Sarcopenia and aspartic acid magnesium

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Abstract  Sarcopenia from aging, inactivity, weightlessness, disease and malnutrition causes social problems due to associated physical dysfunction, a decrease in QOL, and risk of death. As a means of alleviating sarcopenia, appropriate exercise and nutrition therapy is required. However, nourishment therapy is especially important for a person in a situation where exercise is not feasible for maintaining skeletal muscle mass and muscular strength. Therefore, the authors studied the inhibitory effect of aspartic acid magnesium on disuse muscle atrophy. Herein we have described two of the mechanisms of action of aspartic acid magnesium on disuse muscle atrophy suppression. Initially, inactivity causes a decrease in capillary and oxygen supply, inhibits ATP synthesis, and produces active oxygen. As a result, the proteolysis system is activated and muscle atrophy progresses. We have described the effect of aspartic acid magnesium uptake on this mechanism, and the imbalance of the intracellular mineral with the inactive proteolysis system. The above is the content of this review.

Keywords : sarcopenia, aspartic acid magnesium, antimycin A, ATP, mineral imbalance

Introduction

Skeletal muscle atrophy is caused by aging, inactivity, microgravity, disease and malnutrition. Muscle atrophy derived from these causes is collectively called sarcopenia. Sarcopenia is a concept that was proposed by Rosenberg [1] in 1987. In 2010, the European Working Group on Sarcopenia in Older People defined sarcopenia as a syndrome characterized by the progressive and generalized loss of skeletal muscle mass and strength with a risk of adverse outcomes such as physical disability, poor quality of life and death [2]. Disuse muscle atrophy (DMA) is positioned secondarily to sarcopenia. DMA is atrophy of the skeletal muscle that results from inactivity and weightlessness exposure. Muscle weakness in the elderly stemming from a decrease in the amount of activity in daily life in Japan has been a problem. Appropriate exercise and nourishment prescriptions are needed for lessening the incidence of sarcopenia. It is generally known that appropriate prescription-based exercise is needed to improve disuse muscular atrophy. However, in order to maintain and improve the muscle mass and strength of a person who cannot exercise due to certain circumstances, an appropriate nutritional formula and therapy becomes even more essential.

In recent years, various supplements have been developed for the purpose of increasing muscle mass and strength. Therefore, in this review, we would like to introduce the effect of one such supplement, aspartic acid magnesium, on disuse muscle atrophy [3-6].

Disuse muscle atrophy and protein degradation system

Skeletal muscle protein degradation increases in the cases of plaster cast fixation and being bedridden - when skeletal muscle is unable to exert tension and results in a drop in muscular strength at the same time.

Previously, as part of the intracellular proteolytic system during disuse muscular atrophy, the autophagy-lysosomal system, calpain system and the ubiquitin-proteasome system, and research related to each system at the time of muscle atrophy, were reported [7-8]. Myofibrils are very dense with a huge structure composed of a number of proteins such as actin and myosin. Therefore, as for proteolysis of muscular fiber, calpain first cuts the myofibrillar Z band and then isolated in myosin and an actin filaments. Following this, the myosin and actin filaments are broken down into a small peptides by proteasome. The peptide is, furthermore, broken down into an amino acid by a lysosomal system. This is how proteolysis system of muscular fiber resolution works.

However, this series of proteolysis processes is not without a problem. In other words, muscle protein degradation increases even with an inhibitor of the lysosomal system and calpain system. As a result, muscle atrophy is induced.

Skeletal muscle protein degradation system trigger

In recent years, the factors that cause the activation of these proteolytic systems have been noted. In previous reports, factors such as a mineral imbalance and decrease in ATP have been considered as triggers for protein deg-
The degradation of muscle proteins begins with a breakdown of intracellular mineral homeostasis. Excretion of a mineral is not necessarily excreted according to excess and/or a deficiency in the body. Physiologically, other minerals will be influenced if a certain mineral is excreted. That is, it is supposed that there is a correlation between minerals. For example, calcium (Ca) adversely flows in a cell when intracellular magnesium (Mg) is excreted for exercise or stress. An imbalance of such a mineral occurs in a muscle cell. As a result, reactive oxygen species (ROS) are produced by the imbalance of the mineral as shown in Fig 1. It is thought that the production of ROS induces muscle atrophy (Fig. 1).

