



Alteration in skeletal muscle mass in women with subclinical hypercortisolism

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Abstract

Purpose Despite the well-known deleterious effects of cortisol on skeletal muscle, whether subtle cortisol excess in subclinical hypercortisolism (SH) affects skeletal muscle mass is unknown. Our objective was to understand the effects of the cortisol level on skeletal muscle mass in patients with SH.

Methods We compared skeletal muscle mass and fat mass (FM) between 21 patients with SH (12 women and 9 men) and 224 controls (67 women and 157 men) with nonfunctioning adrenal incidentaloma (NFAI). Medical records were reviewed, and we measured body composition parameters using bioelectrical impedance analysis and serum cortisol levels after the overnight 1-mg dexamethasone suppression test (DST).

Results After adjusting for confounding factors, 1-mg DST levels were inversely correlated with appendicular skeletal muscle mass (ASM) ($\gamma = -0.245$, $P = 0.040$), lower limb ASM ($\gamma = -0.244$, $P = 0.040$), and appendicular skeletal muscle index (ASMI; height-adjusted ASM) ($\gamma = -0.229$, $P = 0.048$) in all women, but not men. ASM and ASMI were significantly lower by 6.2% ($P = 0.033$) and 5.9% ($P = 0.046$), respectively, in women with SH compared with those with NFAI, but not men. Conversely, FM and percent fat mass were similar between the two groups. Compared with women with NFAI, among those with SH, lower limb, but not upper limb, ASM was lower by 6.8% ($P = 0.020$).

Conclusions This study showed that women with SH had lower skeletal muscle mass, especially of the lower limb, and suggested that subtle cortisol excess also has adverse effects on skeletal muscle metabolism.

Keywords Subclinical Hypercortisolism · Cortisol · Skeletal Muscle · Sarcopenia

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Introduction

Sarcopenia is characterized by loss of skeletal muscle mass as well as diminished muscle strength and/or physical performance [1]. Patients with sarcopenia exhibit reduced physical capability, impaired cardiopulmonary performance, and greater morbidity and mortality [2]. Its prevalence is exponentially increasing with the elderly

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population because aging is one of its most common causes [3]. Data from a national survey in Korea showed that the prevalence of sarcopenia is 12.1% in men and 11.9% in women [4]. Thus, sarcopenia has become an increasingly serious medical and economic problem.

With the advances in imaging technologies, adrenal incidentaloma (AI), an unexpected adrenal mass discovered during a radiologic examination performed for reasons unrelated to adrenal diseases, has become a common clinical problem [5]. The majority of those tumors are non-functional AI (NFAI) without clinically relevant hormonal hyperfunction. However, subclinical hypercortisolism (SH), defined as a mild alteration of cortisol secretion without clinical signs or symptoms of Cushing's syndrome (CS) such as muscle weakness and skin fragility, is a common finding in patients with AI [5, 6]. A recent review analyzing the results of studies published within the last 15 years showed that patients with SH have an increased rate of comorbidities such as hypertension, impaired glucose metabolism, and increased visceral fat [6]. Furthermore, it also showed that patients with SH have a higher incidence of cardiovascular events and related mortality [6].

Hypercortisolism resulting in muscle weakness is one of the characteristic clinical features of overt CS which has been studied so far in small series of patients using electromyography, histology, imaging, and functional testing [7–10]. Muscle weakness has been reported to be present in 40–70% of patients with CS [11, 12] and it appears to be more pronounced in women [13]. The most commonly impaired part of the body is the proximal musculature of the lower limbs [14]. Cortisol has profound effects on muscle through 11 β -hydroxysteroid dehydrogenase type 1 (11 β -HSD1) to blunt muscle protein synthesis, activate muscle proteolysis, impair mitochondrial function, and decrease sarcolemmal excitability [13–15]. Despite the well-established concept that hypercortisolism in patients with overt CS can lead to muscle weakness, to our knowledge, there is no report on the effects of subtle cortisol excess on skeletal muscle in patients with SH. Here we examined the associations of subtle cortisol excess with skeletal muscle mass in patients with SH compared with those in patients with NFAI.

