Review

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Sarcopenia from mechanism to diagnosis and treatment in liver disease

Srinivasan Dasarathy[1,*], Manuela Merli[2]

Summary

Sarcopenia or loss of skeletal muscle mass is the major component of malnutrition and is a frequent complication in cirrhosis that adversely affects clinical outcomes. These include survival, quality of life, development of other complications and post liver transplantation survival. Radiological image analysis is currently utilized to diagnose sarcopenia in cirrhosis. Nutrient supplementation and physical activity are used to counter sarcopenia but have not been consistently effective because the underlying molecular and metabolic abnormalities persist or are not influenced by these treatments. Even though alterations in food intake, hypermetabolism, alterations in amino acid profiles, endotoxemia, accelerated starvation and decreased mobility may all contribute to sarcopenia in cirrhosis, hyperammonemia has recently gained attention as a possible mediator of the liver-muscle axis. Increased muscle ammonia causes: cataplerosis of α-ketoglutarate, increased transport of leucine in exchange for glutamine, impaired signaling by leucine, increased expression of myostatin (a transforming growth factor beta superfamily member) and an increased phosphorylation of eukaryotic initiation factor 2α. In addition, mitochondrial dysfunction, increased reactive oxygen species that decrease protein synthesis and increased autophagy mediated proteolysis, also play a role. These molecular and metabolic alterations may contribute to the anabolic resistance and inadequate response to nutrient supplementation in cirrhosis. Central and skeletal muscle fatigue contributes to impaired exercise capacity and responses. Use of proteins with low ammoniagenic potential, leucine enriched amino acid supplementation, long-term ammonia lowering strategies and a combination of resistance and endurance exercise to increase muscle mass and function may target the molecular abnormalities in the muscle. Strategies targeting endotoxemia and the gut microbiome need further evaluation.

Keywords: Sarcopenia; Hyperammonemia; Myostatin; mTORC1; Leucine; Clinical outcomes.

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Introduction

Malnutrition in liver disease has been used for decades to describe the phenotype of skeletal muscle loss with or without loss of fat mass [1]. The majority of "malnourished" patients with cirrhosis experience skeletal muscle wasting or sarcopenia, a major predictor of adverse clinical outcomes [2–4]. Although alterations in body composition in cirrhosis have been reported using a number of methods, radiographic image analysis is believed to be the most precise technique to quantify muscle mass and define sarcopenia [5,6]. Over the past few years, a number of investigators have reported that sarcopenia occurs in 30–70% of cirrhotic patients [2,7–10]. The clinical significance of sarcopenia in liver disease, primarily cirrhosis, is due to the high prevalence and adverse impact on clinical outcome measures including survival, quality of life, development of other complications of cirrhosis, and post liver transplant outcomes [1,4,10–14]. Etiology and severity of the underlying liver disease, duration of illness, age and co-morbidities contribute to the severity of sarcopenia [1,4,9,15,16]. Despite being widely recognized as a major complication of cirrhosis, most therapies to date are based on the principle of "deficiency replacement" rather than targeted treatments, and have generally been ineffective [17]. Nutritional supplementation has been a particular therapeutic focus because reduced dietary intake was believed to be the major cause of malnutrition and sarcopenia. However, these approaches have been frequently inadequate in improving survival [18–20]. An integrated metabolic-molecular approach in a comprehensive array of models has shown that hyperammonemia is a mediator of the

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Key point

Sarcopenia is a frequent complication in cirrhosis. It is the major component of malnutrition and is not reversed after liver transplantation; in fact, it may worsen.

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liver-muscle axis [21,22]. Physical activity has been suggested to improve functional capacity but the effect on skeletal muscle mass is still unclear [23]. In recent years, a combination of sarcopenia with obesity has been increasingly recognized, especially in patients with non-alcoholic fatty liver disease (NAFLD) and post liver transplantation. Whether sarcopenia is mechanistically related to obesity and NAFLD, however, is still under debate [24,25]. The major deficiency in the field of sarcopenia in cirrhosis is the lack of understanding of the mechanisms involved. A number of excellent recent reviews have described the clinical relevance of sarcopenia in cirrhosis but have not focused on the possible mechanisms and on the relevance of novel therapeutic targets that have the potential for clinical translation [1,17,26–29].

In the present review we will provide an overview of the clinical relevance of sarcopenia in liver cirrhosis, but the emphasis will be on the possible molecular and metabolic perturbations involved and the promising novel therapeutic approaches that could be made possible by these discoveries.

Diagnosis of sarcopenia in cirrhosis

Most studies to date have used the term "malnutrition" to identify primarily skeletal muscle loss determined by one or more criteria that are not always uniform or precise and an alteration in energy metabolism and potentially fat mass depletion. The diagnosis of skeletal muscle loss requires analysis of the body composition using one or more of a number of available techniques (Table 1) as well as the normal values to define the appropriate cut-off values for sarcopenia [3,6,29]. Even though few studies have directly compared different methods, computed tomography (CT; Supplementary Fig. 1) with one of the image analysis programs is being increasingly used since skeletal muscle can be directly viewed and quantified [5,10,30–33]. Magnetic Resonance Imaging (MRI) has also been proposed as a valuable method although objective data in cirrhosis are scarce [34]. Abdominal CT and MRI scans would be difficult to justify for quantifying muscle mass due to the cost and/or radiation exposure. However, most cirrhotic patients have surveillance scans for focal liver lesions, hepatocellular carcinoma, vascular disease and pre-transplant evaluation.

Muscle mass depends on gender (lower in females) and age (lower with increasing age), and cut-off values for gender and age have been recently reported [32]. Handgrip strength (a measure of muscle function) has been utilized in cirrhotic patients, but it may not be accurate when normalized to body mass index in cirrhosis due to fluctuations body water content.

Quantifying muscle mass by measurements in a single anatomic area like the limb or abdominal muscles are believed to provide a reasonably accurate measure of whole body muscle mass [35]. In cirrhosis, as in most chronic diseases, a preferential loss of type II or fast fibers is expected but *in vivo* measurements of fiber type loss in cirrhotic patients is still lacking. Appendicular muscle mass (limb muscles) is strongly influenced by the activity level. Measurements of psoas and abdominal muscle mass on CT images at L3 or L4 vertebra are used due to their relative independence from the activity level. However these muscles contain both type I and type IIA fibers [36], which also needs to be considered. Another consideration is the quality of skeletal muscle that has been reported based on the CT attenuation that is lower in the muscles of cirrhotics compared to controls [31] and is indicative of fatty infiltration with adverse clinical outcomes [37,38]. Whether muscle quality can be determined by measuring contractile function or by the CT attenuation values needs to be ascertained (Table 2). The possible impact of these parameters on clinical outcomes has not been systematically evaluated.

Clinical impact of sarcopenia in cirrhosis

A number of cross sectional and longitudinal studies using different methods to quantify muscle mass have reported that median survival and probability of survival are lower in patients who have cirrhosis with sarcopenia than those without sarcopenia (Table 2) [7,9,10,33,39–52]. Some of these reports suggest that sarcopenia adds to the prognostic value of the model for end-stage liver disease (MELD) scoring system [40,53]. The cause(s) of higher mortality is however not as evident though both increased risk of infection and encephalopathy may be contributory factors [54]. Sarcopenia may also impair diaphragmatic work due to reduced muscle mass and this event may favor pulmonary complications especially in the context of surgery (liver resection or liver transplantation).

Sepsis related mortality is higher in sarcopenic than non-sarcopenic cirrhosis [10,13,55]. For appropriate antibody and cytokine responses, adequate amino acid supply is necessary that is impaired when skeletal muscle mass is decreased but a direct causal or mechanistic link between sarcopenia and impaired immune function has not been shown [56]. Furthermore, it is also possible that factors that cause sarcopenia, including hormonal and biochemical alterations as well as circulating endotoxins, also contribute to the impaired immune function and increase the risk of infection. Lack of mobility or frailty in sarcopenia may also play a role [57]. Interestingly, cirrhotic patients with refractory ascites seem particularly prone to malnutrition and sarcopenia. Ascites is known to increase resting energy expenditure [58] while food intake is decreased due to raised abdominal pressure and early satiety. Treating refractory ascites by

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transjugular intrahepatic portosystemic shunt has been shown to improve body composition in malnourished cirrhotic patients [6,31].

Quality of life is lower in sarcopenic cirrhosis patients, but it is unclear whether this is due to the loss of muscle mass or impaired contractile function and subsequent limited mobility, or increased risk of other complications. This is still a field that needs well-designed studies [1,11,12]. All domains of the quality of life are lower in malnourished patients when measures that primarily quantify skeletal muscle mass are utilized [1].

Hepatocellular carcinoma (HCC) is a frequent complication in the natural history of chronic liver disease and recent studies have reported that sarcopenia is an independent prognostic factor decreasing survival and increasing treatment related mortality in patients with HCC [37,59].

Liver transplantation is currently the definitive therapy to cure end-stage liver disease and sarcopenia adversely impacts outcomes in patients on the transplant list, in the peri-transplant period and post transplantation [7,9,45,60]. Survival is lower in sarcopenic cirrhotic patients before liver transplantation while increased length of hospitalization, prolonged intensive care unit stay, and longer time of intubation have been reported after transplantation compared to patients without sarcopenia [9,27,45].

It is important to emphasize that clinical outcomes also depend on other factors, but sarcopenia is recognized as a major contributor to adverse outcomes in the management of the cirrhotic patient undergoing liver transplantation.

