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Decreased Follistatin Levels as a Risk of Acute Sarcopenia Marker in Elderly

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Background: Acute sarcopenia is an acute muscle loss that has been associated to the frailty and vulnerability of the elderly. Follistatin has been known as a significant marker for sarcopenia, however, studies of follistatin in humans have shown varying results and there have been no studies to date regarding the relationship between follistatin and acute sarcopenia. The aim of this study was to determine changes in follistatin levels as a risk of acute sarcopenia in elderly.

Materials and methods: This study was a prospective observational study involving hospitalized elderly. The follistatin level was examined with enzyme-linked immunosorbent assay (ELISA). Meanwhile the determination of acute sarcopenia was done through the measurement of changes in hand grip strength and calf circumference parameters. The data obtained was descriptively analyzed, followed by bivariate and multivariate analysis. A $p<0.05$ was considered significant.

Results: There were 66 subjects in this study. A total of 10 subjects (15.2%) had acute sarcopenia on the 7th day of hospitalization. The cut-off point of decreased follistatin levels was 4.870 with a sensitivity of 82.1% and a specificity of 60%. There was an association between decreased follistatin levels and acute sarcopenia ($p=0.01$; RR: 6.90; 95% CI: 1.638-29.069). Multivariate analysis results showed that decreased follistatin levels was a significant factor that might influence the occurrence of acute sarcopenia.

Conclusion: Since this study showed that decreased follistatin levels might be a risk of acute sarcopenia in the elderly, thus it could be used as a marker of acute sarcopenia, which should be further investigated.

Keywords: *decreased follistatin levels, acute sarcopenia, elderly*

Introduction

Muscles contribute greatly to total body weight and will change qualitatively and quantitatively with aging. Sarcopenia according to the Asian Working Group for Sarcopenia (AWGS) is defined as a loss of skeletal muscle mass due to aging and loss of muscle strength with or without a decrease in physical performance.[1] The European Working Group on Sarcopenia in Older People 2nd Update (EWGSOP2) divides sarcopenia into acute and chronic sarcopenia to emphasize the need for periodic assessment of sarcopenia in at-risk individuals.[2] Acute sarcopenia is a condition of acute muscle loss with the elderly being more susceptible to the effects of hospitalization.[3] It has been

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reported that the incidence of acute sarcopenia in the elderly in hospital is 14.7-75%.[4,5]

Follistatin is a glycoprotein that can promote muscle growth.[6] Expression of natural follistatin in plasma promotes muscle hypertrophy due to satellite cell activation.[6] As a hepatokine, follistatin is not expressed in contracting skeletal muscle, instead it was expressed in the liver.[7] Level of follistatin has been related to the occurrence of sarcopenia. In experimental rats, administration of follistatin resulted in muscle hypertrophy.[8-10] In an experimental model of rats given exercise, natural follistatin levels were significantly increased in plasma and liver tissue.[11] Several studies reporting the significant association of the follistatin levels and the physical performance of the elderly.[12-15] Follistatin levels were said to be increased in both humans and experimental mice after physical exercise.[6] The state of bed rest underlying acute sarcopenia with minimal physical activity is thought to be related to follistatin levels. Hence, follistatin has been suggested as a possible marker of sarcopenia.

Patients with sarcopenia progression spent an average of 5.1 days of hospitalization.[5] Muscle mass and muscle strength started to significantly decrease after only 5 days of immobilization.[16] The effect of bed rest was more significant in the elderly compared to non-elderly showing that bed rest for 5 days can result in loss of lean mass and strength.[17] Follistatin production is stated largely influenced by muscle contraction so that physical inactivity can change its profile and response.[11] Elderly who were hospitalized with elderly men, age ≥65 years, dependence on physical activity, malnutrition and length of stay 7 days were factors associated with the development of acute sarcopenia.[18] Acute illness can lead to an increased inflammatory burden, a decrease in activity and a decrease in muscle use that can lead to an acute decrease in muscle function and mass.[17]

The role of follistatin as a biomarker and clinical correlation in human muscle still needs further attention through various studies with varying results. Asians tend to have lower muscle mass, weaker grip strength and lower walking speeds than people from Western countries.[19] Some studies of follistatin in humans have been carried out outside Asia and as far as researchers know to date, there have been no studies conducted regarding the role of follistatin in acute sarcopenia. An understanding of follistatin levels in the elderly can be used as a reference in preparing intervention guidelines during treatment to prevent morbidity and mortality due to acute sarcopenia in the elderly. The aim of this study was to determine changes in follistatin levels as a risk for acute sarcopenia in the elderly.

