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Pharmacological effects of denervated muscle atrophy due to metabolic imbalance in different periods

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Abstract: Denervation-induced skeletal muscle atrophy can potentially cause the decline in the quality of life of patients and an increased risk of mortality. Complex pathophysiological mechanisms with dynamic alterations have been documented in skeletal muscle atrophy resulting from innervation loss. Hence, an in-depth comprehension of the key mechanisms and molecules governing skeletal muscle atrophy at varying stages, along with targeted treatment and protection, becomes essential for effective atrophy management. Our preliminary research categorizes the skeletal muscle atrophy process into four stages using microarray analysis. This review extensively discusses the pathways and molecules potentially implicated in regulating the four stages of denervation and muscle atrophy. Notably, drugs targeting the reactivare oxygen species stage and the inflammation stage assume critical roles. Timely intervention during the initial atrophy stages can expedite protection against skeletal muscle atrophy. Additionally, pharmaceutical intervention in the ubiquitin-proteasome pathway associated with atrophy and autophagy lysosomes can effectively slow down skeletal muscle atrophy. Key molecules within this stage encompass MuRF1, MAFbx, LC3II, p62/SQSTM1, etc. This review also compiles a profile of drugs with protective effects against skeletal muscle atrophy at distinct post-denervation stages, thereby augmenting the evidence base for denervation-induced skeletal muscle atrophy treatment.

Introduction

As a pivotal effector organ within the peripheral nervous system, the structural integrity and functional preservation of skeletal muscle are subject to control and regulation by the nervous system. Peripheral nerve injury arising from acute trauma results in the loss of innervation from skeletal muscle. This initiates a sequence of pathological changes, including reduced muscle fiber cross-sectional area, myofibril degradation with destruction of sarcomeres, diminished contraction speed, and fibrosis, ultimately culminating atrophy (Dumitru *et al.*, 2018). The

regeneration rate of peripheral nerves is slow after injury. As a result, the target muscles of severe patients become irreversibly atrophied before being re-innervated by the nerves. This can even lead to disabling in the patients, bringing a heavy burden to the family and society (Gu *et al.*, 2011). Therefore, a series of problems in target muscle repair and functional reconstruction after peripheral nerve injury could be resolved by exploring novel and effective methods for the treatment of skeletal muscle atrophy.

Skeletal muscle atrophies upon innervation loss due to perturbations in the balance between protein synthesis and degradation. Notably, protein degradation pathways become activated, while protein synthesis pathways are repressed, constituting the underlying cause of skeletal muscle atrophy. Effective alleviation of skeletal muscle atrophy is promoted by inhibition of protein degradation and stimulation of protein synthesis (Lang *et al.*, 2017; Li *et al.*, 2017; Cui *et al.*, 2019). Several molecules are involved in the process of denervated muscular atrophy, and the mechanism of

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regulating denervated muscular atrophy is also extremely complicated. Recent years have witnessed many studies on denervated muscular atrophy at home and abroad, but most of these studies have focused on a single event, a single gene or protein (Castets *et al.*, 2019; Janice Sanchez *et al.*, 2019). A more comprehensive understanding of the denervated skeletal muscle atrophy process is attainable by elucidating pivotal regulators and therapeutic targets. Shen *et al.* (2019) employed microarray analysis to scrutinize target muscles at varying post-denervation time points, revealing that the 28-day period following a neural injury can be segmented into four distinct transcriptional phases, demarcated by three nodal transitions (Shen *et al.*, 2019). These phases encompass the oxidative stress stage, inflammation stage, atrophy stage, and atrophic fibrosis stage. Consistent with previous research, the phenotype of skeletal muscle atrophy manifests between 36 h and 3 days, aligning with the aforementioned atrophy period (Shen *et al.*, 2019; Qiu *et al.*, 2021). Consequently, these research findings facilitate more optimal utilization of diverse methods for denervated skeletal muscle atrophy treatment. Several treatment modalities are currently proposed, encompassing electrical stimulation (Dow *et al.*, 2005; Tamaki *et al.*, 2017) and noncoding RNA (He *et al.*, 2016; Li *et al.*, 2017; Hitachi *et al.*, 2019). Furthermore, extensive utilization of various food and drug extracts is evident in the treatment of denervated skeletal muscle atrophy.