Mechanisms of skeletal muscle loss in cirrhosis

Alterations in protein turnover, energy disposal and metabolic changes induce muscle depletion in cirrhotic patients

As seen above, a number of studies and reviews have provided descriptive data on the high prevalence and adverse clinical impact of sarcopenia in cirrhosis [1,4,7,10,26,60]. Skeletal muscle is the major protein store in the human body [61]. Skeletal muscle mass is maintained by a balance between protein synthesis, protein breakdown and regenerative capacity regulated by muscle satellite cell function [1]. Broadly, two types of studies have contributed to the current understanding of the pathogenesis of sarcopenia in cirrhosis: metabolic-tracer kinetics and molecular signaling pathway studies [21,22,62–66]. An integrated approach using both strategies to examine how metabolic perturbations alter molecular signaling and vice-versa has allowed identification of novel potential therapeutic targets.

Whole body turnover studies using labeled phenylalanine and leucine as primed constant infusion have yielded conflicting results with unaltered, increased or decreased protein breakdown and protein synthesis [62–64]. Arteriovenous difference studies and release of 3-methylhistidine to quantify protein synthesis and breakdown suggest impaired skeletal muscle protein synthesis [67]. Explanations for these conflicting observations included heterogeneity in etiology, duration, age, and severity of liver disease. Heterogeneity in methods used to determine protein turnover and in the contribution of different organs to whole body turnover also explain these differences. Whole body substrate utilization studies using indirect calorimetry have shown that cirrhosis is a state of accelerated starvation because fatty acid oxidation and gluconeogenesis are increased early in the postabsorptive or fasting state [30,68,69]. Since glucose is a preferred substrate in many tissues, and fatty acid carbon cannot be used for gluconeogenesis, amino acids are used for gluconeogenesis [70]. The primary source of amino acids for gluconeogenesis is proteolysis in the skeletal muscle that generates both aromatic and branched chain amino acids (BCAA). Only BCAA are catabolized in the skeletal muscle due to the localization of the branched chain ketodehydrogenase and oxidation of the carbon skeleton as an energy source [71]. As a consequence, plasma BCAA concentrations are lower in cirrhotic patients. In contrast, aromatic amino acids are primarily metabolized in the liver but due to both portosystemic shunting and hepatocellular dysfunction, their plasma concentrations are increased in chronic liver disease [62,72–75]. This interpretation that accelerated starvation and increased gluconeogenesis are bioenergetics perturbations in cirrhosis is supported by the low respiratory quotient in sarcopenia cirrhotics [30]. Most therapies to date have focused on treating the amino acid imbalance rather than targeting the mechanisms that contribute to these alterations that finally result in sarcopenia.

Table 1. Methods to quantify skeletal muscle evaluation in cirrhosis.

	Methods for quantification
Single muscle or groups of muscle	Anthropometry, DEXA, bioelectrical impedance analysis, impedance plethysmography, ultrasonography, CT or MRI,
Quality of muscle	CT scan attenuation
Muscle function	Handgrip strength
Fiber type	Muscle biopsy
Contractile function	Measurement of maximum strength, maintenance of strength, fatigability

CT, computed tomography; DEXA, dual energy X-ray absorptiometry; MRI, magnetic resonance imaging.

Potential mediators of the liver – muscle axis in cirrhosis

One of the major reasons for the very limited understanding of sarcopenia in cirrhosis has been the difficulty in identifying the mediator(s) of the liver-muscle axis. A number of potential mediators have been proposed including increased ammonia, decreased testosterone and growth hormone, and endotoxemia [21,22,76,77]. Even though there is evidence to support each of these potential media-

Key point

Other perturbations that contribute to sarcopenia include endotoxemia, increased aromatase activity to lower testosterone, and mitochondrial dysfunction.

Review

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Table 2. Sarcopenia adversely impacts outcome in cirrhosis.

Author (year)	N	Method to define sarcopenia	Outcome
Wang (2016) [39]	292	Effect of grip strength, muscle mass, muscle quality, SPPB on transplant wait list mortality	Grip strength (HR 0.74), SPPB (HR 0.89), muscle quality (0.77) but not muscle mass (0.91) decreased survival
Kalafateli (2016) [40]	232	L3 psoas area (CT) and Royal Free Hospital Global Assessment	Post OLT infection (OR 6.55), ventilator requirement (OR 8.5), ICU stay >5 d (OR 7.46) higher in sarcopenic patients.
Hanai (2016) [41]	149	CT measure of psoas muscle area at L3/L4	Greater rate of muscle (>3.1%/year) loss increases mortality (HR 2.73)
Kim (2014) [42]	89		Increased mortality hazard risk 5.4 for sarcopenia
Masuda (2014) [43]	204		Sarcopenia 2 fold increased risk of death, 5.3 fold increased risk of sepsis
DiMartini (2013) [44]	338		Increased mortality only in men for each unit decrease in skeletal muscle index
Montano-Loza (2012) [10]	112		Increased mortality hazard risk 2.26 for sarcopenia
Tandon (2012) [7]	142		Increased mortality, hazard risk 2.36 for sarcopenia
Englesbe (2010) [45]	163		Lower post OLT survival, HR 3.7/1000 mm^2 psoas area.
Durand (2014) [33]	376	CT measure of psoas at umbilicus	Increased mortality for each unit decrease in muscle area
Hamaguchi (2014) [46]	200		Median post OLT survival in sarcopenic patients 17.6 m and in non-sarcopenic patients 33.9 m
Hara (2016) [47]	161	Bioelectrical impedance analysis	73 deaths over mean 1005 days follow up
Kaido (2013) [48]	124		Post living donor transplant lower with sarcopenia
Selberg (2002) [142]	305		Survival lower with phase angle <5.4°
Merli (2010) [9]	38	Anthropometrics (MAMA/TSF)	MAMC <5th percentile: relative risk of death 1.79.
Shahid (2005) [49]	61		Increased postoperative mortality.
Lai (2014) [50]	50	Frailty index, SPPB	45% greater mortality for each point increase in frailty index 19% increase in mortality for each point decrease in physical performance
Carey (2010) [51]	294	6 min walk test	Each 100 m reduction in 6 min walk test reduces survival hazard risk 0.48
Alvares da-Silva (2005) [52]	121	Hand grip, anthropometrics	Increased mortality for lower hand grip strength

CT, computed tomography; HR, hazard ratio; ICU, intensive care unit; MAMA, mid arm muscle area; OR, odd ratio; SPPB, short physical performance battery; TSF, triceps skinfold thickness.

Key point

Hyperammonemia mediated upregulation of myostatin is believed to be a mechanism of impaired protein synthesis and increased autophagy, that contribute to sarcopenia.

tors, hyperammonemia has been studied most extensively [17,21,22,78].

Of the hepatic metabolic functions, ammonia disposal by ureagenesis is critical. Both hepatocellular dysfunction and portosystemic shunting that are components of the pathophysiological changes in cirrhosis contribute to impaired ureagenesis [79]. Ammonia is generated by a number of mechanisms including amino acid metabolism, purine metabolism, enterocyte glutaminase activity and urealysis in the gut [80]. Neurotoxicity is the best-studied cytotoxic effect of ammonia [80,81]. Independent investigators have reported increased skeletal muscle ammonia uptake and conversion to glutamate and glutamine in patients and models of liver disease [82–85]. Despite the well recognized cytotoxic effects of ammonia in the neurons and astrocytes, skeletal muscle effects have only been recently reported [21,22,86,87]. Studies in human skeletal muscle, the hyperammonemic portacaval anastomosis (PCA) rat, mice during hyperammonemia and *in vitro* studies in myotubes cultures suggest that ammonia accumulates in the skeletal muscle and activates a program of molecular alterations that contribute to sarcopenia [21,22,86,87]. Even though the mechanism of entry of ammonia into the skeletal muscle has not been well studied, ammonia transporters including the Rh B and C proteins are expressed in the muscle [88]. Following entry, ammonia activates a series of signaling responses whose exact mechanisms are as yet unclear.

Hyperammonemia contributes to muscle depletion: intracellular signaling

In murine myotubes and murine cells cultures, the response to hyperammonemia-mediated activation of p65-NF-κB is an increased expression of myostatin, a TGFβ superfamily member (Fig. 1) [22,89]. Increased expression of myostatin in the skeletal muscle and plasma of cirrhotic patients has been reported [89,90] and these results should be confirmed in future studies. Myostatin is a known inhibitor of protein synthesis and potentially activates the ubiquitin proteasome and autophagy mediated proteolysis [21,22,91]. The ubiquitin-mediated proteolysis is not activated but autophagy has been found to be increased in muscle in experimental models of cirrhosis or during hyperammonemia [21,22]. Other potential mechanisms for activation of autophagy include ammonia mediated mitochondrial dysfunction and generation of reactive oxygen species [92]. Even though these molecular signaling responses have been reported only in neural tissue, similar perturbations may also occur in the skeletal muscle [93].

Interestingly, skeletal muscle metabolic responses to hyperammonemia are being increasingly recognized albeit in preliminary data [93]. Physiologically, glutamine and glutamate serve as anaplerotic

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substrates to generate α ketoglutarate (αKG) and ammonia in most tissues to maintain sufficient concentrations of the tricarboxylic acid (TCA) cycle intermediates [94]. This reaction is catalyzed by the bidirectional enzyme, glutamate dehydrogenase (GDH). The reaction preferentially occurs in the direction generating αKG, because the GDH Km for ammonia is very high (~1 mM), a value that is significantly supraphysiological [95]. However, in cirrhosis, due to impaired ureagenesis and decreased hepatic ammonia disposal, the skeletal muscle functions as a metabolic partner for the liver and skeletal muscle ammonia concentrations are much higher potentially favoring cataplerosis or loss of critical TCA cycle intermediate, αKG [22]. This results in a number of potential consequences including lower flux of the TCA cycle, impaired mitochondrial function and decreased adenosine triphosphate (ATP) synthesis. Since protein synthesis, especially translation initiation, is an energy intense process, low ATP concentrations may also cause reduced protein synthesis. Another consequence of hyperammonemia that can explain a number of clinical observations is that oxodehydrogenases are inhibited by ammonia in a tissue specific manner [96]. These include pyruvate dehydrogenase, that catalyzes the conversion of pyruvate to acetyl coenzyme A (CoA), and αKG dehydrogenase that catalyzes conversion of αKG to succinyl CoA. An overview of these pathways is shown in Fig. 2. A number of clinical studies and meta-analyses have failed to show significant benefit of nutritional supplementation in malnourished cirrhotic patients [18–20,26]. This may be due to the impaired acetyl CoA generation that necessitates formation of acetyl CoA from non-pyruvate sources including amino acids and fatty acids. Continued mitochondrial dysfunction, generation of reactive oxygen species, and impaired bioenergetics in the skeletal muscle all contribute to impaired protein synthesis and activate a metabolic, adaptive response, autophagy.