Materials and methods

Study Design

This study was a prospective cohort observational study with a 7-day follow-up period involving elderly who were admitted to Prof. Dr. IGNG Ngoerah Teaching Hospital, Denpasar, Bali, Indonesia. Sampling was carried out through consecutive sampling from April to September 2021 in a non-isolated room at Prof. Dr. IGNG Ngoerah Teaching Hospital. The protocol of this study has been approved by Ethic Committee of the Faculty of Medicine, Universitas Udayana (No: 454/UN14.2.2.VII.14/LT/2021) and subjects had given informed consent before the study started.

The inclusion criteria were elderly aged more than 60 years old who had just been admitted to hospital since the first day in a non-isolated room. Subjects with cirrhosis were excluded from the study since follistatin levels were significantly decreased in patients with cirrhosis.[20] Elderly with edema in the calf and trauma or musculoskeletal injuries of the upper or lower extremities which could interfere with the measurement of hand grip and calf circumference for the diagnosis of sarcopenia were also excluded from the study. Measurements were carried out twice, on the first day of subjects' hospital admission and on the 7th day in the hospital.

Follistatin Levels Measurement

Follistatin levels were drawn from subjects' blood and measured with the sandwich enzyme-linked immunosorbent assay (ELISA) method with assay from Bioassay Technology Laboratory (Cat. #E1010Hu, Shanghai Korain Biotech, Shanghai, China). This kit was used for quantitative detection of human follistatin in serum, and the values were recorded in ng/mL. Follistatin that was presented in the sample was added and bound to the biotinylated human follistatin antibody. Initial follistatin levels were follistatin taken at the beginning or within 24 hours of hospital admission. Changes in follistatin levels were decreased or increased levels of follistatin after samples were taken on the 7th day compared to initial follistatin levels.

Hand Grip Strength Measurement

Hand grip strength was measured with a handheld dynamometer Camry model EH101 (Zhongshan Camry

Electronic, Zhongshan China). The highest value was recorded for at least 2 trials using each hand or dominant hand with a 30 second pause between experiments.[1,21] The handgrip was measure with subjects in a sitting position and the elbow flexed at 90° then the values were recorded to the nearest 0.1 kg.[1,21] Low hand grip strength based on AWGS <28.0 kg in men or <18.0 kg in women.[1]

Calf Circumference Measurement

Calf circumference was measured according to the recommendations of the International Society for the Advancement of Kinenthropometry (ISAK) with a non-elastic measuring tape that circled the calf horizontally and affixed to the skin but did not compress the underlying tissue to obtain the largest circumference measurement. The sample was in sitting position and the leg upright straight with the ground of the foot beneath it and then measured the highest value for at least 2 trials. The value was recorded to the nearest 0.1 cm.[21-23] Low calf circumference based on AWGS <34.0 cm in men or <33.0 cm in women.[1] Calf muscle area in the gastrocnemius and soleus muscles decreased by 12% after bed rest for 5 weeks or there was a 2.4% decrease in calf muscle area in 1 week.[24] Some studies suggested that calf circumference measurement could replace bioelectrical impedance analysis (BIA) or dual-energy x-ray absorptiometry (DXA) for the diagnosis of sarcopenia muscle mass and were better at describing muscle mass than other anthropometric or circumference measurements.[25,26]

Sarcopenia Determination

Sarcopenia was defined as low muscle strength and low muscle mass based on the AWGS that diagnosed within 24 hours of hospital admission with low muscle strength parameters (low hand grip strength <28.0 kg in men or <18.0 kg in women) and low muscle mass (low calf circumference <34.0 cm in men or <33.0 cm in women).[1] In this study, the result of sarcopenia determination was divided into sarcopenia and non-sarcopenia.