The enhanced medical application of traditional Chinese medicine (TCM) is attributed to its multifaceted functionalities and its facile interaction with substances within the human body. Substantial strides have been made in studying the prevention and treatment of skeletal muscle atrophy using various drugs. Noteworthy achievements include effective myotube atrophy management through triptolide, an ingredient isolated from ancient Chinese herbal medicine. This agent triggers IRS-1 degradation and activates the FOXO3 pathway activation (Wang *et al.*, 2020). Furthermore, withaferin A (WFA), a natural protein-specific binding waveform, mitigates nuclear factor kappa B (NF-κB)-mediated pro-inflammatory signaling, thereby alleviating cancer cachexia-induced skeletal muscle atrophy (Straughn and Kakar, 2019). This article highlights distinct stages of skeletal muscle atrophy, protein synthesis pathways, and other pathways. It systematically summarizes the role of food and drug extracts and other effects on the treatment of skeletal muscle atrophy, which can provide a reference for the clinical management of denervated skeletal muscle atrophy.

Drugs that Play a Protective Role by Inhibiting Protein Degradation

Inhibition of reactive oxygen species

An earlier study involving rats subjected to neural exchange suggested that heightened mitochondrial permeability transitions and increased apoptosis result in elevated mitochondrial ROS production, signifying mitochondrial dysfunction following denervation (Adhihetty *et al.*, 2007). Reactive oxygen species (ROS) are accountable for maintaining the homeostasis of various physiological processes. A mounting body of evidence posits oxidative stress as a crucial regulator of muscle wasting (Jackman and Kandarian, 2004; Powers *et al.*, 2005). Meanwhile, an initial stage in denervated skeletal muscle exposes aberrant ROS production. Thus, curbing excessive ROS production can ameliorate skeletal muscle atrophy. Compared to a control group, a delay in muscle atrophy mediated by ROS inhibition was reported by Kim *et al.* (2018). On injecting *Oenothera odorata* root extract (EVP) in both H_2O_2-treated C_2C_{12} myoblasts and sciatic-denervated mice (Kim *et al.*, 2018). They probed deeper into the mechanistic underpinnings of how reactive oxygen species oversee skeletal muscle atrophy. EVP suppressed superoxide dismutase 1 (SOD1) expression and augmented HSP70 expression in H_2O_2-treated C_2C_{12} myoblasts and sciatic-neutralized mice. In addition, EVP regulates apoptotic signals, including caspase-3, B-cell lymphoma protein 2 (Bcl-2), Bcl-2-associated X (Bax), and ceramides. Administration of recombinant heat shock protein 70 delays peripheral muscle denervation in the SOD1 (G93A) mouse model of amyotrophic lateral sclerosis (Gifondorwa *et al.*, 2012). O'Leary and Hood have previously shown that seven days of muscle disuse increases the expression of Beclin-1, as well as LC3-II, a known component of autophagy (O'Leary and Hood, 2009). These studies provide evidence for EVP's protective effect and regulatory mechanism on denervation muscle atrophy. The glutathione content and malondialdehyde (MDA) levels are considered indicators of oxidative stress and were assayed in the denervated gastrocnemius muscle. As a free radical scavenger, adding vitamin E can restore glutathione levels and reduce MDA levels, further confirming the conclusion that oxidative stress accelerates muscle atrophy (Demiryurek and Babul, 2004). Indeed, increased oxidative damage after denervation resulted in increased ROS production, decreased surface hydrophobicity, and decreased enzymatic activities of glyceraldehyde-3-phosphate dehydrogenase and creatine kinase, while increased mitochondrial ROS may also play a signaling role (Pierce *et al.*, 2006). Another compelling candidate for ROS-mediated signaling is NF-κB. The involvement of NF-κB in muscle atrophy is recognized. The inhibitor of apoptosis 1 (cIAP1) protein, a positive regulator of NF-κB signaling, exhibited upregulation in denervated muscles compared to non-denervated controls 14 days post-denervation. Genetic or pharmacological inhibition of cIAP1 curtailed canonical NF-κB signaling, thus attenuating denervation-induced muscle atrophy (Lala-Tabbert *et al.*, 2019). Furthermore, mitochondrial ROS has been demonstrated to upregulate the expression of ubiquitin ligase, atrogin-1/MAFbx, potentially contributing to atrophy through enhanced protein degradation by the 26S proteasome system (Li *et al.*, 2005). We found that pyrroloquinoline quinone (PQQ)-mediated reduction in the expression of reactive oxygen species and inflammatory factors results in a decrease in the expression of MuRF1 and MAFbx, ultimately improving denervated skeletal muscle atrophy (Qiu *et al.*, 2018; Ma *et al.*, 2019). Kuo from Taiwan revealed PQQ impairs denervation-induced skeletal muscle atrophy by activating PGC-1α and integrating mitochondrial electron transport chain complexes (Kuo *et al.*, 2015). Previous research by our team reported a reduction of