Reduced ATP in the muscle, impaired mitochondrial function, low concentrations of TCA cycle intermediates, increased gluconeogenesis and an increased fatty acid oxidation in the skeletal muscle during hyperammonemia suggest a bioenergetics crisis with a starvation like response. Decreased cellular ATP is consistent with activation of the cellular energy sensor, 5' adenosine monophosphate-activated protein kinase (AMPK) and impaired mTORC1 signaling [66].

Increased cataplerosis and muscle catabolism of branched chain amino acids as a source of energy may be responsible for low circulating branched chain amino acids with skeletal muscle concentrations of BCAA expected to be decreased in the muscle of cirrhotics due to increased utilization. Reduced cellular amino acid concentrations activate adaptive responses that include increased skeletal muscle autophagy that has been reported in both cirrhosis and hyperammonemia in myotubes. Another response to intracellular amino acid deficiency is the integrated stress response mediated by activation of amino acid deficiency sensor, general control non-depressed 2 (GCN2) via phosphorylation of eukaryotic initiation factor 2 that are increased during hyperammonemia and cirrhosis [93]. Surprisingly, skeletal muscle concentrations of branched chain have been mostly reported to be unaltered except for a single study that reported lower muscle concentrations of BCAA [73–75]. Preliminary studies in hyperammonemic myotubes increased cellular transport and concentrations of leucine despite which supplementation with leucine enriched BCAA resulted in reversal of GCN2 activation. This rescued impaired mTORC1 signaling in patients with cirrhosis [66] and in myotubes during hyperammonemia. Other amino acids with therapeutic potential include L citrulline that is a precursor for L arginine and stimulates mTORC1 and protein synthesis [97]. The beneficial effects of citrulline are believed to be due to decreased ureagenesis resulting in amino acid sparing, but it is not known if impaired ureagenesis will aggravate hyperammonemia and its consequences in cirrhosis and need to be studied systematically.

Published data suggest that hyperammonemia is a mediator of the liver-muscle axis and the skeletal muscle does not function only as a metabolic sink for ammonia [22]. Ammonia uptake and disposal via glutamine synthesis in the muscle and transport into the circulation may be involved in sarcopenia. At the same time, if there is low muscle mass, non-hepatic disposal of ammonia is impaired which may cause further adverse effects. Consistently, some investigators have reported that encephalopathy is more frequent in sarcopenic than non-sarcopenic cirrhotics [12,14].

Other potential mediators of the liver – muscle axis in cirrhosis: testosterone, growth hormone

Other mediators of the liver-muscle axis include the low testosterone due to increased aromatase activity in liver disease [98]. Decreased growth hormone concentrations or impaired growth hormone response in the muscle are also likely contributors to sarcopenia in cirrhosis [99,100]. Both growth hormone and testosterone are known to inhibit myostatin expression and signaling responses [101,102] but it is not known if these hormonal alterations of cirrhosis also contribute to the impaired protein synthesis and increased myostatin expression in cirrhosis. A recent randomized trial showed that testosterone supplementation in male cirrhotics did result in an increase in lean body mass but not survival [103].

Hepatocellular and immune dysfunction as well as portosystemic shunting worsen the endotoxemia due to impaired gut barrier function and potentially altered gut microbiome in cirrhosis [104].

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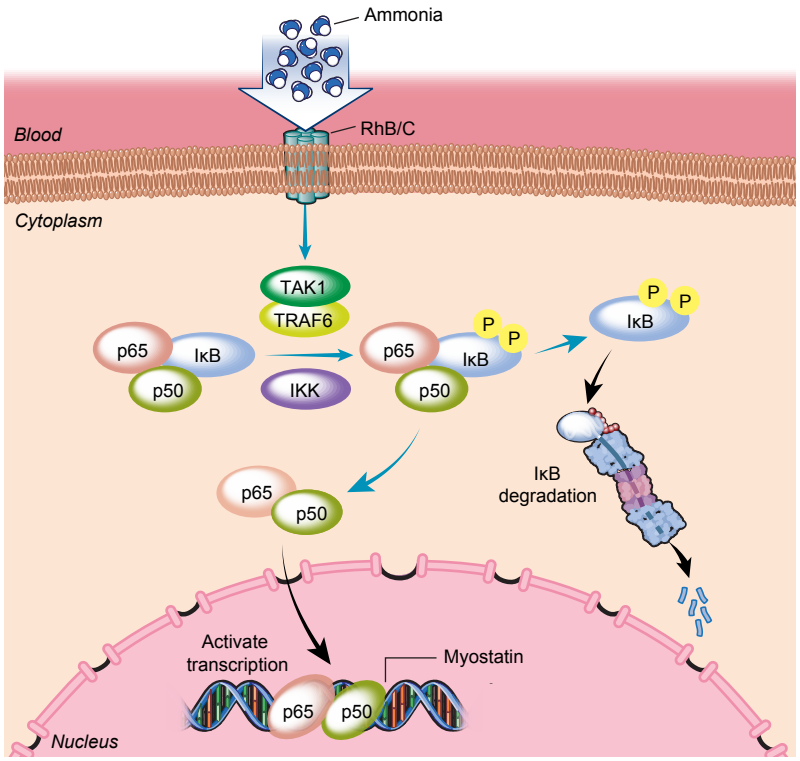

Fig. 1. Myostatin is transcriptionally upregulated by hyperammonemia in the skeletal muscle. Ammonia enters the skeletal muscle via the transport proteins Rh B and G. In the muscle, ammonia activates transforming growth factor β activated kinase 1 (TAK1) that activates TRAF6. Activated TRAF6 (k63 polyubiquitination) activates inhibitor of kappa B (IκB) kinase (IKK) that in turn phosphorylates nuclear factor kappa B (NF-κB) inhibitor protein IκB. Phospho IκB is degraded via a proteasome pathway releasing p65-NF-κB that enters the nucleus and transcriptionally upregulates myostatin.

Endotoxemia via tumour necrosis factor (TNF)α dependent and potentially TNF independent pathways may also impair protein synthesis and potentially activate autophagy [105,106]. Careful molecular studies on these mediators are not available and the cross talk between hyperammonemia and other putative mediators such as those described above are not presently known. The next decade is likely to see major advances in our understanding of the molecular-metabolic interaction and how it contributes to or causes sarcopenia in liver disease.

Finally, sarcopenic obesity has been reported in patients with NAFLD and after liver transplantation [24,25,107]. It is possible that the combination of skeletal muscle loss and increased fat mass may contribute to the development of metabolic components including insulin resistance, diabetes mellitus, hyperlipidemia and possibly NAFLD but whether there is a common underlying mechanism for both sarcopenia and obesity is still not known [108].

Management strategies

There is compelling evidence that sarcopenia is associated with adverse consequences while there are limited data showing that increasing muscle mass improves survival in the non-transplanted and post liver transplant population of cirrhotics [31,32]. Therefore, reversing muscle mass is a priority area for therapeutic interventions in cirrhotic patients (Fig. 3). Interventions that focus only on deficiency replacement have generally been ineffective while targeted therapies have the potential to reverse muscle loss [1,18–20,26,66,87]. The major strategies that have been used to improve muscle mass include supplemental calorie and protein intake, increased physical activity, supplemental hormone therapy, and mechanistic targeted treatments [17,26,109–111]. The critical outcome measures include survival, hospitalization, quality of life, development of and recovery from other complications of cirrhosis. It is not clear if the improved clinical outcomes are due to an increase in muscle mass, amelioration in skeletal muscle contractile dysfunction or a combination of the two. Despite the current focus being on reversing sarcopenia, it is also important to take into consideration skeletal muscle function that include maximum contractile strength, maintenance of contraction, and muscle fatigue in response to persistent and repetitive contraction [78].

Supplemental nutrition

Since caloric and protein intake are frequently decreased in cirrhosis, Guidelines and Consensus papers have consistently recommended to provide adequate amounts of calories and proteins either by frequent feeding, through oral dietary supplementation or when indicated, by enteral or parenteral nutrition [112–115]. Regimens providing extra calories via high caloric feeding, and/or enteral feeding have been extensively studied (Table 3) [114,116–123]. Interestingly, few studies suggest improvement in nitrogen retention or nutritional status using very heterogeneous criteria that measure primarily fat and non-fat mass [17,19,20,122,124,125]. On the other hand, a recent randomized controlled trial measured total body protein utilizing perioperative immunonutrition enriched in n-3 fatty acids, arginine, and nucleotides *vs.* an isocaloric diet in patients undergoing liver transplantation. Protein content was measured by neutron activation analysis, from study entry until immediately prior to LT but did not find any change in total body protein. Postoperative outcomes were also not influenced by the nutritional supplementation [114].