Acute sarcopenia was defined as a decrease in muscle strength and a decrease in muscle mass that occurs 7 days from the beginning of hospital admission with a decrease in muscle strength parameters (decrease of 3 kg of hand grip strength from the initial hand grip strength at hospital admission) and a decrease in muscle mass (decrease in calf circumference 2.4% from the initial calf circumference at hospital admission).[3,24] The total subjects at initial hospital admission (both sarcopenia and non-sarcopenia) were followed for 7 days to assess the acute sarcopenia.

Other factors associated with acute sarcopenia then defined such as elderly categorized to low-risk with elderly aged 60-69 years and high-risk with elderly ≥70 years.[27] Cognitive impairment was measured with the abbreviated mental test (AMT). Delirium was measured with the confusion assessment method (CAM). Depression was measured with the geriatric depression scale short form (GDS-15). Nutritional status was measured with a mini nutritional assessment (MNA). Activities of daily living (ADL) were measured with the Barthel activities of daily living index. Infectious diseases were assessed through the patient's medical records. Chronic comorbid disease was measured with the Charlson comorbidity index (CCI).

Statistical Analysis

Kolmogorov-Smirnov test was used for the normality test. Analysis with chi-square test was used to determine the association of follistatin levels with sarcopenia. Changes in follistatin levels were determined by the best cut-off point based on receiver operating characteristic (ROC) curve and then a dichotomous category was determined based on that cut-off point. Bivariate analysis was performed to determine the factors associated with acute sarcopenia. Logistic regression analysis was performed to determine the most important variable in acute sarcopenia. Statistical Package for the Social Sciences (SPSS) ver. 23 (IBM Corporation, Armonk, NY, USA) was used for data analysis. The $p<0.05$ was used to determine the study's significance level with a 95% confidence interval (CI).

Results

Characteristics of Research Subjects

The demographic characteristics data of the research subjects are listed in Table 1. This study involved 66 subjects with 19 (28.8%) subjects meeting the criteria for sarcopenia. All subjects were then observed for 7 days, and it was found that 10 (15.2%) subjects were meeting the criteria for acute sarcopenia. The clinical characteristics of the subjects are listed in Table 2.

Follistatin Levels and Sarcopenia

Median of follistatin levels on admission day was 18.24 (10.00-48.42). The results of the low and high follistatin bivariate analysis on sarcopenia at the time of initial hospital

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Table 1. Demographic characteristics of subjects.

Characteristics	Value
Age (years)[a]	67 (60-84)
60-69 years old[b]	42 (63.6%)
≥70 years old[b]	24 (36.4%)
Gender[b]	
Man	27 (40.9%)
Woman	39 (59.1%)
Employment history[b]	
Trader	18 (27.3%)
Housewife	15 (22.7%)
Self-employed	14 (21.2%)
Farmer	12 (18.2%)
Laborer	2 (3.0%)
Health workers	1 (1.5%)
Others	4 (6.1%)

[a]Data presented as median (min-max), [b]Data presented as n (%).

admission showed that the results were not statistically significant with p=0.415, as shown in Table 3.

Changes in Follistatin Levels and Acute Sarcopenia

Decreases in follistatin levels were analyzed using Receiver Operator Characteristic (ROC) (Figure 1). The best cut-off point of 4.870 (AUC=0.721; p=0.027) with a sensitivity of 82.1% and a specificity of 60%. Changes in follistatin levels that decreased >4.870 and decreased ≤4.870 based on the ROC cut-off point for acute and non-acute sarcopenia after the chi square test showed significant results with p=0.01 (RR: 6.90; 95% CI: 1.638-29.069) as shown in Table 4.