Methods

Study participants and protocol

The study population was based on the Asan Medical Center (AMC) and Samsung Medical Center (SMC) cohort, which is a subset of the “Co-work of Adrenal Research” study (clinicaltrial.gov no. NCT01382420), a randomized, parallel-group, multicenter, open-label trial conducted at

three medical centers in Korea [16]. From July 2011 to June 2014, we recruited 985 consecutive patients with newly diagnosed AI from two medical centers: AMC ($n = 616$) and SMC ($n = 369$). AI diagnosis was based on the detection of an adrenal mass (size ≥ 1 cm) on computed tomography (CT) performed for an unrelated disease. In all patients, we measured the levels of basal morning (0900 h) ACTH and cortisol, 24-h urinary-free cortisol (UFC), cortisol level (0900 h) after a 1-mg overnight dexamethasone suppression test (1-mg DST), dehydroepiandrosterone sulfate (DHEA-S), 24-h urinary fractionated metanephrines, plasma aldosterone, and plasma renin activity at baseline. We excluded 249 subjects who were suspected to have adrenal CS ($n = 19$), Cushing's disease ($n = 1$), primary aldosteronism ($n = 60$), pheochromocytoma ($n = 56$), congenital adrenal hyperplasia ($n = 3$), adrenal carcinoma ($n = 10$), adrenal metastasis ($n = 3$), adrenal tuberculosis ($n = 2$), pseudo-Cushing's syndrome ($n = 4$), history of drugs (such as steroid, estrogen, or thyroid hormone) or a disease (such as hyperthyroidism) that might affect muscle mass ($n = 67$), or other benign adrenal mass ($n = 24$). Among the remaining 736 subjects, data on bioelectrical impedance analysis (BIA) from the Health Promotion Center was available for 245 subjects, who were therefore eligible for inclusion in this study. We used the criteria for diagnosing SH as 1-mg DST > 5.0 $\mu\text{g/dL}$ (138 nmol/L) or 1-mg DST > 2.2 $\mu\text{g/dL}$ (61 nmol/L) along with one parameter among low levels of ACTH [< 10 pg/mL (2.2 pmol/L)] and DHEA-S [< 35 $\mu\text{g/dL}$ (0.95 $\mu\text{mol/L}$) in women or < 80 $\mu\text{g/dL}$ (2.17 $\mu\text{mol/L}$) in men], as described previously [16]. We classified 21 patients with SH (12 women and 9 men) and 224 patients with NFAI (67 women and 157 men) (Fig. 1).

The following patient information was obtained using an interviewer-assisted questionnaire: smoking habits (current smoker), alcohol intake (≥ 3 U/d), regular outdoor exercise (≥ 30 min/d), history of medication use, previous medical or surgical procedures, and reproductive status including menstruation.

This study was approved by the Institutional Review Boards of AMC and SMC. Written informed consent was provided by all enrolled participants.

Bioelectrical impedance analysis

Body composition was measured using a direct segmental multi frequency BIA (In-Body720; Biospace Co., Ltd, Seoul, Korea). In both the AMC and SMC, the In-Body 720 automatically estimates weight, body mass index (BMI), FM (kg), percent FM [pFM (%)], and skeletal muscle mass of the arms and legs. Appendicular skeletal muscle mass (ASM) was calculated as the sum of skeletal muscle mass in the arms and legs. Upper limb ASM (UL-ASM) was calculated as the sum of the skeletal muscle mass values in