Another approach has been to shorten the duration of post-absorptive or fasting state in cirrhosis because of the accelerated starvation that results in proteolysis, because after food intake, recovery of muscle mass is incomplete [68]. Daytime and nocturnal feeding have been evaluated and there is evidence that a late evening snack has the most beneficial effects and it is currently believed that a late evening and an early morning protein supplement

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are likely to have the greatest benefit on preventing continued muscle loss in cirrhosis [68,126]. Meta analyses of supplemental nutrition in patients with alcoholic hepatitis and those with cirrhosis were disappointing, however, as nutritional supplementation by various routes did not improve survival [18–20]. Even though the exact reason for very limited improvement in sarcopenia with nutritional supplementation is not yet clear, cirrhosis can be seen as a state of anabolic resistance and caloric supplementation alone seems to be inadequate. As mentioned earlier, despite providing calories, impaired mitochondrial function and bioenergetics in combination with impaired molecular responses to nutrient administration in muscle are potential reasons for lack of benefit. Whether other outcomes including encephalopathy, sepsis and quality of life improve with reversal of sarcopenia are currently unknown.

Protein supplementation is another alternative to improve the availability of essential amino acids. However, cirrhosis and hyperammonemia may accelerate amino acids catabolism with further generation of skeletal muscle ammonia that can impair protein synthesis and increase autophagy further with little or no benefit in reversing sarcopenia. Animal proteins have the added disadvantage of being rich in aromatic amino acids that are not metabolized by the skeletal muscle and may worsen encephalopathy [72,127]. Vegetable proteins are rich in BCAA and may have a beneficial effect by removing one mole of ammonia per mole of BCAA via the αKG → glutamate → glutamine pathway. Therefore, instead of protein supplementation, BCAA have been used in the past as treatment for hepatic encephalopathy in a number of acute and long-term studies [128–130]. A recent Cochrane review suggested benefit in the primary outcome, hepatic encephalopathy but not on survival, quality of life or nutritional parameters [131]. Lack of benefit in nutritional parameters was counter to expected outcomes, since BCAA provide a source of energy to the muscle in addition to being substrates for protein synthesis. Another mechanism by which BCAA may function is by inhibiting the amino acid deficiency sensor, GCN2 and reversing eIF2α phosphorylation [132], impaired protein synthesis and improve muscle mass. Finally, leucine directly activates mTORC1 that stimulates protein synthesis and decreases autophagy [133], both of which have the potential to improve muscle mass. A recent study in human cirrhosis reported that a leucine enriched BCAA mixture was able to reverse the molecular perturbations in the skeletal muscle downstream of myostatin in cirrhotic patients [66]. Tracer kinetic studies with direct quantification of muscle protein synthesis showed similar rates of protein synthesis in response to a single oral dose of leucine enriched BCAA mixture did reverse the GCN2-eIF2α mediated impaired protein synthesis and increased

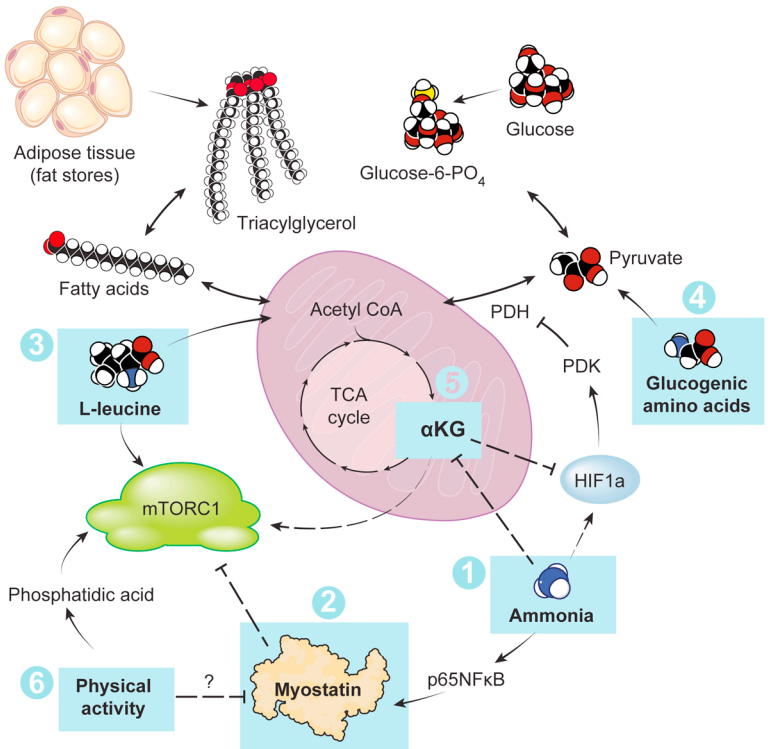

Fig. 2. Biochemical abnormalities in the skeletal muscle that contribute impaired protein synthesis and increased autophagy with consequent sarcopenia. Metabolic and molecular perturbations that can be potentially reversed by intervention at targeted sites. 1. Long-term ammonia lowering strategies. 2. Myostatin blocking agent including antagomirs. 3. L-leucine provides acetyl CoA, activates mTORC1 and protein synthesis. 4. Glucogenic amino acids can be a source of anaplerotic input to provide succinyl CoA replacing the loss of (cataplerosis) of αKG that is converted to glutamate during hyperammonemia (since skeletal muscle cannot generate urea). 5. Cell permeable esters of αKG are a potential strategy to reverse cataplerosis and a novel method to increase muscle ammonia disposal. 6. Physical activity stimulates mTORC1 via phosphatidic acid.

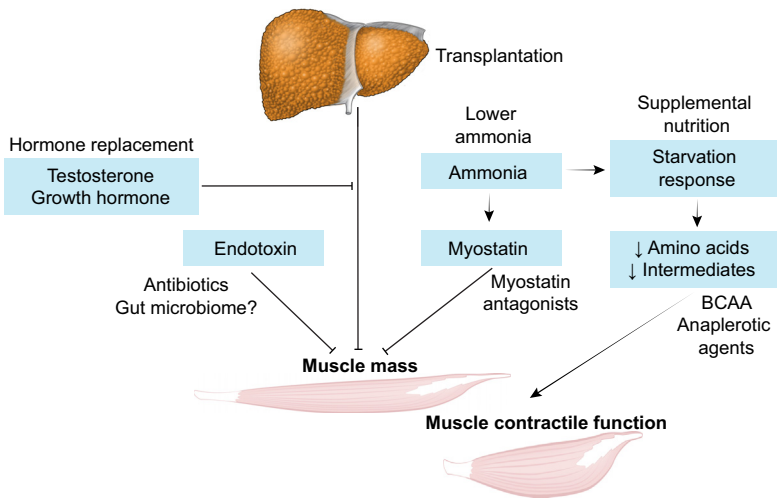

Fig. 3. Overview of strategies to reverse sarcopenia and potentially contractile dysfunction in cirrhosis. Molecular targets are depicted in blue boxes and putative interventions are outside the boxes. Modified from [109] with permission.

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Table 3. Studies about nutritional intervention in adult liver cirrhosis reporting data about changes in parameters dealing with muscle mass.

Author	Treatment	Setting	Duration	Patients (n)	Proteins g/day	Calories kCal/day	Outcome on nutritional parameters	Outcome
Hirsch 1993 [116]	Oral supplement *vs.* control	Cirrhotic patients of alcoholic origin, outpatients	1 year	26 nutritional supplement *vs.* 25 controls	45 ± 10 + 34 g supplement	1580 ± 500 + 1000 kCal supplement	Similar improvement in both groups	Reduced severe infections Reduced hospital admission Similar survival
De Ledinghen 1997 [117]	Short term enteral nutrition *vs.* fasting	Cirrhotic patients after bleeding from esophageal varices	3 days Follow-up 5 weeks	12 enteral *vs.* 10 controls	74 g	2090	No change in nutritional parameters	No change in outcome or rebleeding
Le Cornu 2000 [118]	Oral supplementation to diet *vs.* diet	Malnourished cirrhotic patients in the waiting list for liver transplantation	Variable 77 days	42 supplementation *vs.* 40 controls	80 g	2419 kCal	Treated improved arm circumference and arm muscle circumference + handgrip strength	No difference in outcomes or survival
Marchesini 2003 [119]	Oral supplement of BCAA *vs.* isocaloric and isonitrogen supplement	Advanced cirrhotic outpatients	12 months	59 BCAA *vs.* 56 L-alb *vs.* 56 malto-dextrin	0.8 g/kg/day + BCAA 14 g/day Or L-alb 14 g/day Or no protein supplement	30 kCal/day + 200 kCal	Significant Improvement of mid arm muscle area after BCAA supplement	Lower hospital admission in BCAA
Hu 2003 [120]	Enteral *vs.* parenteral *vs.* controls	Postoperative patients with poor liver function	7 days	65 enteral *vs.* 40 parenteral *vs.* 30 controls	0.16 g N/kg	30 kCal/kg/day	Enteral nutrition caused improved nitrogen balance Minor changes in body weight and arm circumference	Enteral nutrition caused improvement in gut barrier
Dupont 2012 [122]	Enteral 1 month and oral 2 months *vs.* conventional treatment	Alcoholic cirrhotic patients with jaundice but no severe acute alcoholic hepatitis	12 month	44 enteral and oral nutritional supplementation *vs.* 55 controls	Oral diet 60 g Enteral 1.2 g/kg Oral supplement protein 20 g Three times a day	Oral diet 1800 kCal Enteral 30-35/kCal/kg Oral supplement 320 kCal+ Three times a day	No change in arm muscle circumference	Similar complications and survival
Sorrentino 2012 [123]	Parenteral nutrition post paracentesis (PNPS) and late evening snack (LES)	Cirrhotic patients with refractory ascites	12 months	40 PNPS and LES *vs.* 40 only LES *vs.* 40 controls low sodium diet	1.2-1.3 g/kg BW + PN 1.5 g/kg/bw LES 13.5 g/day	30 kCal/kg/day	Maintenance of arm muscle circumference in treated patients *vs.* deterioration of arm muscle circumference at 6 and 12 months in +LES and controls	Lower number of paracenthesis and better survival in patients treated with PN and LES
Plank 2015 [114]	Oral/enteral immune nutrition *vs.* isocaloric control	Before OLT and postoperative	Variable before transplant and 5 days postoperatively Follow-up 12 months	52 immunonutrition *vs.* 49 isocaloric controls	80 g + supplement of 14 g arginine, 4 g omega3 fatty acids, 1.6 ribonucleic acid	1860-1900 kCal	Total body protein unchanged at 12 months	Similar outcomes In both groups

OLT, orthotopic liver transplantation; PN, parenteral nutrition.

mTORC1 signaling [66]. These data provide the first direct evidence of molecular perturbations in the skeletal muscle in cirrhosis and in combination with animal and *in vitro* cell culture data support the role of hyperammonemia as a mediator of the liver-muscle axis.