Bivariate and Multivariate Analysis

Bivariate chi square analysis was conducted to determine the relationship of factors associated with acute sarcopenia indicating cognitive impairment (RR: 10.733; 95% CI: 2.358-48.858; p=0.002), delirium (RR: 12.500; 95% CI: 2.727-57.290; p=0.001), depression (RR: 16.200; 95% CI: 1.912-137.288; p=0.004), ADL (RR: 9.000; 95% CI: 1.068-75.837; p=0.034), and CCI (RR: 7.200; 95% CI: 1.392-37.231; p=0.014) was significantly associated with acute sarcopenia, while age (RR: 1.400; 95% CI: 0.326-6.008; p=0.736), gender (RR: 0.957; 95% CI: 0.242-3.774; p=1.000), initial follistatin levels (RR: 1.00; 95% CI: 0.260-3.841; p=1.00), nutritional status (RR: 3.467; 95% CI: 0.675-17.801; p=0.170) and infectious diseases (RR: 1.435; 95% CI: 0.372-5.529; p=0.732) not associated with acute sarcopenia.

Table 2. Clinical characteristics of subjects.

Characteristics	Value
Weight (kg)[a]	50.2 (34.0-90.0)
Height (cm)[a]	161.5 (148.5-177.7)
Body mass index (kg/m^2)[b]	20.2±4.1
Initial hand grip strength (kg)[b]	13.8±4.4
Hand grip strength on day 7th (kg)[b]	11.3±5.0
Changes in hand grip strength (kg)[b]	3.2±2.4
Initial calf circumference (cm)[a]	33.4 (20.7-41.0)
Calf circumference on day 7th (cm)[a]	33.3 (20.2-41.0)
Changes in calf circumference (cm)[a]	0.05 (0.0-0.9)
Initial follistatin level, median (ng/mL)[a]	18.24 (10.00-48.42)
Low <18.24[c]	33 (50.0%)
High ≥18.24[c]	33 (50.0%)
Follistatin level on day 7th (ng/mL)[a]	19.52 (8.29-135.00)
Changes in follistatin level (ng/mL)[a]	0.215 (-21.86-90.61)
Decreased >4.870[c]	16 (24.2%)
Decreased ≤4.870[c]	50 (75.8%)
Sarcopenia[c]	
Yes	19 (28.8%)
No	47 (71.2%)
Acute sarcopenia[c]	
Yes	10 (15.2%)
No	56 (84.8%)
AMT score[a]	8 (4-10)
Cognitive impairment by AMT[c]	
Yes	17 (25.8%)
No	49 (74.2%)
Delirium[c]	
Yes	12 (18.2%)
No	54 (81.8%)
GDS score[a]	4 (1-15)
Depression by GDS[c]	
Yes	29 (43.9%)
No	37 (56.1%)
MNA score[b]	15.7±4.0
Nutritional status by MNA[c]	
Malnutrition	38 (57.6%)
Not malnutrition	28 (42.4%)
ADL score[a]	8 (1-19)
ADL[c]	
Total dependency	14 (21.2%)
Severe dependency	23 (34.8%)
Moderate dependency	14 (21.2%)
Mild dependency	15 (22.7%)
Infectious disease[c]	
Yes	28 (42.4%)
No	38 (57.6%)
CCI[a]	5 (2-9)
>5[c]	28 (42.4%)
≤5[c]	38 (57.6%)

[a]Data presented as median (min-max), [b]Data presednted as mean±SD, [c]Data presented as n(%).

Table 3. Analysis of follistatin levels with sarcopenia and the diagnostic components of sarcopenia.

Variable	Sarcopenia		PR	95% CI	*p*-value
	Yes	No			
Follistatin levels, n (%)					
<18.24	11 (33.3%)	22 (66.7%)	1.563	0.533-4.582	0.415
≥18.24	8 (24.2%)	25 (75.8%)			

Diagnostic Components of Sarcopenia	*p*-value	*r*
Follistatin levels (ng/mL) *vs.* Hand grip strength (kg)	0.945	-0.009
Follistatin levels (ng/mL) *vs.* Calf circumference (cm)	0.827	-0.027

Multivariate analysis with logistic regression analysis using the backward logistic regression method contained 9 stages with the results of decreasing follistatin levels (AOR: 24.888; 95% CI: 1.924-321.865; p=0.014), delirium (AOR: 35.526; 95% CI: 2.724-463.307; p=0.006) and depression (AOR: 22.588; 95% CI: 1.358-375.797, p=0.030) were variables that were significantly more influential in the incidence of acute sarcopenia.