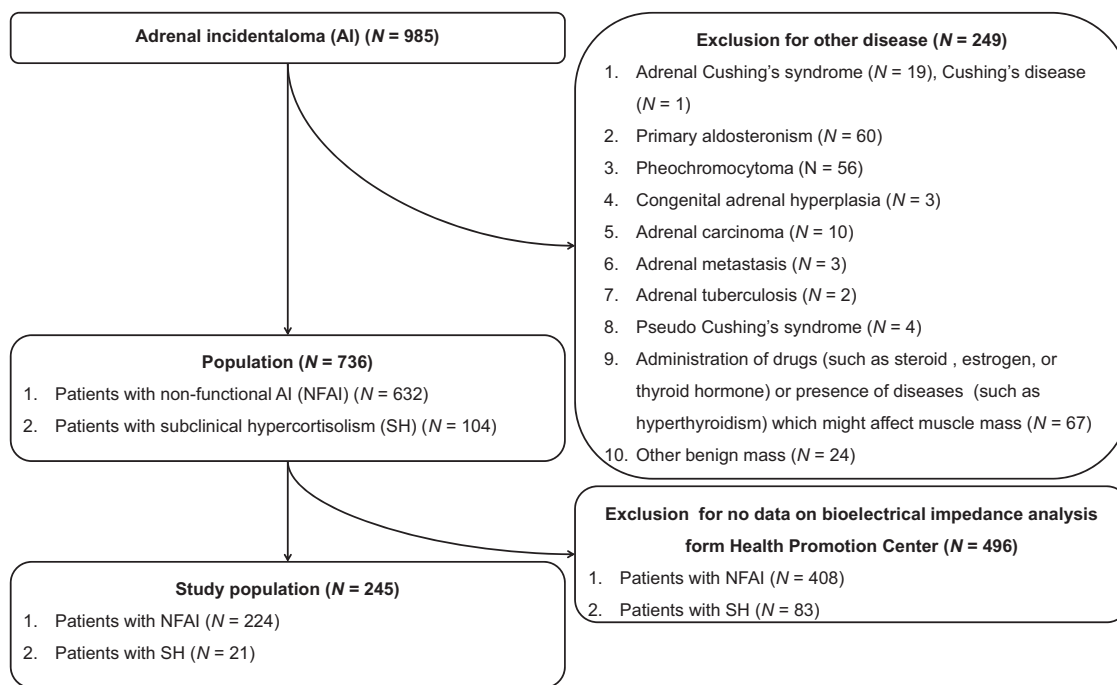


Fig. 1 Flow diagram of study population

both arms, and lower limb ASM (LL-ASM) was calculated as the sum of the skeletal muscle mass values in both legs. Appendicular skeletal muscle index (ASMI), height-adjusted ASM [HA-ASM (kg/m^2)] was defined as ASM divided by body height in meters squared ($\text{ASM}/\text{height}^2$).

All those who visited the Health Promotion Center were subjects of this study. We performed the BIA in the morning after an overnight fast. Before the examination, all subjects were instructed to discharge urine and feces and remain stable without any other exercise or dynamic work. In addition, all subjects wore the same clothes and remained at room temperature of 20–25 °C. During the BIA test, we asked subjects to assume the straight posture. The measurement comprised 30 impedances evaluated in six frequency bands (1, 5, 50, 250, 500, and 1000 kHz) for each of the five parts (right arm, left arm, torso, right leg, and left leg). Furthermore, the amplitude was 90 μA for 1 kHz and 400 μA for others per the manufacturer's instructions.

Hormonal and biochemical measurements

All measurements of hormonal and biochemical parameters were performed at each of the two medical centers. For the hormonal assay, the plasma ACTH levels were measured by immunoradiometric assay using an ELSA-ACTH kit (Cisbio bioassay; Codolet, France) on a Cobra II γ -counter (Packard Instrument Company, Meriden, CT) at the AMC and on a RALS Gamma Counter GAMMA-10 (Shin Jin Medics Inc., Seoul, Republic of Korea) at the SMC. Serum cortisol and

