The relationship between follistatin and sarcopenia in elderly

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ABSTRACT

Introduction: Sarcopenia is a multifactorial condition characterized by progressive and comprehensive mass loss accompanied by skeletal muscle strength or function. It can be triggered by physical inactivity, hormonal changes, energy, protein intake, oxidative stress and inflammatory processes. The state of inactivity underlying sarcopenia is related to follistatin levels.

Methods: an analytical cross-sectional study conducted in Posyandu or Elderly Associations in Denpasar City with subjects aged ≥60 years old started from January 2021 to September 2021. The data collected were anthropometric measurements, muscle mass, grip strength, walking speed and serum follistatin levels. Sarcopenia was diagnosed based on the AWGS criteria. Bivariate analysis was performed using the Mann-Whitney analysis to determine the difference in the mean of the independent variables and the Spearman’s test to determine the relationship between sarcopenia and follistatin levels.

Results: 75 subjects were selected, and 15 (20%) subjects were diagnosed with sarcopenia. The results of the Spearman correlation test of upper arm circumference, waist circumference, calf circumference, SMI, walking speed, hand grip strength and follistatin levels only found a significant negative relationship between waist circumference and follistatin levels r = -0.309, p = 0.016. No differences in mean follistatin levels with sarcopenia and non-sarcopenia group p = 0.516. There was no significant relationship between follistatin levels and sarcopenia p = 0.615.

Conclusion: There is no correlation between follistatin levels and sarcopenia in the elderly. However, follistatin is inversely correlated with waist circumference.

Keywords: sarcopenia, follistatin, elderly.


INTRODUCTION

The elderly population will continue to grow as life expectancy increases. The elderly population in 2020 was about 727 million.1 By 2050, the elderly population is expected to triple to approximately 1.5 billion (16% of the world’s population).2 The projection of the Indonesian population in 2010 to 2035, it is predicted that the elderly group will have a proportion of 10% of the entire population of Indonesia in 2020.2 In 2020, Bali had an elderly population of 11.58%, making it the 4th provision for the highest percentage of elderly people in Indonesia.3

The aging process results in changes in both physical and mental a person. Physical changes associated with a progressive decrease in the physiological function of all organs, including muscles, are called sarcopenia. Sarcopenia will reduce the mobility of the elderly resulting in increased morbidity and mortality.4,5 Sarcopenia is a syndrome characterized by progressive and comprehensive loss of mass accompanied by skeletal muscle strength or function, which is at the risk of causing physical disability, low quality of life, and death.6,7 Sarcopenia arises due to the disruption of the complex balance between anabolic and catabolic factors of muscle protein with age, thus affecting the tropism and efficiency of skeletal muscle.8 It is known that there is a multifactorial process that triggers sarcopenia, namely loss of alpha motor nerve input in the spinal column, physical inactivity, hormonal changes, energy, protein intake, oxidative stress, and inflammatory processes. Subclinical inflammation and oxidative stress are the basic mechanisms of sarcopenia because they can trigger the release of catabolic stimuli.9

The mechanism of sarcopenia involves decreased protein synthesis and increased protein degradation.10 The regulation of muscle mass is regulated by proteins such as follistatin, myostatin and activin A.11 Follistatin is a glycoprotein that can inhibit the activity of myostatin and activin A. Studies biomolecular and laboratory studies of follistatin showed significant results. Administration of follistatin in experimental rats caused muscle hypertrophy in rats.12 Follistatin can be affected by physical activity based on the study by Hansen et al. reported significant results with increased follistatin levels in both humans and experimental rats after the administration of physical exercise.13
The state of inactivity underlying sarcopenia is related to follistatin levels. Therefore, the researcher intends to examine the relationship between follistatin levels and sarcopenia in the elderly. This is very important to study because sarcopenia can cause morbidity and mortality and reduce the life expectancy of the elderly.