Exercise and physical activity

The type of exercise determines the muscle related outcomes [134]. Resistance exercise increases skeletal muscle mass by inducing muscle injury and regeneration and protein synthesis [135]. Endurance

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exercise improves functional capacity but does not necessarily reverse sarcopenia. A combination of resistance and endurance exercise have the potential to improve muscle mass and functional capacity but such studies have not been performed in cirrhosis. Randomized studies have reported improvement in short-term outcomes in response to exercise in cirrhotics [23]. Since direct comparisons of outcomes in healthy subjects and cirrhotic patients in response to exercise have not been reported, it is not possible to determine if the anabolic resistance to nutrients is also observed with exercise. There is evidence that protein kinase Cζ –phosphatidic acid mediates signal transduction of mechanical activity to signaling responses by activating mTORC1 signaling and protein synthesis [136]. However, it is not known if these physiological responses are blunted in cirrhosis and if ammonia is the mediator of such blunted responses. A recent study in a comprehensive array of models including hyperammonemic rats, human subjects and *ex vivo* muscle preparations does suggest that hyperammonemia also alters contractile response and increases fatigue in cirrhosis [78]. Whether immobilization and injury responses in the cirrhotic skeletal muscle are altered has not been studied but may explain the rapid deconditioning observed during hospitalization.

Anabolic hormones

Testosterone and growth hormone have been used in the past to improve nutritional status and, potentially, muscle mass in cirrhosis but have not been consistently beneficial [99,100,103,137,138]. Despite adverse effects, these therapies are not effective in reversing nutritional status or sarcopenia. Increased aromatase activity contributes to conversion of testosterone to estradiol that blunts its effect [98]. Aromatase resistant androgens like oxandrolone may therefore be beneficial but have not been borne out in clinical practice [137]. Lack of therapeutic benefit with hormone replacement may also be due to impaired signaling responses including mTORC1 response downstream of androgen and growth hormone receptors may be responsible for failure of these therapies. Increasing the understanding of molecular and metabolic perturbations in the skeletal muscle not only provides explanations for the lack of clinical benefit of standard therapies but also is likely to help identify novel, specific therapeutic targets for reversing sarcopenia.

Ammonia lowering strategies

Current methods to lower ammonia include non-absorbable disaccharides and antibiotics to prevent gut generation of ammonia [139]. The primary outcomes of these treatments are reversal of encephalopathy and lowering of blood ammonia concentrations. It is, however, well known that blood ammonia concentrations do not always correlate with the severity of encephalopathy, the most studied response to hyperammonemia [140]. Skeletal muscle turnover is a slow process and lowering ammonia transiently may not necessarily lower muscle ammonia concentrations or reverse the ongoing metabolic and molecular perturbations rapidly. Studies on long-term ammonia lowering strategies, quantifying muscle ammonia concentrations and signaling responses to these interventions are necessary before such an approach can be used to reverse muscle loss and impaired contractile function. Novel and potential methods to lower muscle ammonia include the use of cell permeable esters of αKG that can provide a direct anaplerotic influx with removal of ammonia as glutamine. However, glutamine disposal will then become limiting and strategies for long-term ammonia disposal to protect the skeletal muscle are necessary. Isoleucine and valine as anaplerotic substrates have been suggested because they can remove one mole of ammonia per mole of amino acid but the molecular and functional responses to these interventions have not been evaluated in preclinical or clinical studies to lower muscle ammonia or reverse sarcopenia [82,83].

Key point

Therapies including nutrient supplementation and exercise are not consistently effective since they target replacing deficiency rather than the underlying mechanisms.

Novel molecular targeted strategies

Myostatin antagonists [91], direct mTORC1 activators [66,133], antioxidants, and mitochondrial protective agents have the potential to benefit skeletal muscle protein turnover but have not been adequately evaluated. Careful mechanistic studies are necessary with preclinical testing before these interventions can be translated to clinical practice.

Post liver transplantation sarcopenia

The underlying molecular mechanisms and mediators need to be ascertained before therapies can be recommended. Direct mTORC1 inhibitors that block protein synthesis responses and accelerate autophagy are largely used after liver transplantation, at least in the United States [17]. Calcineurin inhibits muscle growth and hypertrophy [141] and calcineurin inhibitors are used in the vast majority of post transplant patients. The contribution of these medications to post transplant sarcopenia and sarcopenic obesity needs to be evaluated. Whether anabolic resistance of cirrhosis is reversed by liver transplantation is not known and integrated metabolic-molecular studies with muscle biopsies are needed before specific therapies and preventive measures can be developed. Finally, the reversibility of hyperammonemia induced signaling responses and impaired protein synthesis after liver transplantation is not known. It is possible that epigenetic changes in the regulatory molecules result in long-term or persistent sarcopenia even after transplantation or ammonia lowering therapies.

Key point

Therapies targeting mitochondrial function, including: mitochondrial antioxidants, mTORC1 signaling, and myostatin, hold promise for the future.

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Conclusion

In summary, there is compelling evidence to show that sarcopenia is the major complication of cirrhosis and adversely affects outcomes during the entire course of a cirrhotic patient's life. Evidence to show that sarcopenia can be reversed is much more limited and it is not clear if reversing sarcopenia will indeed improve outcomes as expected. Nutritional supplementation is not consistently effective in improving outcomes but long-term BCAA with leucine are promising therapies to prevent and treat sarcopenia in cirrhosis. Long-term reduction of muscle ammonia, novel approaches to enhance muscle ammonia disposal, and strategies to block myostatin hold potential for the future. Identification of molecular and metabolic perturbations in the cirrhotic skeletal muscle will allow development of targeted therapies that focus in reversing the anabolic resistance in these patients.

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Conflict of interest

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Authors' contributions

Dr. Dasarathy generated the initial draft, edited the manuscript, generated the figures and tables and approved the final draft. Dr Merli assisted with the initial draft, edited the draft, edited the figures and approved the final manuscript.

Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jhep.2016.07.040.