UFC levels (after dichloromethane extraction) were measured by RIA using the Coat-A-Count® cortisol kit (Siemens Healthcare Diagnostics, Los Angeles, CA) on a Cobra II γ -counter (Packard Instrument) at the AMC and by RIA using a cortisol RIA kit (Beckman Coulter, Prague, Czech Republic) on a RALS GAMMA-10 γ -counter (Shin Jin Medics) at the SMC. The DHEA-S level was measured by RIA using the Coat-A-Count® DHEA- SO_4 kit (Siemens Healthcare Diagnostics) on a Cobra II γ -counter (Packard Instrument) at the AMC and by RIA using a DHEA-S RIA CT kit (Asbach Medical Products, Obrigheim, Germany) on a RALS GAMMA-10 γ -counter (Shin Jin Medics) at the SMC. We performed the 1-mg DST twice and used the mean value for analysis in those with a value of $>1.8 \mu\text{g}/\text{dL}$ (50 nmol/L) [5, 16]. Differences in serum cortisol assays among the centers were monitored on a quarterly basis by an external quality control program of the Korean Society of Nuclear Medicine using standard specimens with low, middle, and high cortisol levels (<http://qc.ksnmt.or.kr/ria>). All the centers satisfied the criteria for comparability with an absolute Z-score value of <2 . The intra- and interassay coefficients of variation for all assays were ~5% and 10%, respectively.

Statistical analysis

Owing to the known sex differences in muscle weakness of patients with CS, analyses were performed separately among men and women. Data are presented as mean \pm SD, median (interquartile range), or as numbers (percentages)

unless otherwise specified. Baseline characteristics were compared using a Student’s *t*-test or Mann–Whitney *U*-test for continuous variables or the χ^2 -test for categorical variables. The association of 1-mg DST with ASM, UL-ASM, LL-ASM, ASMI, FM, and pFM was investigated through Spearman’s test after adjustment for potential confounders, including age, sex, BMI, menopausal status (for women), current smoking, alcohol use, and regular outdoor exercise. In these analyses, 1-mg DST values were log-transformed because of their skewed distribution. Multivariable-adjusted least-square mean levels (95% confidence intervals) of LM, ASM, ASMI, FM, pFM, UL-ASM, and LL-ASM according to absence or presence of SH were estimated and compared using analysis of covariance after adjustment for confounders. All statistical analyses were performed using SPSS statistical software (SPSS Inc., Chicago, IL). *P* < 0.05 was considered statistically significant.

Results

The baseline characteristics of the 245 study subjects (79 women and 166 men) are listed in Table 1. In men, multiple lesions were more common (*P* = 0.035) and the lesions

were larger (*P* = 0.024) in the SH group than in the NFAI group. In both women and men, ACTH levels (*P* < 0.001 for both women and men) and DHEA-S (*P* = 0.001 for both women and men) were significantly lower in the SH group than in the NFAI group. The 1-mg DST values were significantly higher in the SH group than in the NFAI group (*P* < 0.001 for both women and men). The other variables were comparable between the two groups.

In women, the 1-mg DST levels were inversely correlated with LM ($\gamma = -0.271, P = 0.017$), ASM ($\gamma = -0.277, P = 0.015$), LL-ASM ($\gamma = -0.281, P = 0.013$), and ASMI ($\gamma = -0.230, P = 0.044$) (Table 2). Higher 1-mg DST levels in women had tendency of lower UL-ASM ($\gamma = -0.220, P = 0.054$) (Table 2). Further adjustments in addition to medical center, age, BMI, menopausal state (for women), current smoking, alcohol use, and regular outdoor exercise showed similar results. In both sexes, there was no significant correlation of ACTH, UFC, and DHEA-S levels with body composition parameters after adjustment for confounders (data not shown). In contrast, 1-mg DST levels in men were not significantly correlated with body composition parameters (Table 2).

