Regulation of Skeletal Muscle Function by Amino Acids, Especially Non-Proteinogenic Amino Acids

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Summary
Amino acids are compounds that contain an amino group (-NH₂) and a carboxyl group (-COOH) and are components of proteins and materials for various bioactive molecules. The skeletal muscle, which is the largest organ in the human body, representing ~40% of the total body weight, plays important roles in exercise, energy expenditure, and glucose/amino acid usage-processes that are modulated by various amino acids and their metabolites. In this review, we address the metabolism and function of amino acids, especially non-proteinogenic amino acids, in the skeletal muscle. Leucine, a BCAA, and its metabolite, β-hydroxy-β-methylbutyrate (HMB), both activate mammalian target of rapamycin complex 1 (mTORC1) and increase protein synthesis, but the mechanisms of activation appear to be different. The metabolite of valine (another BCAA), β-aminoisobutyric acid (BAIBA), is increased by exercise, is secreted by the skeletal muscle, and acts on other tissues, such as white adipose tissue, to increase energy expenditure. In addition, several amino acid-related molecules reportedly activate skeletal muscle function. Oral 5-aminolevulinic acid (ALA) supplementation can protect against mild hyperglycemia and help prevent type 2 diabetes. β-alanine levels are decreased in the skeletal muscles of aged mice. β-alanine supplementation increased the physical performance and improved the executive function induced by endurance exercise in middle-aged individuals. Further studies focusing on the effects of amino acids and their metabolites on skeletal muscle function will provide data essential for the production of food supplements for older adults, athletes, and individuals with metabolic diseases.

Key Words skeletal muscle, PGC1α, exercise, energy expenditure, branched-chain amino acid (BCAA), leucine, β-hydroxy-β-methylbutyrate (HMB), β-aminoisobutyric acid (BAIBA)

Introduction: Amino acids and health
Amino acids are compounds that contain an amino group (-NH₂) and a carboxyl group (-COOH) and are components of proteins and materials for various bioactive molecules. As amino acids are known to be biologically safe, they are used for food and non-food purposes. Amino acids, and other food components, can be divided according to their biological regulatory functions; for example, leucine increases the anabolism of muscle proteins (1), arginine has a vasodilation action and enhances immunity (2), and gamma aminobutyric acid (GABA) regulates blood pressure (3). In addition to these three functions, amino acids can also be used as medical diagnostic tools and to predict the risk of various diseases, such as cancer, by measuring their levels in blood (4). Thus, the effects of amino acids on health are a highly important issue. In this review, we discuss the roles of amino acids, especially non-proteinogenic amino acids, in the skeletal muscle.

Skeletal muscle and prevention of metabolic diseases
The skeletal muscle is the largest organ in the human body, representing ~40% of the total body weight, which stores energy in the form of proteins (amino acids). The skeletal muscle exhibits plasticity in response to the environment; proper exercise combined with adequate nutrition leads to muscle hypertrophy. Conversely, motor incapacity and aging cause muscle atrophy, which leads to decreased energy expenditure (obesity), decreased blood glucose uptake by the skeletal muscle and increased blood glucose levels (diabetes), and a lower quality of life (5). In aged societies, such as those in developed countries, muscle atrophy suppression is important for health and longevity. Moreover, exercise has been shown to affect not only the skeletal muscle but also various other organs (6, 7). An understanding of the metabolism of the skeletal muscle during exercise and of the mechanisms of muscle atrophy is important for the prevention and treatment of metabolic diseases and muscle dysfunction.

β-hydroxy-β-methylbutyrate (HMB) in comparison to Leucine
β-hydroxy-β-methylbutyrate (HMB) is a metabolite of leucine. Generally, leucine is metabolized to α-ketoisocaproic acid (KIC) by branched-chain amino acid aminotransferase 2 (BCAT2) in the skeletal muscle. Most of the KIC is converted to isovaleryl-CoA, and only 5–10% of KIC is converted to HMB in the liver (8). HMB
has been used as an ergogenic supplement to increase muscle mass and strength in humans. Several studies have revealed that HMB and leucine stimulate protein synthesis and reduce muscle protein breakdown. HMB and leucine increase the phosphorylation of FOXO1 and decrease nuclear FOXO1 levels, resulting in the downregulation of muscle atrophy-related muscle RING-finger protein-1 (MURF1) (9). Suppression of the FOXO1 pathway by HMB or leucine may prevent muscle atrophy, as FOXO1 is an important transcription factor in this process (10). Similarly to leucine, HMB increases protein synthesis through the activation of mTORC1. Treating myoblasts with HMB increases Akt phosphorylation and, subsequently, activates mTORC1 signaling. To our knowledge, no report has described the activation of mTORC1 by HMB by Sestrin 2 or Sestrin 1. Thus, the signals produced by HMB to activate mTORC1 may be distinct from those of leucine.