References

[1] Periyalwar P, Dasarathy S. Malnutrition in cirrhosis: contribution and consequences of sarcopenia on metabolic and clinical responses. Clin Liver Dis 2012;16:95–131.
[2] Alberino F, Gatta A, Amodio P, Merkel C, Di Pascoli L, Boffo G, et al. Nutrition and survival in patients with liver cirrhosis. Nutrition 2001;17:445–450.
[3] Merli M, Romiti A, Riggio O, Capocaccia L. Optimal nutritional indexes in chronic liver disease. JPEN J Parenter Enteral Nutr 1987;11:130S–134S.
[4] Merli M, Riggio O, Dally L. Does malnutrition affect survival in cirrhosis? PINC (Policentrica Italiana Nutrizione Cirrosi). Hepatology 1996;23:1041–1046.
[5] Giusto M, Lattanzi B, Albanese C, Galtieri A, Farcomeni A, Giannelli V, et al. Sarcopenia in liver cirrhosis: the role of computed tomography scan for the assessment of muscle mass compared with dual-energy X-ray absorptiometry and anthropometry. Eur J Gastroenterol Hepatol 2015;27:328–334.
[6] Dasarathy J, Alkhouri N, Dasarathy S. Changes in body composition after transjugular intrahepatic portosystemic stent in cirrhosis: a critical review of literature. Liver Int 2011;31:1250–1258.
[7] Tandon P, Ney M, Irwin I, Ma MM, Gramlich L, Bain VG, et al. Severe muscle depletion in patients on the liver transplant wait list: its prevalence and independent prognostic value. Liver Transpl 2012;18:1209–1216.
[8] Campillo B, Paillaud E, Uzan I, Merlier I, Abdellaoui M, Perennec J, et al. Value of body mass index in the detection of severe malnutrition: influence of the pathology and changes in anthropometric parameters. Clin Nutr 2004;23:551–559.
[9] Merli M, Giusto M, Gentili F, Novelli G, Ferretti G, Riggio O, et al. Nutritional status: its influence on the outcome of patients undergoing liver transplantation. Liver Int 2010;30:208–214.
[10] Montano-Loza AJ, Meza-Junco J, Prado CM, Lieffers JR, Baracos VE, Bain VG, et al. Muscle wasting is associated with mortality in patients with cirrhosis. Clin Gastroenterol Hepatol 2012;10:166–173 173 e161.
[11] Shiraki M, Nishiguchi S, Saito M, Fukuzawa Y, Mizuta T, Kaibori M, et al. Nutritional status and quality of life in current patients with liver cirrhosis as assessed in 2007–2011. Hepatol Res 2013;43:106–112.
[12] Huisman EJ, Trip EJ, Siersema PD, van Hoek B, van Erpecum KJ. Protein energy malnutrition predicts complications in liver cirrhosis. Eur J Gastroenterol Hepatol 2011;23:982–989.
[13] Merli M, Lucidi C, Giannelli V, Giusto M, Riggio O, Falcone M, et al. Cirrhotic patients are at risk for health care-associated bacterial infections. Clin Gastroenterol Hepatol 2010;8:979–985.
[14] Merli M, Giusto M, Lucidi C, Giannelli V, Pentassuglio I, Di Gregorio V, et al. Muscle depletion increases the risk of overt and minimal hepatic encephalopathy: results of a prospective study. Metab Brain Dis 2013;28:281–284.
[15] DiCecco SR, Wieners EJ, Wiesner RH, Southorn PA, Plevak DJ, Krom RA. Assessment of nutritional status of patients with end-stage liver disease undergoing liver transplantation. Mayo Clin Proc 1989;64:95–102.
[16] Figueiredo FA, Dickson ER, Pasha TM, Porayko MK, Therneau TM, Malinchoc M, et al. Utility of standard nutritional parameters in detecting body cell mass depletion in patients with end-stage liver disease. Liver Transpl 2000;6:575–581.
[17] Dasarathy S. Consilience in sarcopenia of cirrhosis. J Cachexia Sarcopenia Muscle 2012;3:225–237.
[18] Ney M, Vandermeer B, van Zanten SJ, Ma MM, Gramlich L, Tandon P. Meta-analysis: oral or enteral nutritional supplementation in cirrhosis. Aliment Pharmacol Ther 2013;37:672–679.
[19] Koretz RL, Avenell A, Lipman TO. Nutritional support for liver disease. Cochrane Database Syst Rev 2012;5:CD008344.
[20] Antar R, Wong P, Ghali P. A meta-analysis of nutritional supplementation for management of hospitalized alcoholic hepatitis. Can J Gastroenterol 2012;26:463–467.
[21] Qiu J, Tsien C, Thapalaya S, Narayanan A, Weihl CC, Ching JK, et al. Hyperammonemia-mediated autophagy in skeletal muscle contributes to sarcopenia of cirrhosis. Am J Physiol Endocrinol Metab 2012;303:E983–E993.
[22] Qiu J, Thapaliya S, Runkana A, Yang Y, Tsien C, Mohan ML, et al. Hyperammonemia in cirrhosis induces transcriptional regulation of myostatin by an NF-kappaB-mediated mechanism. Proc Natl Acad Sci U S A 2013;110:18162–18167.
[23] Jones JC, Coombes JS, Macdonald GA. Exercise capacity and muscle strength in patients with cirrhosis. Liver Transpl 2012;18:146–151.
[24] Hong HC, Hwang SY, Choi HY, Yoo HJ, Seo JA, Kim SG, et al. Relationship between sarcopenia and nonalcoholic fatty liver disease: the Korean Sarcopenic Obesity Study. Hepatology 2014;59:1772–1778.
[25] Choudhary NS, Saigal S, Saraf N, Mohanka R, Rastogi A, Goja S, et al. Sarcopenic obesity with metabolic syndrome: a newly recognized entity following living donor liver transplantation. Clin Transplant 2015;29:211–215.
[26] Dasarathy S. Treatment to improve nutrition and functional capacity evaluation in liver transplant candidates. Curr Treat Options Gastroenterol 2014;12:242–255.

Please cite this article in press as: Dasarathy S, Merli M. Sarcopenia from mechanism to diagnosis and treatment in liver disease. J Hepatol (2016), http://dx.doi.org/10.1016/j.jhep.2016.07.040

[27] Montano-Loza AJ. Clinical relevance of sarcopenia in patients with cirrhosis. World J Gastroenterol 2014;20:8061–8071.
[28] Toshikuni N, Arisawa T, Tsutsumi M. Nutrition and exercise in the management of liver cirrhosis. World J Gastroenterol 2014;20:7286–7297.
[29] Kallwitz ER. Sarcopenia and liver transplant: The relevance of too little muscle mass. World J Gastroenterol 2015;21:10982–10993.
[30] Glass C, Hipskind P, Tsien C, Malin SK, Kasumov T, Shah SN, et al. Sarcopenia and a physiologically low respiratory quotient in patients with cirrhosis: a prospective controlled study. J Appl Physiol 1985;2013:559–565.
[31] Tsien C, Shah SN, McCullough AJ, Dasarathy S. Reversal of sarcopenia predicts survival after a transjugular intrahepatic portosystemic stent. Eur J Gastroenterol Hepatol 2013;25:85–93.
[32] Tsien C, Garber A, Narayanan A, Shah SN, Barnes D, Eghtesad B, et al. Post-liver transplantation sarcopenia in cirrhosis: a prospective evaluation. J Gastroenterol Hepatol 2014;29:1250–1257.
[33] Durand F, Buyse S, Francoz C, Laouenan C, Bruno O, Belghiti J, et al. Prognostic value of muscle atrophy in cirrhosis using psoas muscle thickness on computed tomography. J Hepatol 2014;60:1151–1157.
[34] Cruz-Jentoft AJ, Baeyens JP, Bauer JM, Boirie Y, Cederholm T, Landi F, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. Age Ageing 2010;39:412–423.
[35] Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol 1985;2004:2333–2338.
[36] Schiaffino S, Reggiani C. Fiber types in mammalian skeletal muscles. Physiol Rev 2011;91:1447–1531.
[37] Fujiwara N, Nakagawa H, Kudo Y, Tateishi R, Taguri M, Watadani T, et al. Sarcopenia, intramuscular fat deposition, and visceral adiposity independently predict the outcomes of hepatocellular carcinoma. J Hepatol 2015;63:131–140.
[38] Hamaguchi Y, Kaido T, Okumura S, Kobayashi A, Fujimoto Y, Ogawa K, et al. Muscle steatosis is an independent predictor of postoperative complications in patients with hepatocellular carcinoma. World J Surg 2016;40:1959–1968.
[39] Wang CW, Feng S, Covinsky KE, Hayssen H, Zhou LQ, Yeh BM, et al. A comparison of muscle function, mass, and quality in liver transplant candidates: results from the functional assessment in liver transplantation study. Transplantation 2016;100:1692–1698.
[40] Kalafateli M, Mantzoukis K, Choi Yau Y, Mohammad AO, Arora S, Rodrigues S, et al. Malnutrition and sarcopenia predict post-liver transplantation outcomes independently of the Model for End-stage Liver Disease score. J Cachexia Sarcopenia Muscle 2016. http://dx.doi.org/10.1002/jcsm.12095, [Epub ahead of print].
[41] Hanai T, Shiraki M, Ohnishi S, Miyazaki T, Ideta T, Kochi T, et al. Rapid skeletal muscle wasting predicts worse survival in patients with liver cirrhosis. Hepatol Res 2016;46:743–751.
[42] Kim TY, Kim MY, Sohn JH, Kim SM, Ryu JA, Lim S, et al. Sarcopenia as a useful predictor for long-term mortality in cirrhotic patients with ascites. J Korean Med Sci 2014;29:1253–1259.
[43] Masuda T, Shirabe K, Ikegami T, Harimoto N, Yoshizumi T, Soejima Y, et al. Sarcopenia is a prognostic factor in living donor liver transplantation. Liver Transpl 2014;20:401–407.
[44] DiMartini A, Cruz Jr RJ, Dew MA, Myaskovsky L, Goodpaster B, Fox K, et al. Muscle mass predicts outcomes following liver transplantation. Liver Transpl 2013;19:1172–1180.
[45] Englesbe MJ, Patel SP, He K, Lynch RJ, Schaubel DE, Harbaugh C, et al. Sarcopenia and mortality after liver transplantation. J Am Coll Surg 2010;211:271–278.
[46] Hamaguchi Y, Kaido T, Okumura S, Fujimoto Y, Ogawa K, Mori A, et al. Impact of quality as well as quantity of skeletal muscle on outcomes after liver transplantation. Liver Transpl 2014;20:1413–1419.
[47] Hara N, Iwasa M, Sugimoto R, Mifuji-Moroka R, Yoshikawa K, Terasaka E, et al. Sarcopenia and sarcopenic obesity are prognostic factors for overall survival in patients with cirrhosis. Intern Med 2016;55:863–870.
[48] Kaido T, Ogawa K, Fujimoto Y, Ogura Y, Hata K, Ito T, et al. Impact of sarcopenia on survival in patients undergoing living donor liver transplantation. Am J Transplant 2013;13:1549–1556.
[49] Shahid M, Johnson J, Nightingale P, Neuberger J. Nutritional markers in liver allograft recipients. Transplantation 2005;79:359–362.
[50] Lai JC, Feng S, Terrault NA, Lizaola B, Hayssen H, Covinsky K. Frailty predicts waitlist mortality in liver transplant candidates. Am J Transplant 2014;14:1870–1879.
[51] Carey EJ, Steidley DE, Aqel BA, Byrne TJ, Mekeel KL, Rakela J, et al. Six-minute walk distance predicts mortality in liver transplant candidates. Liver Transpl 2010;16:1373–1378.
[52] Alvares-da-Silva MR, Reverbel da Silveira T. Comparison between handgrip strength, subjective global assessment, and prognostic nutritional index in assessing malnutrition and predicting clinical outcome in cirrhotic outpatients. Nutrition 2005;21:113–117.
[53] Montano-Loza AJ, Duarte-Rojo A, Meza-Junco J, Baracos VE, Sawyer MB, Pang JX, et al. Inclusion of sarcopenia within MELD (MELD-Sarcopenia) and the prediction of mortality in patients with cirrhosis. Clin Transl Gastroenterol 2015;6 e102.
[54] van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, Metselaar HJ, IJzermans JN. Systematic review and meta-analysis of the impact of computed tomography assessed skeletal muscle mass on outcome in patients awaiting or undergoing liver transplantation. Am J Transplant 2016;16:2277–2292.
[55] Caregaro L, Alberino F, Amodio P, Merkel C, Bolognesi M, Angeli P, et al. Malnutrition in alcoholic and virus-related cirrhosis. Am J Clin Nutr 1996;63:602–609.
[56] Roubenoff R. Sarcopenia: effects on body composition and function. J Gerontol A Biol Sci Med Sci 2003;58:1012–1017.
[57] Yende S, Iwashyna TJ, Angus DC. Interplay between sepsis and chronic health. Trends Mol Med 2014;20:234–238.
[58] Dolz C, Raurich JM, Ibanez J, Obrador A, Marse P, Gaya J. Ascites increases the resting energy expenditure in liver cirrhosis. Gastroenterology 1991;100:738–744.
[59] Meza-Junco J, Montano-Loza AJ, Baracos VE, Prado CM, Bain VG, Beaumont C, et al. Sarcopenia as a prognostic index of nutritional status in concurrent cirrhosis and hepatocellular carcinoma. J Clin Gastroenterol 2013;47:861–870.
[60] Merli M, Giusto M, Giannelli V, Lucidi C, Riggio O. Nutritional status and liver transplantation. J Clin Exp Hepatol 2011;1:190–198.
[61] Daniel PM. The metabolic homoeostatic role of muscle and its function as a store of protein. Lancet 1977;2:446–448.
[62] Tessari P, Kiwanuka E, Vettore M, Barazzoni R, Zanetti M, Cecchet D, et al. Phenylalanine and tyrosine kinetics in compensated liver cirrhosis: effects of meal ingestion. Am J Physiol Gastrointest Liver Physiol 2008;295: G598–G604.
[63] Tessari P, Vettore M, Millioni R, Puricelli L, Orlando R. Effect of liver cirrhosis on phenylalanine and tyrosine metabolism. Curr Opin Clin Nutr Metab Care 2010;13:81–86.
[64] McCullough AJ, Mullen KD, Tavill AS, Kalhan SC. In vivo differences between the turnover rates of leucine and leucine's ketoacid in stable cirrhosis. Gastroenterology 1992;103:571–578.
[65] Thapaliya S, Runkana A, McMullen MR, Nagy LE, McDonald C, Naga Prasad SV, et al. Alcohol-induced autophagy contributes to loss in skeletal muscle mass. Autophagy 2014;10:677–690.
[66] Tsien C, Davuluri G, Singh D, Allawy A, Ten Have GA, Thapaliya S, et al. Metabolic and molecular responses to leucine-enriched branched chain amino acid supplementation in the skeletal muscle of alcoholic cirrhosis. Hepatology 2015;61:2018–2029.
[67] Morrison WL, Bouchier IA, Gibson JN, Rennie MJ. Skeletal muscle and whole-body protein turnover in cirrhosis. Clin Sci (Lond) 1990;78:613–619.
[68] Tsien CD, McCullough AJ, Dasarathy S. Late evening snack: exploiting a period of anabolic opportunity in cirrhosis. J Gastroenterol Hepatol 2012;27:430–441.
[69] Merli M, Eriksson LS, Hagenfeldt L, Wahren J. Splanchnic and leg exchange of free fatty acids in patients with liver cirrhosis. J Hepatol 1986;3:348–355.
[70] Chen X, Iqbal N, Boden G. The effects of free fatty acids on gluconeogenesis and glycogenolysis in normal subjects. J Clin Invest 1999;103:365–372.
[71] Blonde-Cynober F, Aussel C, Cynober L. Abnormalities in branched-chain amino acid metabolism in cirrhosis: influence of hormonal and nutritional factors and directions for future research. Clin Nutr 1999;18:5–13.
[72] Bembr A. The amino acid composition of animal tissue protein. J Biol Chem 1943;148:431–439.
[73] Iob V, Coon WW, Sloan M. Free amino acids in liver, plasma, and muscle of patients with cirrhosis of the liver. J Surg Res 1967;7:41–43.
[74] Montanari A, Simoni I, Vallisa D, Trifiro A, Colla R, Abbiati R, et al. Free amino acids in plasma and skeletal muscle of patients with liver cirrhosis. Hepatology 1988;8:1034–1039.
[75] Plauth M, Egberts EH, Abele R, Muller PH, Furst P. Characteristic pattern of free amino acids in plasma and skeletal muscle in stable hepatic cirrhosis. Hepatogastroenterology 1990;37:135–139.
[76] Mowat NA, Edwards CR, Fisher R, McNeilly AS, Green JR, Dawson AM. Hypothalamic-pituitary-gonadal function in men with cirrhosis of the liver. Gut 1976;17:345–350.
[77] Kovarik M, Muthny T, Sispera L, Holecek M. The dose-dependent effects of endotoxin on protein metabolism in two types of rat skeletal muscle. J Physiol Biochem 2012;68:385–395.