After adjustment for confounders, women with SH (*n* = 12) had significantly lower LM (by 6.2%, *P* = 0.048),

Table 1 Baseline characteristics of subjects (*n*=245)

	Women (<i>n</i> = 79)			Men (<i>n</i> = 166)		
	NFAI (<i>n</i> = 67)	SH (<i>n</i> = 12)	<i>P</i>	NFAI (<i>n</i> = 157)	SH (<i>n</i> = 9)	<i>P</i>
Age (y), mean ± SD	53.3 ± 7.1	54.4 ± 10.8	0.728	54.7 ± 7.6	56.1 ± 10.0	0.606
Height (cm), mean ± SD	158.9 ± 4.9	156.6 ± 4.3	0.126	169.7 ± 6.7	171.8 ± 6.0	0.350
Weight (kg), median [IQR]	60.0 [56.1; 67.5]	55.0 [51.7; 62.4]	0.071	74.3 [68.2; 80.1]	78.7 [70.0; 84.7]	0.454
BMI (kg/m ²), median [IQR]	24.0 [22.0; 26.0]	22.9 [20.0; 26.2]	0.181	26.0 [24.0; 28.0]	26.6 [24.0; 28.0]	0.813
Menopause, <i>n</i> (%)	49 (73.1%)	9 (75.0%)	>0.999			
Exercise (>30 min), <i>n</i> (%)	20 (29.9%)	6 (50.0%)	0.336	55 (35.0%)	2 (22.2%)	0.642
Current smoker, <i>n</i> (%)	3 (4.5%)	0 (0.0%)	>0.999	55 (35.0%)	5 (55.6%)	0.399
Alcohol, <i>n</i> (%)	2 (3.0%)	0 (0.0%)	>0.999	12 (7.6%)	1 (11.1%)	>0.999
Site (%)			0.676			0.123
Bilateral, <i>n</i> (%)	4 (6.0%)	0 (0.0%)		18 (11.5%)	3 (33.3%)	
Left, <i>n</i> (%)	35 (52.2%)	7 (58.3%)		89 (56.7%)	5 (55.6%)	
Multiple lesions, <i>n</i> (%)	6 (9.0%)	0 (0.0%)	0.610	20 (12.7%)	4 (44.4%)	0.035
Size (cm), median [IQR]	1.5 [1.1; 2.0]	1.8 [1.4; 2.0]	0.208	1.5 [1.2; 1.9]	1.7 [1.5; 2.7]	0.024
ACTH (pg/mL), median [IQR]	25.8 [15.7; 40.1]	15.0 [10.8; 23.8]	0.001	34.0 [21.4; 48.3]	8.5 [6.9; 19.3]	<0.001
Cortisol (µg/dL), median [IQR]	10.9 [8.9; 13.8]	11.1 [7.5; 13.1]	0.628	12.7 [9.7; 14.9]	11.4 [8.3; 14.2]	0.308
DHEA-S (µg/dL), median [IQR]	65.8 [40.7; 93.8]	22.9 [18.0; 36.4]	<0.001	104.5 [80.5; 154.8]	43.3 [36.0; 75.0]	0.001
UFC (µg/d), median [IQR]	44.0 [30.5; 55.9]	62.0 [31.1; 82.6]	0.238	47.9 [34.3; 71.0]	36.0 [32.7; 59.6]	0.266
1-mg DST (µg/dL), median [IQR]	1.1 [1.0; 1.6]	4.2 [3.3; 5.0]	<0.001	1.0 [1.0; 1.4]	3.4 [3.2; 4.4]	<0.001

BMI body mass index, *DHEA-S* dehydroepiandrosterone–sulfate, *IQR* interquartile range, *NFAI* nonfunctioning adrenal incidentaloma, *SH* subclinical hypercortisolism, *UFC* urine-free cortisol, *1-mg DST* cortisol levels after an overnight 1-mg dexamethasone suppression test

Significant results (*P* < 0.05) are in bold

Table 2 Correlation of cortisol levels after overnight 1-mg DST with LM, ASM, UL-ASM, LL-ASM, ASMI, FM, and pFM