On the other hand, a decrease in the number of capillaries is known to induce disuse muscle atrophy. As a result, blood flow volume falls, and the muscle cell becomes hypoxic. Furthermore, oxidative phosphorylation is delayed, and a decrease in ATP production delays the pump function of the cell. As a result, the Ca in the cell increases, and disuse muscle atrophy is induced.

Recently, the authors revealed that magnesium aspartate acts effectively in suppressing the induction of such disuse muscular atrophy.

**Effect of aspartic acid magnesium on skeletal muscle atrophy induced by a decrease in ATP**

During inactivity, the number of capillaries is reduced, resulting in a reduction in the oxygen supply as well. Accordingly, ATP synthesis is reduced and muscular atrophy is caused by the inhibition of ATP synthesis\(^{11,12}\). Tissue becomes ischemic due to a decrease in the number of capillaries, which, in turn, stops oxidative phosphorylation. Following this, adenosine triphosphate (ATP) synthesis stagnates, the activity of Na-K dependent ATPase is impaired, and sodium flows into the cell. Excessive intracellular sodium is replaced with calcium by a sodium-calcium exchanger increasing intracellular calcium concentration.

Intracellular calcium activates calpain and enhances protein degradation, resulting in disuse muscle atrophy\(^{13,14}\). Aspartic acid generates fumaric acid by a purine nucleotide circuit as shown in Fig. 1. Aspartic acid combines with inosine monophosphate (IMP) by the catalyst of an adenylosuccinic acid synthetic enzyme, forming adenylosuccinic acid. Adenylosuccinic acid is broken down by the catalyst of an adenylosuccinic acid lyase, generating adenosine monophosphate (AMP) and a fumaric acid. AMP is converted to ATP by AMPK. With aspartic acid magnesium uptake, ATP production in the circuit is maintained, and is suggested to suppress muscle atrophy.

The authors suspected that a reduction in adenosine triphosphate (ATP) synthesis induced atrophy of skeletal muscle. Magnesium aspartate in the purine nucleotide circuit prompted ATP synthesis, and was thought to suppress the atrophy of skeletal muscle. Using a mouse atrophy induction model, the authors observed the suppression of muscle atrophy due to aspartic acid magnesium and ATP. Many articles reveal that ATP decreases with atrophy of the skeletal muscle. Also, it was examined whether skeletal muscle atrophy was induced when ATP synthesis was inhibited. The authors used antimycin as an ATP synthesis inhibitor.

The electron transfer system, which exists in a mitochondria internal membrane, is the system to synthesize ATP. The system is comprised of four enzyme complexes called complex I, II, III, and IV. These complexes deliver an electron and transport a hydrogen ion from inside the membrane to the outside. ATP synthase makes ATP synthesis using a concentration gradient of protons arising as a result\(^{15}\). Antimycin A inhibits complex III of the electronic transmission system. Complex III is comprised of a cytochrome b566, cytochrome b560, cytochrome c1, Fe-S cluster. Ubiquinol is oxidized in Qi by ubiquinone, and an electron moves to cytochrome b566. The electron moves to cytochrome b560. Ubiquinone is returned to the Qi part of cytochrome b560 and becomes ubiquinol. This series of electron transfers is called a quinone cycle. Antimycin A block the electron transfer process to the Qi site from cytochrome b560 of the quinone cycle. Thus antimycin A inhibits ATP synthesis by inhibiting the electron transfer system\(^{15-17}\).

![Aspartic acid and purine nucleotides circuit](Image)

**Fig. 1** Aspartic acid and purine nucleotides circuit
Effect of L/D-Asp intake on muscle wet weight per body weight in tail suspension

Nagata et al. reported that D-aspartic acid increases the mRNA of the steroidal hormone-producing regulatory protein\(^{18,19}\). Moreover, administration of D-aspartic acid has been reported to increase testosterone synthesis in the testis. Also, L-aspartic acid has been reported that may be converted to D-aspartic acid in vivo\(^{18,19}\). D-aspartic acid is generated from L-aspartic acid that is administered orally. The converted D-aspartic acid induces testosterone synthesis. As a result, protein assimilation of skeletal muscle is enhanced, and it is believed that the atrophy of muscle is suppressed.