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Review

[78] McDaniel J, Davuluri G, Hill EA, Moyer M, Runkana A, Prayson R, et al. Hyperammonemia results in reduced muscle function independent of muscle mass. Am J Physiol Gastrointest Liver Physiol 2016;310:G163–G170.

[79] Shangraw RE, Jahoor F. Effect of liver disease and transplantation on urea synthesis in humans: relationship to acid-base status. Am J Physiol 1999;276:G1145–G1152.

[80] Olde Damink SW, Jalan R, Dejong CH. Interorgan ammonia trafficking in liver disease. Metab Brain Dis 2009;24:169–181.

[81] Hadjihambi A, Rose CF, Jalan R. Novel insights into ammonia-mediated neurotoxicity pointing to potential new therapeutic strategies. Hepatology 2014;60:1101–1103.

[82] Dam G, Ott P, Aagaard NK, Vilstrup H. Branched-chain amino acids and muscle ammonia detoxification in cirrhosis. Metab Brain Dis 2013;28:217–220.

[83] Holecek M. Evidence of a vicious cycle in glutamine synthesis and breakdown in pathogenesis of hepatic encephalopathy-therapeutic perspectives. Metab Brain Dis 2014;29:9–17.

[84] Lockwood AH, McDonald JM, Reiman RE, Gelbard AS, Laughlin JS, Duffy TE, et al. The dynamics of ammonia metabolism in man. Effects of liver disease and hyperammonemia. J Clin Invest 1979;63:449–460.

[85] Ganda OP, Ruderman NB. Muscle nitrogen metabolism in chronic hepatic insufficiency. Metabolism 1976;25:427–435.

[86] Dasarathy S, Muc S, Hisamuddin K, Edmison JM, Dodig M, McCullough AJ, et al. Altered expression of genes regulating skeletal muscle mass in the portacaval anastomosis rat. Am J Physiol Gastrointest Liver Physiol 2007;292:G1105–G1113.

[87] Dasarathy S, McCullough AJ, Muc S, Schneyer A, Bennett CD, Dodig M, et al. Sarcopenia associated with portosystemic shunting is reversed by follistatin. J Hepatol 2011;54:915–921.

[88] Takeda K, Takemasa T. Expression of ammonia transporters Rhbg and Rhcg in mouse skeletal muscle and the effect of 6-week training on these proteins. Physiol Rep 2015;3, [pii: e12596, doi: 12510.14814/phy12592.12596].

[89] Garcia PS, Cabbabe A, Kambadur R, Nicholas G, Csete M. Brief-reports: elevated myostatin levels in patients with liver disease: a potential contributor to skeletal muscle wasting. Anesth Analg 2010;111:707–709.

[90] Merli M, Giusto M, Molfino A, Bonetto A, Rossi M, Ginanni Corradini S, et al. MuRF-1 and p-GSK3beta expression in muscle atrophy of cirrhosis. Liver Int 2013;33:714–721.

[91] Han HQ, Zhou X, Mitch WE, Goldberg AL. Myostatin/activin pathway antagonism: molecular basis and therapeutic potential. Int J Biochem Cell Biol 2013;45:2333–2347.

[92] Kosenko E, Venediktova N, Kaminsky Y, Montoliu C, Felipo V. Sources of oxygen radicals in brain in acute ammonia intoxication in vivo. Brain Res 2003;981:193–200.

[93] Davuluri G, Krokowski D, Guan BJ, Kumar A, Thapaliya S, Singh D, et al. Metabolic adaptation of skeletal muscle to hyperammonemia drives the beneficial effects of L-leucine in cirrhosis. J Hepatol 2016. http://dx.doi.org/10.1016/j.jhep.2016.06.004, pii: S0168-8278(16)30266-5. [Epub ahead of print].

[94] Owen OE, Kalhan SC, Hanson RW. The key role of anaplerosis and cataplerosis for citric acid cycle function. J Biol Chem 2002;277:30409–30412.

[95] Ertan H. Some properties of glutamate dehydrogenase, glutamine synthetase and glutamate synthase from Corynebacterium callunae. Arch Microbiol 1992;158:35–41.

[96] Lai JC, Cooper AJ. Neurotoxicity of ammonia and fatty acids: differential inhibition of mitochondrial dehydrogenases by ammonia and fatty acyl coenzyme A derivatives. Neurochem Res 1991;16:795–803.

[97] Breuillard C, Cynober L, Moinard C. Citrulline and nitrogen homeostasis: an overview. Amino Acids 2015;47:685–691.

[98] Dasarathy S, Mullen KD, Dodig M, Donofrio B, McCullough AJ. Inhibition of aromatase improves nutritional status following portacaval anastomosis in male rats. J Hepatol 2006;45:214–220.

[99] Bucuvalas JC, Cutfield W, Horn J, Sperling MA, Heubi JE, Campaigne B, et al. Resistance to the growth-promoting and metabolic effects of growth hormone in children with chronic liver disease. J Pediatr 1990;117:397–402.

[100] Moller S, Becker U, Gronbaek M, Juul A, Winkler K, Skakkebaek NE. Short-term effect of recombinant human growth hormone in patients with alcoholic cirrhosis. J Hepatol 1994;21:710–717.