denervation-induced oxidative stress and inflammation mediated by salidroside, inhibiting muscle proteolysis and ultimately alleviating muscle atrophy caused by denervation (Huang *et al.*, 2019).

Inhibition of pro-inflammatory factors
Apigenin is a natural plant flavonoid acclaimed for its anti-obesity, anti-inflammatory, antioxidant, and anti-cancer attributes. Apigenin was attributed to dose-dependent inhibition of collagenase activity implicated in rheumatoid arthritis (RA) and the suppression of lipopolysaccharide (LPS)-induced nitric oxide (NO) production in RAW 264.7 macrophage cells (Lee *et al.*, 2007). In denervated gastrocnemius and soleus muscles, apigenin treatment impeded the upregulation of tumor necrosis factor alpha (TNF-α) and interleukin (IL)-6 expression (Choi *et al.*, 2018). Furthermore, Shiota *et al.* (2015) reported that apigenin exerted inhibitory effects on atrogin-1/MAFbx expression and curbed the reduction in myotube diameter induced by LPS stimulation in the C_2C_{12} murine cell line (Shiota *et al.*, 2015).

Ficus carica L. (FCL.), a flowering plant, contains flavonoids, psoralen, and bergapten, renowned for their antioxidant, anti-inflammatory, and anti-apoptotic characteristics. They are also implicated in quelling IL-1β and IL-6 production in atrophic muscles, attenuating muscle inflammation by inhibiting nuclear factor NF-κB activation (Dai *et al.*, 2020). A study by Aarti Yadav *et al.* identified quercetin as a dietary antioxidant flavonoid that curbed inflammation in myotubes, fully reinstating TNF and an a-induced reduction of myotube diameter (Kim *et al.*, 2018). This reflects muscle morphology at the cellular level. Quercetin improves motor function and muscle mass in aged people (Yadav *et al.*, 2022). These reported effects of flavonoids are in other skeletal muscle atrophy models rather than in denervation-induced skeletal muscle atrophy. However, apigenin, functioning as a bioactive flavone, augmented the fiber-cross sectional area by 0.1% in the gastrocnemius muscle of sciatic denervated mice over 2 weeks. The enhanced cross-sectional area was substantiated by diminished levels of TNF-α in the gastrocnemius and IL-6 in the soleus muscle (Choi *et al.*, 2018). Buyang Huangwu Tang (BYHWT), a classic Chinese medicine formula, ameliorated the inflammatory response in denervation-dependent skeletal muscle atrophy rat models (Zhou *et al.*, 2020). Wu *et al.* (2019) found that negative regulation of pro-inflammatory cytokine mediated by salidroside attenuates denervation-induced skeletal muscle atrophy. The discernible pathway involves the inflammation caused by denervation in skeletal muscle, leading to the generation of inflammatory cytokines, particularly IL-6. This, in turn, augments the phosphorylation of STAT3 and the expression of SOCS3, which consequently triggers the activation of the proteolytic pathway in the muscle. Salidroside curbed muscle proteolysis and muscle atrophy by tempering the inflammatory response triggered by innervation. This likely transpired through the inactivation of the STAT3/SOCS3 pathway (Wu *et al.*, 2019).