METHODS

Study Design and Sampling
This is an analytical cross-sectional study to determine the relationship between myokines and metabolic syndrome in the elderly. The research was conducted in Banjars around Denpasar City, which has elderly Pospayu or elderly associations, from January 2021 to September 2021. The research sample is all residents aged more than or equal to 60 years living or in Denpasar City who meets the inclusion criteria and does not meet the exclusion criteria. The sample size formula calculates the sample size. The minimum number of samples needed is 74 samples. Samples were taken successively using consecutive sampling from the elderly population living or residing in Denpasar City that met the research criteria until it met the required number of samples. The inclusion criteria in this study were aged more than or equal to 60 years. Exclusion criteria were patients suffering from acute infection, patients with malignancy, and patients who had taken aspirin or NSAIDs for at least the last 2 weeks.

Data Collection
The data collected were anthropometric measurements, muscle mass, grip strength, walking speed, and serum follistatin levels. Sarcopenia parameters used were based on the Asian Working Group for Sarcopenia (AWGS), which consisted of muscle mass, muscle strength, and physical performance. Muscle mass was measured using BIA (Tanita MC 980 Plus) with a skeletal muscle index <7.0 kg/m² in men and <5.7 kg/m² in women, classified as low muscle mass. Muscle strength was measured based on grip strength (Camry), with grip strength <26 kg in men and <18 kg in women classified as low grip strength. Physical performance was measured by walking speed as far as 5 meters with walking speed <0.8 m/s which was classified as low walking speed. Sarcopenia is defined as low muscle mass, low muscle strength, and/or low physical performance. Meanwhile, serum follistatin levels were measured by enzyme-linked immunosorbent assay (ELISA).

Statistical Analysis
Bivariate analysis was performed using the t-test to determine the difference in the distribution of the control variables, the difference in the mean of the independent variables and Spearman’s test to determine the relationship between sarcopenia and follistatin. Data were analyzed using SPSS software version 23.0.

RESULTS

Characteristics of Research Subjects
The study was conducted in Denpasar City from January 2021 to September 2021. During the study period, 75 subjects were selected according to the inclusion and exclusion criteria. The research subjects comprised 30 men (50%) and 45 women (45%) with an average age of 68.57 years. Twenty-eight people (37.3%) had an elementary school educational background, and 23 (30.7%) worked as entrepreneurs. Based on marital status, 2 people (2.7%) were not married, 58 people (77.3%) were married, and 15 people were divorced (20%).

Based on the clinical character table from 75 subjects in this study, 15 (20%) subjects were diagnosed with sarcopenia. The median knee height of the research subjects was 48.5 cm, with the lowest value of 41 cm and the highest at 70 cm, and the median upper arm circumference was 27 cm, with the lowest value of 21 cm and the highest at 37 cm. The average BMI of the research subjects was 23.60 kg/m², with a median height of 160 m and an average weight of 60.913 kg. Based on blood pressure, the research subjects’ median systolic blood pressure was 150 mmHg, with the lowest value of 100 mmHg and the highest value of 190 mmHg. While diastolic blood pressure, the median value was 80 mmHg, with the lowest value 60 mmHg and the highest value 110 mmHg. A total of 61 people (81.3%) had a functional status in the category of independent dependence and a CACI score <2, as many as 56 people (74.7%).

Analysis of the Relationship of Follistatin Levels with Sarcopenia
Based on the data normality test, the results showed that the levels of follistatin and sarcopenia were not normally distributed. To determine the distribution of differences between the variable levels of follistatin with sarcopenia by performing a Mann-Whitney analysis. The analysis results show no difference in the mean follistatin levels in the sarcopenia and non-sarcopenia groups.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Normal</th>
<th>Sarcopenia</th>
<th>p value for mean</th>
<th>r</th>
<th>p-value for correlation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Follistatin levels</td>
<td>60 (80)</td>
<td>15 (20)</td>
<td>0.516</td>
<td>-0.059</td>
<td>0.615</td>
</tr>
</tbody>
</table>