The authors examined the effect of L-aspartic acid and D-aspartic acid administration on disuse muscle atrophy. As shown in Fig. 2, the mean and standard deviation of W group, L-asp group and D-asp group are 0.25 ± 0.03 mg/g, 0.25 ± 0.01 mg/g, and 0.25 ± 0.02 mg/g in the non-TS group. Mean and standard deviation of W group, L-asp group and D-asp group are 0.19 ± 0.02 mg/g, 0.23 ± 0.02 mg/g, and 0.22 ± 0.03 mg/g in the TS group. After making multiple comparisons to examine the influence that difference of drinking water gave to soleus muscle wet weight of the TS group and non-TS group, difference of drinking water was not seen in the non-TS group (Fig. 2A).

As shown in Fig. 2B, the average of muscle weight of soleus of the L-aspartic acid TS group is significantly higher in comparison to average of muscle weight of soleus of the WTS group. As a result of having performed a two way ANOVA of plantaris muscle wet weight between the drinking water groups, a statistically significant difference was not observed. Furthermore, as a result of having performed a two way ANOVA of variance of plantaris muscle wet weight between the TS groups, a statistically significant difference was not observed.

Effect of L/D-Asp uptake on soleus muscle protein content in tail suspension

As shown in Fig. 3, Soleus muscle protein content is 0.02 mg/g, and 0.22 ± 0.03 mg/g in the TS group. After making multiple comparisons to examine the influence that difference of drinking water gave to soleus muscle wet weight of the TS group and non-TS group, difference of drinking water was not seen in the non-TS group (Fig. 2A).

As shown in Fig. 2B, the average of muscle weight of soleus of the L-aspartic acid TS group is significantly higher in comparison to average of muscle weight of soleus of the WTS group. As a result of having performed a two way ANOVA of plantaris muscle wet weight between the drinking water groups, a statistically significant difference was not observed. Furthermore, as a result of having performed a two way ANOVA of variance of plantaris muscle wet weight between the TS groups, a statistically significant difference was not observed.

**Fig. 2** Effect of the L/D-aspartic acid (L/D-asp) intake to give to soleus muscle wet weight per body weight in non-TS group (A) and TS group (B)

W: Water
L-asp: L- 72mg L-aspartic acid/1l water
D-asp: D- 72mg D-aspartic acid/1l water

**Fig. 3** Effect of L / D- aspartic acid on the soleus muscle protein mass

L-asp: L-aspartic acid
D-asp: D-aspartic acid
L-aspTS: L-aspartic acid+tail suspension
D-aspTS: D-aspartic acid+ tail suspension
1.72 ± 0.21mg in the L-aspartic acid group, 1.92 ± 0.07mg in the D-aspartic acid group, 1.65 ± 0.17mg in L-aspartic acid TS group and 1.25 ± 0.12 in the D-aspartic acid TS group. Results of a two-way ANOVA of soleus muscle protein content did not show a significant difference between the drinking water groups (Fig. 3). On the other hand, a statistically significant difference at the 1% level between the TS groups was shown, revealing that there are more interactions. Therefore, when a test of the simple main effects was conducted, a significant difference was observed in the TS group.

Accordingly, a multiple comparison test was also performed. And results showed that the L-aspTS group, when compared to the D-aspTS group, showed a statistically significantly higher value. Also a test for simple main effects for, as a result, a significant difference was observed in the group administered with D-asp. Then, as a result of multiple comparison test, as compared to D-aspTS group D-asp groups were significantly higher.