Variables	γ	P^a	γ	P^b	γ	P^c
Women ($n=79$)						
LM (kg)	-0.271	0.017	-0.258	0.026	-0.233	0.051
ASM (kg)	-0.27	0.015	-0.257	0.026	-0.245	0.040
UL-ASM (kg)	-0.220	0.054	-0.220	0.058	-0.204	0.089
LL-ASM (kg)	-0.281	0.013	-0.257	0.026	-0.244	0.040
ASMI (kg/m ²)	-0.230	0.044	-0.233	0.044	-0.229	0.048
FM (kg)	-0.063	0.586	-0.069	0.557	0.159	0.185
pFM (%)	0.007	0.952	-0.007	0.954	0.159	0.185
Men ($n=166$)						
LM (kg)	0.029	0.712	0.059	0.455	0.074	0.354
ASM (kg)	0.041	0.606	0.069	0.382	0.069	0.386
UL-ASM (kg)	0.053	0.505	0.103	0.194	0.082	0.303
LL-ASM (kg)	0.032	0.681	0.051	0.522	0.059	0.458
ASMI (kg/m ²)	-0.006	0.942	0.037	0.643	0.011	0.894
FM (kg)	-0.028	0.724	0.043	0.587	0.069	0.387
pFM (%)	-0.039	0.621	0.009	0.909	0.011	0.892

The 1-mg DST values were log-transformed because of their skewed distributions. ASMI is a height-adjusted ASM [HA-ASM (kg/m²)] that is defined as ASM divided by body height in meters squared (ASM/height²). Significant results ($P < 0.05$) are in bold

ASM appendicular skeletal muscle mass, ASMI appendicular skeletal muscle index, FM fat mass, LM lean mass, LL-ASM lower limb ASM, pFM percent FM, UL-ASM upper limb ASM, 1-mg DST cortisol level after overnight 1-mg dexamethasone suppression test

^a P values determined by Spearman's test with respect to the log-transformed 1-mg DST adjusted for medical center

^b P values determined by Spearman's test with respect to the log-transformed 1-mg DST adjusted for medical center, age, menopausal status (for women), and body mass index

^c P values determined by Spearman's test with respect to the log-transformed 1-mg DST adjusted for medical center, age, menopausal status (for women), body mass index, current smoking, alcohol use, and regular outdoor exercise

ASM (by 6.2%, $P = 0.033$), and ASMI (by 5.9%, $P = 0.046$) than those with NFAI ($n = 67$) (Fig. 2). However, no statistical difference was observed in FM and pFM between the groups. In contrast, after adjustment for confounders, no statistical difference was observed in body composition between men with SH ($n = 9$) and those with NFAI ($n = 157$) (Fig. 3).

Finally, we assessed the differences in UL-ASM and LL-ASM according to the presence of SH (Fig. 4). After adjustment for confounders, women with SH had significantly lower LL-ASM (by 6.8%, $P = 0.020$), but not UL-ASM ($P = 0.158$) than those with NFAI (Fig. 4). In contrast, no statistical difference was observed in either UL-ASM or LL-ASM between men with SH and those with NFAI (Fig. 4).

Discussion

This study showed that women, but not men, with SH had markedly lower skeletal muscle mass than those with NFAI.

Consistently, 1-mg DST levels in women were inversely correlated with ASM and ASMI. In particular, the magnitudes of these inverse correlations of 1-mg DST in women were larger in the lower limb than in the upper limb. These differences in the magnitude of the effects of 1-mg DST could be confirmed by the finding that women with SH had lower LL-ASM, but not UL-ASM, than those with NFAI. Thus, we obtained the following two novel findings: (1) subtle cortisol excess could be associated with a lower skeletal muscle mass and (2) the negative correlations of subtle cortisol excess with skeletal muscle mass were observed only in women and were more pronounced in lower limbs than upper limbs.