Table 2. Correlation of upper arm circumference, waist circumference, calf circumference, SMI, walking speed, hand grip strength to follistatin levels.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Normal</th>
<th>Follistatin</th>
<th>Sarcopenia</th>
</tr>
</thead>
<tbody>
<tr>
<td>Upper arm circumference</td>
<td>0.004</td>
<td>0.977</td>
<td>0.273</td>
</tr>
<tr>
<td>Waist circumference</td>
<td>-0.309</td>
<td>0.016*</td>
<td>0.280</td>
</tr>
<tr>
<td>Calf circumference</td>
<td>0.205</td>
<td>0.117</td>
<td>-0.061</td>
</tr>
<tr>
<td>SMI</td>
<td>-0.034</td>
<td>0.799</td>
<td>0.032</td>
</tr>
<tr>
<td>Walking speed</td>
<td>-0.144</td>
<td>0.271</td>
<td>-0.234</td>
</tr>
<tr>
<td>Hand Grip Strength</td>
<td>-0.164</td>
<td>0.210</td>
<td>-0.068</td>
</tr>
</tbody>
</table>

Note: * = statistically significant

Table 1. Differences in Mean Follistatin Levels with Sarcopenia.
Analysis of the relationship between levels of follistatin and sarcopenia used Spearman analysis because data distribution on levels of follistatin and sarcopenia was not normal. The result of the analysis of the relationship between follistatin levels and sarcopenia is that there is no significant relationship between follistatin levels and sarcopenia.

The results of the Spearman correlation test for upper arm circumference, waist circumference, calf circumference, SMI, walking speed, hand grip strength and follistatin levels indicate a significant negative relationship between waist circumference and follistatin levels. The interpretation of the results is that the greater the waist circumference, the lower the follistatin levels in the elderly group who do not experience sarcopenia.

**DISCUSSION**

A total of 75 subjects were included in this study, consisting of 30 men (50%) and 45 women (45%) with an average age of 68.57 years. In this study, 15 (20%) subjects were diagnosed with sarcopenia. This study, similar to a systematic review and meta-analysis study by Petermann-Rocha et al., stated that the global prevalence of sarcopenia is 10-27%. Papadopoulou et al., in their systematic review and meta-analysis study, stated that the prevalence of elderly sarcopenia in the population living in the community was 11% in men and 9% in women, in the population living in nursing homes was 51% in men and 31% in women, and the hospital population was 23% in men and 24% in women.

The results showed no significant difference in the mean levels of follistatin between the sarcopenia and non-sarcopenia groups. The analytical test also showed no significant relationship between follistatin levels and sarcopenia. There was also no significant relationship between follistatin levels and each component of sarcopenia, such as SMI, hand grip strength and walking speed. The results of this study are consistent with the results of a systematic review study by Marguti KM et al. in 2017, which found that follistatin (and 4 other biomarkers) was not associated with sarcopenia. The study by Choi K et al. in 2020 also found that follistatin levels were not associated with the incidence of sarcopenia in patients with hepatocellular carcinoma (HCC).

Different results were obtained in several other studies. A study by Du Y et al. in 2021 found that follistatin levels were associated with the incidence of sarcopenia in postmenopausal women (OR 1.66; 95% CI 1.19-3.57; p= 0.022), where sarcopenia patients had higher follistatin levels than patients with sarcopenia compared to patients who are not sarcopenia. A 2018 Fife et al. also found a significant relationship between follistatin levels and muscle performance in elderly women.

This study showed no significant relationship between follistatin levels and each component of sarcopenia diagnoses, such as hand grip strength, skeletal muscle index and walking speed. This study, similar to research by Liaw et al. and Fife et al., also stated that follistatin levels were not associated with hand grip strength. Research by Hoffmann et al. also stated that there is no relationship between follistatin levels and muscle mass. A study by Sordi et al. tried to correlate the levels of follistatin and myostatin (follistatin antagonist) with idiopathic musculoskeletal inflammatory myopathy, namely dermatomyositis and polymyositis are inflammatory diseases of the skeletal muscles that cause weakness in skeletal muscles.