Discussion

The incidence of acute sarcopenia in this study was 15.2%, similar to the results of multicenter study which stated that elderly without a diagnosis of sarcopenia on admission, 14.7% met the diagnosis of sarcopenia on discharge.[5] Another study stated that the incidence can up to 75%, however this study was only conducted in one center with a homogenous sample of elderly patients who had undergone major colorectal surgery.[4]

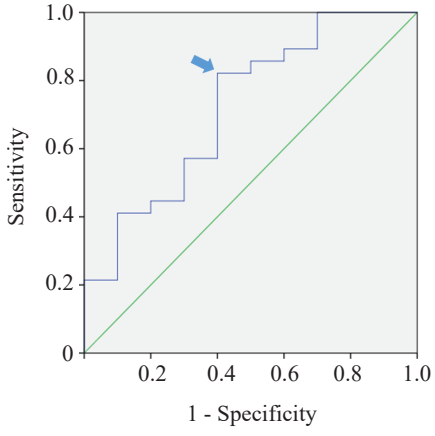

Figure 1. ROC analysis of changes in follistatin levels to acute sarcopenia.

Follistatin levels in this study obtained a median of 18.24 (10.00-48.42) ng/mL that was similar to one study with follistatin levels of 18.97±6.1 ng/mL in sarcopenia condition.[28] Changes in follistatin levels showed a decrease in follistatin levels to acute sarcopenia with the best cut-off point was 4.870. This study showed no relationship between low and high follistatin to sarcopenia that is similar to the results of one study with elderly female population living in an elderly care setting.[13] Our study showed different results to one study which reported that follistatin was significantly associated with sarcopenia.[28] However, that study was a community population of healthy post menopausal women and were still able to carry out daily activities or exercise well.[28]

There was no relationship between follistatin levels and hand grip strength in this study, which is similar to other studies.[12-14] This study also showed that there was no relationship between follistatin levels and calf circumference which indicated a component of muscle mass in sarcopenia similar to other studies.[12,13] One study tried to link levels of circulating follistatin with dermatomyositis and polymyositis diseases (idiopathic musculoskeletal inflammatory myopathies) that cause weakness in skeletal muscles.[29] That study found no relationship between follistatin levels in patients with polymyositis/ dermatomyositis compared with healthy control subjects.[29]

The AWGS diagnostic criteria for sarcopenia include low muscle strength plus low muscle mass with or without poor physical performance.[1] The presence of physical performance criteria seems to have a better relationship with follistatin. Follistatin associated with the timed up and go test in the elderly, high follistatin was correlated with a rise from a seat test, follistatin correlated with walking speed in

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Table 4. Analysis of changes in follistatin levels with acute sarcopenia.

Variable	Acute Sarcopenia		RR	95% CI	*p*-value
	Yes	No			
Changes in follistatin levels, n (%)					
Decrease >4.870	6 (37.5%)	10 (62.5%)	6.9	1.638-29.069	0.01*
Decrease ≤4.870	4 (8.0%)	46 (92.0%)			

*Considered as significant if $p<0.05$.

the elderly, and also follistatin had a weak relationship with the physical performance in the elderly.[12-15]

Human studies of follistatin had more results significantly in physical performance than muscle strength or muscle mass.[12-18] Injection of follistatin directly in experimental rat accelerates muscle regeneration and it was a potent regulator of insulin in taking glucose which then induces rat muscle hypertrophy.[8-10] Injection of follistatin directly into quadriceps muscle in 6 human patients with Becker muscular dystrophy in human experimental cohort study, result with improvement of muscle hypertrophy and histology.[30] Thus, direct injection of follistatin still seems to show consistent results in rat or human muscle, in contrast to follistatin that naturally circulates in human blood that are much more significant on physical performance than muscle strength or muscle mass.