Effect of aspartic acid magnesium intake on skeletal muscle wet weight under electron transport system inhibitor antimycin A dosage

As shown in Fig. 4, the mean and standard deviation of Phosphate buffered saline injection - Water drinking group (PBS-W group), Phosphate buffered saline injection - L-aspartic acid magnesium drinking group (PBS-DMg group), 1mg Antimycin A injection - Water drinking group (1AA-W group), and 1mg Antimycin A injection - L-aspartic acid magnesium drinking group (1AA-DMg group), 2 mg Antimycin A injection - Water drinking group (2 AA-W group), and 2 mg Antimycin A injection - L-aspartic acid magnesium drinking group (2 AA-DMg group) were 0.23 ± 0.01 mg/g, 0.22 ± 0.03 mg/g, 0.18 ± 0.01 mg/g, 0.23 ± 0.01 mg/g, 0.21 ± 0.01 mg/g, and 0.22 ± 0.01 mg/g, respectively. The muscle wet weight per body weight of soleus muscle between the PBS-W group and 1AA-W group, and between 1AA-W group 2AA-W group, was significantly statistically different at a 1% risk rate. Also, a statistically significant difference was obtained at a 5% risk rate between the 1AA-W group and 1AA-DMg group. Plantaris muscle wet weight per body weight was not significantly statistically different between any of the groups.

Effect of aspartic acid magnesium intake on muscle protein mass under electronic transmission system inhibitor antimycin A dosage

As shown in Fig. 5, the mean and standard deviation of the PBS-W group, PBS-DMg group, 1AA-W group, 1AA-DMg group, 2AA-W group and 2AA-DMg group were 0.92 ± 0.07 mg, 1.04 ± 0.15 mg, 0.44 ± 0.09 mg, 0.59 ± 0.14 mg, 0.23 ± 0.02 mg and 047 ± 0.06 mg, respectively. Soleus muscle protein mass was significantly statistically different at a 1% risk rate between the control group and the antimycin A-treated group (Fig. 5). In addition, a significant difference was seen at a 1% significance level between the W group and DMg group of the antimycin group given 1 mg/kg. Furthermore, a significant difference was seen at a 5% significance level between the W group and DMg group of the antimycin group given 2 mg/kg. A statistically significant difference wasn’t observed among groups concerning protein content of the plantaris muscle.
Effect of aspartic acid magnesium intake on muscular cell area under electronic transmission system repressor antimycin A dosage

As shown in Fig. 6, the mean and standard deviation of PBS-W group, PBS-DMg group, 1AA-W group, 1AA-DMg group, 2AA-W group and 2AA-DMg group were 1021 ± 240μm², 1032 ± 213μm², 479 ± 112μm², 813 ± 190μm², 541 ± 116μm² and 681±152μm², respectively. A 1% statistically significant decrease in the muscle cell area of the soleus muscle was observed between the control and antimycin A-treated group. In addition, a similarly significant increase was observed in the W group and D-Mg group of the antimycin A-treated group. A statistically significant difference in muscle cell area of the plantar muscle was observed in all the groups.

Effect of aspartic acid magnesium on skeletal muscle atrophy due to mineral imbalance

Dynamics of tissue minerals in muscle atrophy. Kondo et al., who induced disuse muscle atrophy in rats, reported
a change in concentrations of iron (Fe), zinc (Mn), copper (Cu), manganese (Mn) and calcium (Ca) during 8 days of muscle tissue disuse\(^9\). As muscle atrophy occurred rapidly in the first eight days, and then decelerated, the concentration of Fe increased. In addition, there were no changes in the concentration of Cu. On the other hand, concentrations of Mn and Zn increased to day 8, then showed a biphasic change toward decreasing. This change is reported to be related to the rate of progression of muscle atrophy. Furthermore, since Ca concentration was significantly elevated, calpain, a proteolytic system, and XOD were activated\(^29\).

**Dynamics of mineral and Reactive oxygen species (ROS) production mechanism in vivo in disuse muscle atrophy.**

The generation of ROS in disuse muscle atrophy has been reported\(^21-24\). Generally, super oxide (O\(_2^{-}\)), hydrogen peroxide (H\(_2\)O\(_2\)), and a hydroxyl radical (HO) are included in ROS. Among ROS defense factors, glutathione peroxidase includes Se, and SOD includes Mn, Cu and Zn. Furthermore, catalase includes Fe, and MT includes Zn.