[101] Liu W, Thomas SG, Asa SL, Gonzalez-Cadavid N, Bhasin S, Ezzat S. Myostatin is a skeletal muscle target of growth hormone anabolic action. J Clin Endocrinol Metab 2003;88:5490–5496.

[102] Lakshman KM, Bhasin S, Corcoran C, Collins-Racie LA, Tchistiakova L, Forlow SB, et al. Measurement of myostatin concentrations in human serum: Circulating concentrations in young and older men and effects of testosterone administration. Mol Cell Endocrinol 2009;302:26–32.

[103] Sinclair M, Grossmann M, Hoermann R, Angus PW, Gow PJ. Testosterone therapy increases muscle mass in men with cirrhosis and low testosterone: A randomised controlled trial. J Hepatol 2016. http://dx.doi.org/10.1016/j.jhep.2016.06.007. pii: S0168-8278(16)30269-0. [Epub ahead of print].

[104] Bajaj JS, Heuman DM, Hylemon PB, Sanyal AJ, White MB, Monteith P, et al. Altered profile of human gut microbiome is associated with cirrhosis and its complications. J Hepatol 2014;60:940–947.

[105] Lang CH, Frost RA, Nairn AC, MacLean DA, Vary TC. TNF-alpha impairs heart and skeletal muscle protein synthesis by altering translation initiation. Am J Physiol Endocrinol Metab 2002;282:E336–E347.

[106] Keller CW, Fokken C, Turville SG, Lunemann A, Schmidt J, Munz C, et al. TNF-alpha induces macroautophagy and regulates MHC class II expression in human skeletal muscle cells. J Biol Chem 2011;286:3970–3980.

[107] Carias S, Castellanos AL, Vilchez V, Nair R, Dela Cruz AC, Watkins J, et al. Nonalcoholic steatohepatitis is strongly associated with sarcopenic obesity in patients with cirrhosis undergoing liver transplant evaluation. J Gastroenterol Hepatol 2016;31:628–633.

[108] Merli M, Iebba V, Giusto M. What is new about diet in hepatic encephalopathy. Metab Brain Dis 2015, [Epub ahead of print].

[109] Dasarathy S. Cause and management of muscle wasting in chronic liver disease. Curr Opin Gastroenterol 2016;32:159–165.

[110] Sinclair M, Gow PJ, Grossmann M, Angus PW. Review article: sarcopenia in cirrhosis–aetiology, implications and potential therapeutic interventions. Aliment Pharmacol Ther 2016;43:765–777.

[111] Williams TJ, McKenna MJ. Exercise limitation following transplantation. Compr Physiol 2012;2:1937–1979.

[112] Plauth M, Merli M, Kondrup J, Weimann A, Ferenci P, Muller MJ, et al. ESPEN guidelines for nutrition in liver disease and transplantation. Clin Nutr 1997;16:43–55.

[113] Plauth M, Cabre E, Riggio O, Assis-Camilo M, Pirlich M, Kondrup J, et al. ESPEN guidelines on enteral nutrition: liver disease. Clin Nutr 2006;25:285–294.

[114] Plank LD, Mathur S, Gane EJ, Peng SL, Gillanders LK, McIlroy K, et al. Perioperative immunonutrition in patients undergoing liver transplantation: a randomized double-blind trial. Hepatology 2015;61:639–647.

[115] Amodio P, Bemeur C, Butterworth R, Cordoba J, Kato A, Montagnese S, et al. The nutritional management of hepatic encephalopathy in patients with cirrhosis: International Society for Hepatic Encephalopathy and Nitrogen Metabolism Consensus. Hepatology 2013;58:325–336.

[116] Hirsch S, Bunout D, de la Maza P, Iturriaga H, Petermann M, Icazar G, et al. Controlled trial on nutrition supplementation in outpatients with symptomatic alcoholic cirrhosis. JPEN J Parenter Enteral Nutr 1993;17:119–124.

[117] de Ledinghen V, Beau P, Mannant PR, Borderie C, Ripault MP, Silvain C, et al. Early feeding or enteral nutrition in patients with cirrhosis after bleeding from esophageal varices? A randomized controlled study. Dig Dis Sci 1997;42:536–541.

[118] Le Cornu KA, McKiernan FJ, Kapadia SA, Neuberger JM. A prospective randomized study of preoperative nutritional supplementation in patients awaiting elective orthotopic liver transplantation. Transplantation 2000;69:1364–1369.

[119] Marchesini G, Bianchi G, Merli M, Amodio P, Panella C, Loguercio C, et al. Nutritional supplementation with branched-chain amino acids in advanced cirrhosis: a double-blind, randomized trial. Gastroenterology 2003;124:1792–1801.

[120] Hu QG, Zheng QC. The influence of Enteral Nutrition in postoperative patients with poor liver function. World J Gastroenterol 2003;9:843–846.

[121] Les I, Doval E, Garcia-Martinez R, Planas M, Cardenas G, Gomez P, et al. Effects of branched-chain amino acids supplementation in patients with cirrhosis and a previous episode of hepatic encephalopathy: a randomized study. Am J Gastroenterol 2011;106:1081–1088.

[122] Dupont B, Dao T, Joubert C, Dupont-Lucas C, Gloro R, Nguyen-Khac E, et al. Randomised clinical trial: enteral nutrition does not improve the long-term outcome of alcoholic cirrhotic patients with jaundice. Aliment Pharmacol Ther 2012;35:1166–1174.

[123] Sorrentino P, Castaldo G, Tarantino L, Bracigliano A, Perrella A, Perrella O, et al. Preservation of nutritional-status in patients with refractory ascites due to hepatic cirrhosis who are undergoing repeated paracentesis. J Gastroenterol Hepatol 2012;27:813–822.

[124] Matsuoka S, Tamura A, Nakagawara H, Moriyama M. Improvement in the nutritional status and clinical conditions of patients with liver failure using a liver diet combined with a branched chain amino acids-enriched elemental diet. Hepatogastroenterology 2014;61:1308–1312.

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[125] Fialla AD, Israelsen M, Hamberg O, Krag A, Gluud LL. Nutritional therapy in cirrhosis or alcoholic hepatitis: a systematic review and meta-analysis. Liver Int 2015;35:2072–2078.
[126] Okuda H, Shiratori K. Long-term nutritional assessment and quality of life in patients with cirrhosis taking a late evening snack. J Gastroenterol 2007;42:186–187.
[127] Nguyen DL, Morgan T. Protein restriction in hepatic encephalopathy is appropriate for selected patients: a point of view. Hepatol Int 2014;8:447–451.
[128] Metcalfe EL, Avenell A, Fraser A. Branched-chain amino acid supplementation in adults with cirrhosis and porto-systemic encephalopathy: systematic review. Clin Nutr 2014;33:958–965.
[129] Alexander WF, Spindel E, Harty RF, Cerda JJ. The usefulness of branched chain amino acids in patients with acute or chronic hepatic encephalopathy. Am J Gastroenterol 1989;84:91–96.
[130] Gluud LL, Dam G, Borre M, Les I, Cordoba J, Marchesini G, et al. Oral branched-chain amino acids have a beneficial effect on manifestations of hepatic encephalopathy in a systematic review with meta-analyses of randomized controlled trials. J Nutr 2013;143:1263–1268.
[131] Gluud LL, Dam G, Les I, Cordoba J, Marchesini G, Borre M, et al. Branched-chain amino acids for people with hepatic encephalopathy. Cochrane Database Syst Rev 2015;9:CD001939.
[132] Zhang P, McGrath BC, Reinert J, Olsen DS, Lei L, Gill S, et al. The GCN2 eIF2alpha kinase is required for adaptation to amino acid deprivation in mice. Mol Cell Biol 2002;22:6681–6688.
[133] Carroll B, Korolchuk VI, Sarkar S. Amino acids and autophagy: cross-talk and co-operation to control cellular homeostasis. Amino Acids 2015;47:2065–2088.
[134] Fyfe JJ, Bishop DJ, Stepto NK. Interference between concurrent resistance and endurance exercise: molecular bases and the role of individual training variables. Sports Med 2014;44:743–762.
[135] Damas F, Phillips S, Vechin FC, Ugrinowitsch C. A review of resistance training-induced changes in skeletal muscle protein synthesis and their contribution to hypertrophy. Sports Med 2015;45:801–807.
[136] Hornberger TA, Chu WK, Mak YW, Hsiung JW, Huang SA, Chien S. The role of phospholipase D and phosphatidic acid in the mechanical activation of mTOR signaling in skeletal muscle. Proc Natl Acad Sci U S A 2006;103:4741–4746.
[137] Orr R, Fiatarone Singh M. The anabolic androgenic steroid oxandrolone in the treatment of wasting and catabolic disorders: review of efficacy and safety. Drugs 2004;64:725–750.
[138] Rambaldi A, Iaquinto G, Gluud C. Anabolic-androgenic steroids for alcoholic liver disease: a Cochrane review. Am J Gastroenterol 2002;97:1674–1681.
[139] Rose CF. Ammonia-lowering strategies for the treatment of hepatic encephalopathy. Clin Pharmacol Ther 2012;92:321–331.
[140] Lockwood AH. Blood ammonia levels and hepatic encephalopathy. Metab Brain Dis 2004;19:345–349.
[141] Semsarian C, Wu MJ, Ju YK, Marciniec T, Yeoh T, Allen DG, et al. Skeletal muscle hypertrophy is mediated by a Ca2+-dependent calcineurin signalling pathway. Nature 1999;400:576–581.
[142] Selberg O, Selberg D. Norms and correlates of bioimpedance phase angle in healthy human subjects, hospitalized patients, and patients with liver cirrhosis. Eur J Appl Physiol 2002;86:509–516.

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