This study showed a relationship between the change in follistatin (decreased follistatin) levels with acute sarcopenia. The occurrence of sarcopenia might also be related with the workout or exercise routine of the elderly.[17,31] Follistatin levels were increased significantly in rats that were actively given a bicycle and swimming exercises.[6] Woman who had undergone bicycle training showed an increase in follistatin levels.[32] One study reported significant increases in follistatin levels after acute exercise, followed by high-intensity exercise or resistance training.[33]

In this study, the elderly were hospitalized with a lack of physical activity with ADL total dependence was 21.2% and severe dependence was 34.8%. In elderly who were hospitalized, physical activity and length of stay ≥7 days were factors associated with the occurrence of sarcopenia during hospitalization.[3,5,18] In many studies, acute or ongoing physical activity appeared to be associated with follistatin levels, whereas lack of physical activity by elderly with acute sarcopenia appeared to be associated significantly with decreased follistatin levels.[6,32,33]

Follistatin was more sensitive to changes in energy balance such as acute activity or exercise and therefore may be a marker for an acute rather than a chronic muscle loss.[16] Circulating follistatin is increased in the state of exercise and it is associated with increased gluconeogenesis.[34] A review study showed follistatin increased in high-intensity, aerobic and resistance training and could improve glucose uptake in skeletal muscle.[35] The lenght of physical inactivity periods could lead to decreased follistatin levels through decreased muscle contractility, impairment of glucose uptake and decreased gluconeogenesis.[8,11,34,35] Follistatin increase in acute inflammatory conditions and can mimic the acute phase response of the inflammatory response, however, it is still not clear what is the role of follistatin in the inflammatory process.[36] Inflammation can lead to fat accumulation in striated muscle, reducing muscle strength and muscle mass.[37] Follistatin can reduce the production of reactive oxygen species which play a role in the inflammatory process, and therefore migh have a protective effect in acute sarcopenia.[33]

As researchers' knowledge, to date, this current study is the first one to observe the association between changes in follistatin and acute sarcopenia. The cut-off point of decreased follistatin levels found was 4.870, which may be a good starting point for further research. However, further research needs to be done regarding the examination of follistatin and acute sarcopenia by using examination such as BIA or DXA based on the AWGS guidelines or with a CT scan and MRI based on EWGSOP2 guidelines. Longer period of research (last up to 6 months) is also needed for the examination of follistatin and acute sarcopenia involving the physical performance diagnostic criteria of sarcopenia.

Conclusion

Decreased follistatin levels in elderly may indicate a risk of acute sarcopenia in the. Therefore, decreased follistatin

levels could be used as a useful marker of acute sarcopenia, which should be further investigated.

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Authors Contribution

IGPSA designed the study and conceptual framework. IMAW carried out the experiment and wrote the manuscript. All authors discussed the results and commented on the manuscript.