Regulation of ubiquitin-proteasome and autophagy-lysosome pathway
Activation of proteolytic metabolism and inhibition of protein synthesis pathway are the primary causes of skeletal muscle atrophy following denervation. The proteolytic pathway chiefly encompasses the ubiquitin-proteasome system and the autophagy-lysosome system. Clenbuterol (CLE) is a beta-adrenergic receptor stimulant employed in treating muscle spasms and asthma (Bohorov *et al.*, 1987; Pairet *et al.*, 1997). *In vivo* and *in vitro* treatment with CLE was found to prohibit the transcriptional upregulation of atrophy-related Ub ligases (i.e., MAFbx and MuRF1) in denervated soleus muscles (Goncalves *et al.*, 2012). MAFbx overexpression led to a reduction in cultured muscle cell line size, indicating accelerated protein catabolism. Conversely, *MAFbx* gene depletion impeded nerve transection-induced muscle loss (Bodine *et al.*, 2001). Nandrolone decelerates denervation atrophy by repressing MAFbx and MuRF1 (Zhao *et al.*, 2008). The results showed that Nandrolone inhibited the expression of MAFbx and MuRF1 in subacutely denervated muscles. Reduced MAFbx and MuRF1 expression 35 and 56 days after denervation, suggesting that protection against denervation atrophy is time-related. Furthermore, no decrease in MAFbx or MuRF1 expression was observed at 3, 7, 14, or 31 days when nandrolone failed to prevent denervation atrophy at these time points. This underscores a mechanistic association between lowered MAFbx or MuRF1 levels and attenuated atrophy. Irisin is a 112 amino acid glycosylated protein hormone formed by the proteolysis of FNDC5 (Schumacher *et al.*, 2013). The pivotal role of irisin in rescuing skeletal muscle atrophy was substantiated by the significant increase in the denervated muscle mass following irisin injection. Estimating the protein levels of both MAFbx and MuRF1, it was observed that injection of irisin resulted in a marked reduction in MAFbx and MuRF1 protein levels in denervated muscle. These findings advocate that irisin treatment curbed the expression of key indicators of skeletal muscle wasting during denervation-induced muscle atrophy (Reza *et al.*, 2017).

Phenolic compounds derived from plants deliver diverse health benefits, mitigating the risk of cardiovascular disease and cancer (Kris-Etherton *et al.*, 2002). Moreover, the protective impact of phenolic compounds from olive oil on muscle atrophy has been validated. These compounds can also alleviate the insulin resistance of skeletal muscle induced by a high-fat diet (Fujiwara *et al.*, 2017; Szychlinska *et al.*, 2019). In terms of alkylresorcinols (ARs) in denervated muscle atrophy, dietary alkylresorcinol supplementation was reported to thwart muscle atrophy by restraining the expression of the related genes of the ubiquitin-proteasome and autophagy-lysosomal pathway (Hiramoto *et al.*, 2018). This study demonstrated that AR ingestion prevented denervation-induced hindlimb muscle weight and muscle fiber size reductions. However, ubiquitin ligase and autophagy-related gene expression related to muscle proteolysis were somewhat higher in denervated mice on an ARs-supplemented diet (D-AR) compared to

those on a normal diet (D-ND). Furthermore, the abundance of autophagy marker p62 was significantly higher in D-AR than in D-ND. Fish oil rich in n-3 polyunsaturated fatty acids, including eicosapentaenoic acid and docosahexaenoic acid, is known for maintaining muscle mass. However, numerous studies have underscored fish oil's positive role in various skeletal muscle atrophy models, such as cancer cachexia (Whitehouse et al., 2001), acute starvation (Whitehouse and Tisdale, 2001), sepsis (Khal and Tisdale, 2008), arthritis (Castillero et al., 2009), hind-limb immobilization (You et al., 2010). Komiya et al. (2019) documented that fish oil regulates ubiquitin ligase transcriptional levels through the TNF-α signaling pathway but not the FOXO1 pathway. Nevertheless, dietary fish oil intake has demonstrated no significant effect on skeletal muscle mass loss induced by sciatic nerve denervation (Komiya et al., 2019). Furthermore, BYHWT treatment is also associated with decreased levels of skeletal muscle atrophy-specific molecules MAFbx and MuRF1. This finding aligns with the outcomes of inflammatory response and motor endplate alterations following BYHWT treatment (Zhou et al., 2020). NeuroHeal, a new neuroprotective drug for peripheral nerve injury, discovered using artificial intelligence is based on a combination of two approved drugs, acamprosate and ribavirin, which facilitate its preparation for clinical application. Decreased levels of MAFbX and MuRF1 proteins were evident in the NeuroHeal group in denervated muscles. Denervation-induced autophagy activation characterized by LC3II and p62/SQSTM1 levels was diminished by NeuroHeal, indicating blocked autophagy flux (Marmolejo-Martinez-Artesero et al., 2020).