In contrast to the study of sarcopenia in humans with varying or insignificant results, the biomolecular and laboratory follistatin in animal studies have shown significant results. Administration of follistatin in experimental mice causes muscle hypertrophy of mice. Administered follistatin to injured muscles can accelerate repairment and was a very potent muscle regeneration. Follistatin causes muscle hypertrophy in mice and can regulate the action of insulin in taking glucose, which induces hypertrophy in muscles.
ultimately leading to hypertrophy and muscle growth.11 Another study also found that high levels of follistatin can cause a decrease in muscle strength in patients with chronic kidney disease24 and also cause a decrease in walking speed in the elderly.28 This may be due to an increase in follistatin levels, also accompanied by an increase in myostatin levels.24 However, the role of follistatin is not exclusively limited to the activity of myostatin and activin A but can also mediate the action of insulin in muscle with overexpressed levels of follistatin that will increase insulin-mediated glucose uptake in muscle.25 Follistatin mediated skeletal muscle hypertrophy and was regulated independently of myostatin.26

Follistatin is not associated with sarcopenia, as stated by Hoffmann et al. Research that follistatin can act as an acute marker rather than the chronic state of muscle loss and is more sensitive to changes in energy balance such as exercise or acute activity.21 Hansen et al. stated that follistatin levels increased significantly in mice given a bicycle and swimming exercises.13 The study by Domin et al. also stated that follistatin levels increased significantly after acute, either resistance training, endurance training or high-intensity exercise.27

Sarcopenia parameters used were based on the Asian Working Group for Sarcopenia (AWGS), which consisted of muscle mass, muscle strength, and physical performance and sarcopenia is defined as low muscle mass and low muscle strength and/or low physical performance.5 Follistatin studies in humans seem to show more significant results on physical performance than muscle mass19,21 and muscle strength.19,20 As mentioned in several studies, follistatin is correlated with the timed up and go test in the elderly19, follistatin was associated with walking speed in the elderly20, and high follistatin was associated with a rise from a seat test.21 Several studies above show varying results in the components of the physical performance of sarcopenia. In its systematic review study, Marguti et al. stated that follistatin was only weakly correlated with the physical performance of the elderly.16

The subgroup analysis of the sarcopenia and normal groups showed a significant negative relationship between waist circumference and follistatin levels. The greater the waist circumference, the lower the follistatin level in the elderly group who did not experience sarcopenia. According to WHO, waist circumference is used to assess central obesity. Increased waist circumference will be at risk for central obesity, which causes the release of many inflammatory cytokines by visceral fat (abdominal fat). This inflammation will increase the occurrence of sarcopenia. Follistatin is not only affected by muscle mass but also by visceral fat tissue.28 Hansen and Plomgaard stated that inflammatory conditions influence follistatin. However, the role of follistatin in the inflammatory process is still not completely clear.29 A study by Flanagan et al. stated that follistatin levels decreased significantly in obese subjects compared to non-obese controls.30 Follistatin is a hepatokine similar to other hepatokines and is associated with the abnormal development of glucose and lipid metabolism in obesity.31 Hepatokines can increase hepatic fat accumulation and inflammatory signaling that exacerbates glucose metabolism and insulin signaling.32 A study by Han et al. stated that follistatin injection in mice causes muscle hypertrophy of mice and follistatin is a potent regulator of insulin action that can improve muscle glucose uptake in insulin-resistant obese mice.25

Current studies and research on follistatin with sarcopenia or diagnosis components of sarcopenia in the human population had varying results compared to follistatin studies in experimental animals with more significant consistent results. With the results of these different studies, further studies are needed with larger sample size and prospective methods to find out more clearly about the actual mechanism of action of follistatin and its relationship with the incidence of sarcopenia in the elderly.

CONCLUSION

This study concludes that there is no correlation between levels of follistatin and sarcopenia in the elderly. However, follistatin is inversely correlated with waist circumference. Further studies with larger sample size and prospective methods to clearly confirm the actual mechanism of action of follistatin and its relationship with the incidence of sarcopenia in the elderly.

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CONFLICT OF INTEREST

The author reports no conflicts of interest in this work.

ETHICS APPROVAL

This research has received ethical approval from the Faculty of Medicine Ethics Committee, Udayana University/Sanglah Hospital with the number 2245/UN14.2.2.VII.14/LT/2021.

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AUTHOR CONTRIBUTION

All authors contributed equally in the writing of this article.

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