References

1. Chen LK, Woo J, Assantachai P, Auyeung TW, Chou MY, Iijima K, *et al.* Asian Working Group for Sarcopenia: 2019 consensus update on sarcopenia diagnosis and treatment. J Am Med Dir Assoc. 2020; 21(3): 300-307.e2.
2. Cruz-Jentoft AJ, Bahat G, Bauer J, Boirie Y, Bruyere O, Cederholm T, *et al.* Sarcopenia: revised European consensus on definition and diagnosis. Age Ageing. 2019; 48(1): 16-31.
3. Welch C, Greig CA, Masud T, Pinkney T, Jackson TA. Protocol for understanding acute sarcopenia: a cohort study to characterise changes in muscle quantity and physical function in older adults following hospitalisation. BMC Geriatr. 2020; 20: 239. doi: 10.1186/s12877-020-01626-4.
4. Welch C, Greig CA, Hassan-Smith ZK, Pinkney TD, Lord JM, Jackson TA. A pilot observational study measuring acute sarcopenia in older colorectal surgery patients. BMC Res Notes. 2019; 12: 24. doi: 10.1186/s13104-019-4049-y.
5. Martone AM, Bianchi L, Abete P, Bellelli G, Bo M, Cherubini A, *et al.* The incidence of sarcopenia among hospitalized older patients: results from the Glisten study. J Cachexia Sarcopenia Muscle. 2017; 8(6): 907-14.
6. Hansen J, Brandt C, Nielsen AR, Hojman P, Whitham M, Febbraio MA, *et al.* Exercise induces a marked increase in plasma follistatin: evidence that follistatin is a contraction-induced hepatokine. Endocrinology. 2011; 151(1): 164-71.
7. Pedersen BK, Febbraio MA. Muscles, exercise and obesity: skeletal muscle as a secretory organ. Nat. Rev. Endocrinol. 2012; 8(8): 457-65.
8. Han X, Moller LLV, Groote ED, Bojsen-Moller KN, Davey J, Olguin CH, *et al.* Mechanisms involved in follistatin-induced hypertrophy and increased insulin action in skeletal muscle. J Cachexia Sarcopenia Muscle. 2019; 10(6): 1241-57.
9. Sepulveda PV, Lamon S, Hagg A, Thomson RE, Winbanks CE, Qian H, *et al.* Evaluation of follistatin as a therapeutic in models of skeletal muscle atrophy associated with denervation and tenotomy. Sci Rep. 2015; 5: 17535. doi: 10.1038/srep17535.
10. Yaden BC, Croy JE, Wang Y, Wilson JM, Datta-Mannan A, Shetler P, *et al.* Follistatin: a novel therapeutic for the improvement of muscle regenerations. J Pharmacol Exp Ther. 2014; 349(2): 355-71.
11. Oh KJ, Lee DS, Kim WK, Han BS, Lee SC, Bae KH. Metabolic adaptation in obesity and type II diabetes: myokines, adipokines and hepatokines. Int J Mol Sci. 2017; 18(1): 8. doi: 10.3390/ijms18010008.
12. Fife E, Kostka J, Kroc L, Guligowska A, Pigłowska M, Soltysik B, *et al.* Relationship of muscle function to circulating myostatin, follistatin and GDF11 in older women and men. BMC Geriatr. 2018; 18: 200. doi: 10.1186/s12877-018-0888-y.
13. Hofmann M, Halper B, Oesen S, Franzke B, Stuparits P, Tschan H, *et al.* Serum concentrations of insulin-like growth factor-1, members of the TGF-beta superfamily and follistatin do not reflect different stages of dynapenia and sarcopenia in elderly women. Exp Gerontol. 2015; 64: 35-45.
14. Liaw FY, Kao TW, Fang WH, Han DS, Chi YC, Yang WS. Increased follistatin associated with decreased gait speed among old adults. Eur J Clin Invest. 2016; 46(4): 321-7.
15. Margutti KMM, Schuch NJ, Schwanke CHA. Inflammatory markers, sarcopenia and its diagnostic criteria among the elderly: a systematic review. Rev Bras Geriatr Gerontol. 2017; 20(3): 441-53.
16. Wall BT, Dirks ML, Snijders T, Senden JMG, Dolmans J, van Loon LJC. Substantial skeletal muscle loss occurs during only 5 days of disuse. Acta Physiol. 2014; 210(3): 600-11.
17. Welch C, Hassan-Smith ZK, Greig CA, Lord JM, Jackson TA. Acute sarcopenia secondary to hospitalisation –an emerging condition affecting older adults. Aging Dis. 2018; 9(1): 151-64.
18. Sousa AS, Guerra RS, Fonseca I, Pichel F, Amaral TF. Sarcopenia and length of hospital stay. Eur J Clin Nutr. 2015; 70(5): 595-601.
19. Wu YH, Hwang AC, Liu LK, Peng LN, Chen LK. Sex differences of sarcopenia in Asian populations: the implications in diagnosis and management. J Clin Gerontol Geriatr. 2016; 7(2): 37-43.
20. Rinnov AR, Plomgaard P, Pedersen BK, Gluud LL. Impaired follistatin secretion in cirrhosis. J Clin Endocrinol Metab. 2016; 101(9): 3395-400.
21. Setiati S, Mienche, Setyohadi B, Kurniawan J, Laksmi PW, Ariane A, *et al.* Diagnostic performance of calf circumference, thigh circumference, and SARC-F questionnaire to identify sarcopenia in elderly compared to asian working group for sarcopenia's diagnostic standard. Acta Medica Indonesiana. 2019; 51(2): 117-27.
22. Norton K. Standards for anthropometry assessment. In: Norton K, Eston R, editors. Kinanthropometry and Exercise Physiology. 4th ed. South Australia: Routledge; 2020. p.68-137.
23. Spiegeleer AD, Kahya H, Sanchez-Rodriguez D, Piotrowicz K, Surquin M, Marco E, *et al.* Acute sarcopenia changes following hospitalization: influence of pre-admission care dependency level. Age Ageing. 2021; 50(6): 2140-6.
24. LeBanc A, Gogia P, Schneider V, Krebs J, Schonfeld E, Evans H. Calf muscle area and strength changes after five weeks of horizontal bed rest. Am J Sports Med. 1988; 16(6): 624-9.
25. Santos LP, Gonzalez MC, Orlandi SP, Bielemann RM, Barbosa-Silva TG, Heymsfield SB, *et al.* New prediction equations to estimate appendicular skeletal muscle mass using calf circumference: results from NHANES 1999–2006. J Parenter Enteral Nutr. 2019; 43(1): 1-10. doi: 10.1002/jpen.1605.
26. Kawakami R, Miyachi M, Sawada SS, Torii S, Midorikawa T, Tanisawa K, *et al.* Cut-offs for calf circumference as a screening tool for low muscle mass: WASEDA'S Health Study. Geriatr Gerontol Int. 2020; 20(10): 943-950.