The detailed mechanisms underlying the actions of leucine and HMB (i.e., differences and similarities) in the skeletal muscle warrant further clarification.

**Valine metabolites**

BAIBA stems from mitochondrial valine catabolism and is produced by the skeletal muscle during exercise (11). Moreover, it has been shown to communicate the beneficial effects of exercise from the skeletal muscle to other tissues and organs in an endocrine manner. BAIBA increases energy expenditure by activating the β-oxidation pathway of hepatic fatty acids, triggers the browning of white adipose tissue, is inversely correlated with cardiometabolic risk factors (11), and improves insulin resistance and inflammation in the skeletal muscle (12). We previously reported that BAIBA reduces tumor necrosis factor (TNF)-α-induced expression of vascular cell adhesion molecule (VCAM)-1 in human umbilical endothelial cells, suggesting that BAIBA acts to prevent atherosclerosis by physical training (13, 14).

There are two enantiomers of BAIBA in biological systems: L-BAIBA and D-BAIBA. L-BAIBA is generated from catabolic reactions of L-valine, whereas D-BAIBA is produced from thymine in the cytosol. Kitase et al. revealed that the production of L-BAIBA increases during muscle contraction, presumably due to intensive oxidation of L-valine (15). L-BAIBA, secreted by skeletal muscle, has been shown to act on osteocytes; it diminishes the production of reactive oxygen species in mitochondria and protects osteocytes from apoptosis, preventing bone loss (15). However, it is unknown whether systemic D-BAIBA levels are also affected by exercise or whether this regulation is specific to L-BAIBA.

Vinaline catabolic intermediate 3-hydroxy-isobutylate (3-HIB) stimulates fatty acid intake in the skeletal muscle and leads to insulin resistance, which may be due to the accumulation of intramuscular lipids (12, 16). Recently, Yoneshiro et al. reported that insufficient BCAA catabolism in brown adipose tissue led to obesity and diabetic phenotypes in mice (17). BCAA catabolism in the skeletal muscle may also be related to obesity and diabetes. Sufficient metabolism of valine and BCAA may lead to decreased 3-HIB levels, which appears to be important for the prevention of obesity and diabetes.

**Other amino acid metabolites**

Metabolomic analysis of PGCo-α-transgenic mice revealed a marked increase in GABA, BAIBA, and amino acid metabolites (18). GABA intake improves high blood pressure (3), as does regular exercise. Exercise-induced PGCo-α expression increases GABA production and may contribute to the improvement of high blood pressure (18). BAIBA and GABA are potential myokines that are secreted from exercised skeletal muscle and affect various other organs, and they may explain the exercise-mediated improvement of metabolic diseases via the interactions between muscles and other organs.

NO production improves blood flow. Increased endothelial nitric oxide synthase (eNOS) expression and muscle blood vessel formation were observed in PGCo-α-transgenic mice (19). This appears to be a situation in which the transfer of nutrients and oxygen during exercise is enhanced. Moreover, arginine supplementation in mice increases PGCo-α expression (20), and arginine may activate PGCo-α-mediated NO production. NO is produced from arginine; however, arginine taken orally is susceptible to degradation in the intestine, whereas the intake of citrulline, a precursor of arginine, increases blood arginine levels more effectively (21). Thus, citrulline intake may improve blood flow through the skeletal muscle.

The amino acid 5-aminolevulinic acid (ALA) is produced by ALA synthase (ALAS) and is important for the heme-biosynthesis process. PGCo-α activates the expression of the ALAS gene in the liver (22). We observed decreased ALAS expression in skeletal muscle-specific PGCo-α-deficient mice, as assessed by microarray analysis (23). ALA can control glucose metabolism in the skeletal muscle. Decreased ALA levels in mice and muscle cells attenuate mitochondrial function and cause impaired glucose tolerance and insulin resistance, which are recovered by ALA treatment (24). In fact, cohort studies suggest that oral ALA administration can protect against mild hyperglycemia and may help prevent type 2 diabetes (24). Thus, ALA may be a useful supplement to improve skeletal muscle function.

**Closing remarks**

Amino acids are critical for human health. In this review, we discussed the roles of amino acids in the skeletal muscle and the organs that interact with it. We described the relationships between the exercise-activated transcription regulator PGCo-α and amino acids. In addition, we discussed the changes in amino acid metabolites during skeletal muscle aging. Clearly, BCAA and various other amino acids and their metabolites play important roles in the skeletal muscle. Further study of amino acids, especially in the skeletal muscle, will continue to benefit preventive medicine and health sciences.
Disclosure of state of COI
No conflicts of interest to be declared.

REFERENCES