Resveratrol, a natural phytochemical, is widely found in plants, fruits, and red wine (Zhu et al., 2017). Studies reveal that treatment with 0.5% of the food intake of resveratrol can alleviate muscle atrophy caused by innervation in mice. The reduction of the MAFbx-dependent system and the improvement of autophagy defects are responsible for this attenuation (Asami et al., 2018). Geranylgeraniol (GGOH), as a C20-type isoprene found in fruits, vegetables, and grains, serves as an intermediate product of the Mevalonate pathway and a precursor of Geranylgeranyl pyrophosphate (Muraguchi et al., 2011). Miyawaki et al. (2020) reported that GGOH administration increased muscle fiber size in denervation-induced skeletal muscle atrophy in vivo and curtailed denervation-induced MAFbx expression (Miyawaki et al., 2020). The inhibitory impact of GGOH on skeletal

muscle atrophy could potentially be attributed to its androgen-inhibiting effects, subsequently curbing MAFbx and MuRF1 (Pires-Oliveira et al., 2010). In a hind limb unloading model, administration of quercetin to the gastrocnemius muscle curtailed MAFbx and MuRF1 expression, thereby suppressing skeletal muscle mass loss (Mukai et al., 2010). Isoflavones, natural organic compounds, inhibit denervation-induced apoptosis and muscle atrophy. Studies suggest that isoflavones can suppress MuRF1 transcriptional activity and myotube atrophy (Tabata et al., 2019).

In addition to the aforementioned roles of vitamin E in regulating reactive oxygen species during skeletal muscle atrophy, different vitamins also play significant roles in various stages of denervation-induced skeletal muscle atrophy. Vitamin C deficiency was reported to reduce muscle weight, elevate FOXO-1, MAFbx, and MuRF1 expression. Re-administration of vitamin C could restore muscle weight and lower gene expression in skeletal muscle atrophy (Takisawa et al., 2019). Vitamin D, another member of the vitamin family, participates in calcium absorption, utilization, and bone calcification. However, recent findings suggest its involvement in skeletal muscle functions in different situations. In mice, vitamin D receptor deletion resulted in reduced muscle fiber size. In experiments using C2C12 cells, vitamin D suppressed the expression of cathepsin L and MAFbx (Endo et al., 2003; Hirose et al., 2018). We have summarized the protective effects of drugs in the process of catabolism during denervated muscle atrophy in Fig. 1.

The Effect of Drugs on the Protein Synthesis Pathway

Possibly through activation of the adenosine monophosphate-activated protein kinase pathway, Royal jelly, comprising water, protein, sugar, and lipids, can avert the reduction in skeletal muscle fiber diameter following denervation. Treatment of C_2C_{12} myoblasts with Royal jelly promoted differentiation and proliferation (Shirakawa et al., 2020). In the process of denervated skeletal muscle atrophy, the Notch signaling pathway is activated early after denervation and subsequently declined. Meanwhile, the effect of nandrolone on the Notch signaling ensues following nerve transection, which occurs earlier than the protective effect that prevents continued muscle loss (Liu et al., 2011). In denervated mouse models, dietary intake of 8-prenylnaringenin triggers
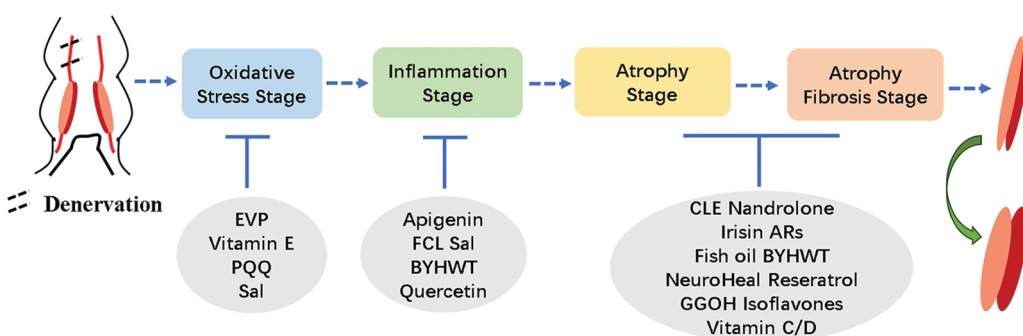