Print ISSN: 2527-4384, Online ISSN: 2527-3442
DOI: 10.21705/mcbs.v7i3.359

27. Kementerian Kesehatan Republik Indonesia. Peraturan Menteri Kesehatan Nomor 25 Tahun 2016 tentang Rencana Aksi Nasional Kesehatan Lanjut Usia Tahun 2016-2019. Jakarta: Kementerian Kesehatan Republik Indonesia; 2019.
28. Du Y, Xu C, Shi H, Jiang X, Tang W, Wu X, *et al.* Serum concentrations of oxytocin, DHEA and follistatin are associated with osteoporosis or sarcopenia in community-dwelling postmenopausal women. BMC Geriatr. 2021; 542: 21. doi: 10.1186/s12877-021-02481-7.
29. Sordi CM, Reis-Neto ET, Keppeke GD, Shinjo SK, Sato EI. Serum myostatin and follistatin levels in patients with dermatomyositis and polymyositis. J Clin Rheumatol. 2022; 28(1): 33-7.
30. Mendell JR, Sahenk Z, Malik V, Gomez AM, Flanigan KM, Lowes LP, *et al.* A phase 1/2a follistatin gene therapy trial for becker muscular dystrophy. Mol Ther. 2015; 23(1): 192-201.
31. Aryana IGPS, Setiati S, Rini SS. Molecular mechanism of acute sarcopenia in elderly patient with COVID–19. Acta Med Indones. 2021; 53(4): 481-92.
32. Jurimae J, Vaiksaar S, Purge P, Tillmann V. Irisin, fibroplast growth factor-21, and follistatin responses to endurance rowing training session in female rowers. Front Physiol. 2021; 12: 689696. doi: 10.3389/fphys.2021.689696.
33. Domin R, Dadej D, Pytka M, Zybek-Kocik A, Ruchala M, Guzik P. Effect of various exercise regimens on selected exercise-induced cytokines in healthy people. Int J Environ Res Public Health. 2021; 18(3):1261. doi: 10.3390/ijerph18031261.
34. Hansen JS, Rutti S, Arous C, Clemmesen JO, Secher NH, Drescher A, *et al.* Circulating follistatin is liver-derived and regulated by the glucagon-to-insulin ratio. J Clin Endocrinol Metab. 2016; 101(2): 550-60.
35. Gonzalez-Gil AM, Elizondo-Montemayor L. The role of exercise in the interplay between myokines, hepatokines, osteokines, adipokines, and modulation of inflammation for energy substrate redistribution and fat mass loss: A review. Nutrients. 2020; 12(6): 1899. doi: 10.3390/nu12061899.
36. Hansen JS, Plomgaard P. Circulating follistatin in relation to energy metabolism. Mol Cell Endocrinol. 2016; 433: 87-93. doi: 10.1016/j.mce.2016.06.002.
37. Aryana IGPS, Hapsari AAAR, Kuswardhani RAT. Myokine regulation as marker of sarcopenia in elderly. Mol Cell Biomed Sci. 2018; 2(2): 38-47.