D, Goldberg AL. SIRT1 Protein, by Blocking the Activities of Transcription Factors FoxO1 and FoxO3, Inhibits Muscle Atrophy and Promotes Muscle Growth. J Biol Chem 2013;288:30515–26
78. Chalkiadaki A, Guarente L. The multifaceted functions of sirtuins in cancer. Nat Rev Cancer 2015;15:608–24
79. Lagouge M, Argmann C, Gerhart-Hines Z et al. Resveratrol improves mitochondrial function and protects against metabolic disease by activating SIRT1 and PGC-1alpha. Cell 2006;127:1109–22
80. Baur JA, Pearson KJ, Price NL et al. Resveratrol improves health and survival of mice on a high-calorie diet. Nature 2006;444:337–42
81. Gerhart-Hines Z, Rodgers JT, Bare O et al. Metabolic control of muscle mitochondrial function and fatty acid oxidation through SIRT1/PGC-1alpha. EMBO J 2007;26:1913–23
82. Libri V, Brown AP, Gambarota G et al. A pilot randomized, placebo controlled, double blind phase I trial of the novel SIRT1 activator SRT2104 in elderly volunteers. PLoS One 2012;7:e51395
83. Hoffmann G, Breitenbücher F, Schuler M et al. A novel sirtuin 2 (SIRT2) inhibitor with p53-dependent pro-apoptotic activity in non-small cell lung cancer. J Biol Chem 2014;289:5208–16
84. Venkatasubramanian S, Noh RM, Daga S et al. Cardiovascular effects of a novel SIRT1 activator, SRT2104, in otherwise healthy cigarette smokers. J Am Heart Assoc 2013;2:e000042
85. Mercken EM, Mitchell SJ, Martin-Montalvo A et al. SRT2104 extends survival of male mice on a standard diet and preserves bone and muscle mass. Aging Cell 2014;13:787–96
●● This study shows the protective effects of synthetic SIRT1 activators on aging and immobilization-induced muscle atrophy
86. Sandri M, Lin J, Handschin C et al. PGC-1alpha protects skeletal muscle from atrophy by suppressing FoxO3 action and atrophy-specific gene transcription. Proc Natl Acad Sci U S A 2006;103:16260–5t
87. Chan MC, Arany Z. The many roles of PGC-1α in muscle — recent developments. Metabolism 2014;63:441–51
88. Hindi SM, Mishra V, Bhatnagar S et al. Regulatory circuitry of TWEAK-Fn14 system and PGC-1α in skeletal muscle atrophy program. FASEB J 2014;28:1398–411
89. Kumar A, Bhatnagar S, Paul PK. TWEAK and TRAF6 regulate skeletal muscle atrophy. Curr Opin Clin Nutr Metab Care 2012;15:233–9
90. Ruas JL, White JP, Rao RR et al. A PGC-1α isoform induced by resistance training regulates skeletal muscle hypertrophy. Cell 2012;151:1319–31

91. Tian M, Asp ML, Nishijima Y et al. Evidence for cardiac atrophic remodeling in cancer-induced cachexia in mice. Int J Oncol 2011;39:1321–6
92. Jamart C, Francaux M, Millet GY et al. Modulation of autophagy and ubiquitin-proteasome pathways during ultra-endurance running. J Appl Physiol 2012;112:1529–37