FIGURE 1. Overview of the effects of drugs at different stages of denervated skeletal muscle atrophy.

Akt phosphorylation, thereby potentially alleviating skeletal muscle atrophy (Mukai et al., 2012).

The Effect of Drugs on Other Pathways

Due to its binding affinity to estrogen receptors (ERs), several physiological functions of genistein are attributed to its estrogenic activity (Kuiper et al., 1998). ERs are present in the skeletal muscle, and ER subtypes ER-α and ER-β have a vital role in the differentiation and inflammation of myoblasts (Ogawa et al., 2011; Velders et al., 2012). The intake of genistein exhibits a protective effect on muscle loss caused by innervation. ER-α is activated during soleus muscle atrophy. Therefore, it may be presumed that genistein targets ER-α, thereby exerting a protective effect against muscle atrophy (Aoyama et al., 2016). Nandrolone enhances strength and functional recovery in re-innervated muscles post-establishment of denervation atrophy (Isaacs et al., 2013). Studies have confirmed the efficacy of growth hormone in treating denervated skeletal muscle atrophy. In cases of peripheral nerve injury, Tuffaha et al. (2016) unveiled that growth hormone therapy could expedite axon regeneration in male rats, foster axon myelination, diminish muscle atrophy, and amplify muscle innervation.

TABLE 1

Some important studies for the function of natural drugs or compounds on skeletal muscle metabolism

Different periods of denervated muscle atrophy	Drugs	Description
Reactive oxygen species	Oenothera odorata root extract (EVP)	EVP repressed SOD1 expression and increased HSP70 expression in H_2O_2-treated C_2C_{12} myoblasts and sciatic-neutralized mice. In addition, EVP regulates apoptotic signals, including caspase-3, Bax, Bcl-2, and ceramides
	Vitamin E	Vitamin E was able to restore glutathione levels and decrease MDA levels, which further substantiated the conclusion that oxidative stress accelerated muscle atrophy
	Pyrroloquinoline quinone (PQQ)	PQQ-mediated reduction in the expression of reactive oxygen species and inflammatory factors results in a decrease in the expression of MuRF1 and MAFbx, ultimately improving denervated skeletal muscle atrophy and impairs denervation-induced skeletal muscle atrophy by activating PGC-1α and integrating mitochondrial electron transport chain complexes
	Salidroside	Salidroside reduced denervation-induced oxidative stress and inflammation, inhibiting muscle proteolysis and ultimately alleviating muscle atrophy
Pro-inflammatory factors	Apigenin	In the denervated gastrocnemius and soleus muscle, the upregulation of TNF-α and IL-6 expression was impeded by apigenin treatment. Apigenin has inhibitory effects on atrogin-1/MAFbx expression and prevents reduction in myotube diameter induced by lipopolysaccharide (LPS) stimulation in the C_2C_{12} murine cell line
	Ficus carica L. (FCL.)	FCL is involved in hindering IL-1β and IL-6 production in atrophic muscles and suppresses inflammation in atrophic muscle by inhibiting nuclear factor NF-κB activation
	Quercetin	Quercetin limited inflammation in myotubes, whereas completely restored TNF a-induced reduction of myotube diameter
	Buyang Huangwu Tang (BYHWT)	BYHWT improved inflammatory response when administered in denervated-dependent skeletal muscle atrophy rat models
	Salidroside	Inhibition of muscle proteolysis and muscle atrophy results from salidroside by reducing the inflammatory response caused by innervation, which may be carried out by inactivating the STAT3/SOCS3 pathway
Ubiquitin proteasome and autophagy lysosome pathway	Clenbuterol (CLE)	In vivo and in vitro treatment with CLE was found to prohibited the transcriptional upregulation of atrophy-related Ub ligases in denervated soleus muscles
	Nandrolone	Administration of Nandrolone decelerates denervation atrophy by repressing MAFbx and MuRF1
	Irisin	Injection of irisin resulted in a marked reduction in MAFbx and MuRF1 protein levels in denervated muscle
	Alkylresorcinols (ARs)	The expression of ubiquitin ligase and autophagy-related genes related to muscle proteolysis was slightly higher in denervated mice fed a diet supplemented with ARs than in denervated mice fed a normal diet