However, minerals not only protect ROS, but also sometimes serve to generate ROS. As an example, an increase in Ca activates calpain. Activated calpain activates XOD, and activated XOD produces a superoxide from O\(_2\);\(^29\). The most dangerous hydroxyl radicals for the living body are produced in the presence of transition metals such as Fe and Cu from superoxide and hydrogen peroxide\(^19\). As it is well-known, calpain is activated by disuse muscle atrophy, and the amount of glutathione peroxidase and catalase to metabolize hydrogen peroxide dramatically increases. Therefore, the generating mechanism of ROS in disuse muscle atrophy is shown in Fig. 7. The hydroxyl radical produced by such a mechanism promotes adipose hyperoxidation. As a result, atrophy of the skeletal muscle is promoted\(^9\).

As noted above, ROS negatively regulates the myogenic regulatory factors (MRF) family\(^10,25\). In addition, ROS also inhibits protein synthesis. Research on muscle atrophy suppression using vitamin E\(^26\) and astaxanthin\(^27,29\) with an antioxidant have been made. In this way, an imbalance of minerals occurs causing the homeostasis collapse of minerals in muscle tissue and cells\(^28\).

The authors examined the effect of mineral intake on the mineral dynamics and muscle atrophy. The authors used a tail suspension (TS) as a muscle atrophy-induced model. The mean and standard deviation (SD) of W group, WTS group, D-Mg group and D-MgTS group were 0.28 ± 0.06 mg/g, 0.25 ± 0.04 mg/g, 0.28 ± 0.03 mg/g and 0.28 ± 0.04 mg/g, respectively. The mean of soleus muscle weight per body weight in tail-suspension for water intake (water treatment system or WTS) group collectively significantly decreased in comparison with the mean of soleus muscle weight to body weight of individual (WTS) group members. However, the mean of soleus muscle weight per body weight of the aspartic acid magnesium intake (D-MgTS) group did not significantly differ from the soleus muscle weight per body weight of the aspartic acid magnesium intake (D-Mg) group (Fig. 8). In addition, skeletal muscle enlargement was not observed in comparing the mean of sole muscle weight per body weight of the W group and D-Mg group. Moreover, with the plantaris muscle, there was no significant difference in the means between the groups. In this way, muscle weight in the soleus muscle under tail suspension rapidly decreased.

![Fig. 7 Mechanism of ROS production in disuse muscle atrophy](image-url)
Fig. 8  Inhibitory effect of aspartic acid magnesium for soleus muscle atrophy in the tail suspension
W: water intake
WTS: water intake+tail suspension
D-Mg: aspartic acid magnesium intake
D-MgTS: aspartic acid magnesium intake+tail suspension

over 7 days, and a strong correlation was observed between muscle weight and muscle cell area (r = 0.81). In the WTS group soleus muscle, Ca significantly increased and Mg decreased in the cell. Therefore, it is suggested that Ca is a trigger that enhances the proteolytic system in muscle cells. On the other hand, in the soleus muscle of the D-MgTS and D-Mg groups, an increase in Ca and decrease in Mg were not observed. In other words, Mg density in the cell was maintained by taking in D-Mg. As a result, an excessive inflow of the Ca producing active oxygen was suppressed, and it is thought that muscle atrophy was controlled.

Concerning cell area measurement results, between the W group and WTS group, the WTS group and D-MgTS group, and the WTS group and the D-ZnTS group, the mean values of the soleus muscle significantly differed. The cell area value of the plantar muscle was not statistically different when comparing the mean values among the above groups. The amount of protein in the soleus muscle, between the W group and WTS group, and the WTS group and D-MgTS group, significantly differed from average values. There was no significant difference between any groups concerning the plantaris muscle. The results of quantitative analysis of Ca, Mg and Zn in skeletal muscle in the soleus muscle showed no significant difference in the D-MgTS group, whereas Ca in the WTS group significantly increased. Mg in soleus muscle showed a decreasing trend in the WTS group; but there was no significant difference in the other groups. Ca, Mg and Zn concentration in the plantaris muscle did not significantly differ in any group. We reviewed the inhibitory effect of aspartic acid magnesium on disuse muscle atrophy based on the experiments conducted so far.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this article.

References