(Continued)

TABLE 1 (continued)

Different periods of denervated muscle atrophy	Drugs	Description
	Fish oil	Fish oil regulates the transcript level of ubiquitin ligases via TNF-α signaling rather than the FOXO1 pathway
	Buyang Huangwu Tang (BYHWT)	Compared to the model group, reduced levels of skeletal muscle atrophy-specific molecules MAFbx and MuRF1 were also documented after BYHWT treatment
	NeuroHeal	Decreased levels of MAFbx and MuRF1 proteins were evident in the NeuroHeal group in denervated muscles. Denervation-mediated activation of autophagy is characterized by LC3II and p62/SQSTM1 levels, while the reduction promoted by NeuroHeal signifies that autophagy flux is blocked
	Resveratrol	Treatment with 0.5% of the food intake of resveratrol can alleviate muscle atrophy in mice caused by innervation. The reduction of the MAFbx-dependent system and the improvement of autophagy defects are responsible for this attenuation
	GGOH	GGOH administration increased the muscle fiber size in denervation-induced skeletal muscle atrophy *in vivo*, and it also suppresses the denervation-induced MAFbx expression
	Isoflavones	Isoflavones can suppress the transcriptional activity of MuRF1 and myotube atrophy
	Vitamin C	Vitamin C deficiency reduces muscle weight and increases the expression of FOXO-1, MAFbx, and MuRF1
	Vitamin D	Deletion of vitamin D receptor reduces muscle fiber size and vitamin D suppressed the expression of cathepsin L and MAFbx
Protein synthesis pathway	Royal jelly (RJ)	The RJ can prevent the decrease in skeletal muscle fiber diameter following denervation and promote C_2C_{12} myoblasts differentiation and proliferation
	Nandrolone	The Notch signaling pathway is activated early after denervation and subsequently declined, meanwhile, the effect of nandrolone on Notch signaling ensues following nerve transection, which occurs earlier than the protective effect that prevents continued muscle loss
	8-Prenylnaringenin	In denervated mouse models, dietary ingestion of 8-Prenylnaringenin activates Akt phosphorylation and thus, can relieve skeletal muscle atrophy
The other pathways	Genistein	The intake of genistein exhibits a protective effect on muscle loss caused by innervation, which may be targeted ER-α, exerting a protective effect against muscle atrophy

Nonetheless, further research is crucial to unveil the precise molecular mechanism (Tuffaha *et al.*, 2016). Bortezomib retards the atrophy of rat thyroid muscle and posterior circular ganglion muscles caused by innervation, though the specific mechanism has not been clarified (Sei *et al.*, 2015). Finally, we summarized the pharmacological protective effects mentioned in the review on denervated skeletal muscle atrophy and present them in Table 1.

Conclusion

Comprehending the intricate pathophysiological course of denervated skeletal muscle atrophy is significant in uncovering more drugs or methodologies to combat this condition. In this review, we introduced the proposition from the previous phase: denervated skeletal muscle atrophy can be categorized into four stages as a foundation and summarized various drugs reported as effective for addressing denervated skeletal muscle atrophy across different stages. Our prior investigations also highlighted the pivotal roles of salidroside, isoquercitrin, and PQQ in alleviating denervated skeletal muscle atrophy by modulating distinct signaling pathways or molecules. However, comprehensive research is imperative to ascertain the ongoing efficacy of these drugs.

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