Despite a well-established deleterious effect of overt cortisol excess on skeletal muscle in patients with overt CS, SH by definition is not associated with proximal muscle weakness [5, 6, 17]. Therefore, much of the research has focused on the comorbidities (hypertension, impaired glucose metabolism, and increased visceral fat) and clinically relevant outcomes (osteoporotic fractures, cardiovascular diseases, and mortality) associated with SH except for

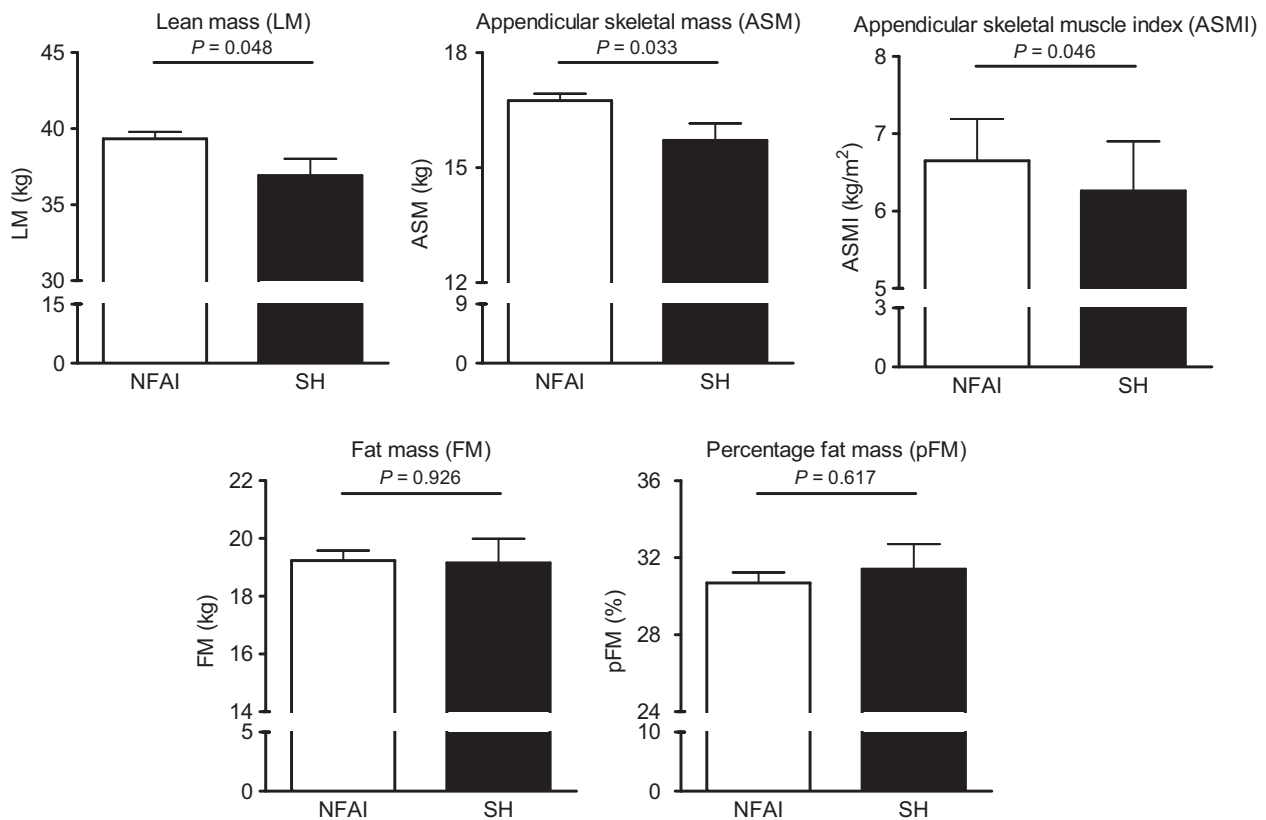


Fig. 2 Lean mass (LM), appendicular skeletal mass (ASM), appendicular skeletal muscle index (ASMI), fat mass (FM), and percent fat mass (pFM) in the women in the nonfunctional adrenal incidentaloma (NFAI) and subclinical hypercortisolism (SH) groups. Data are the

means \pm SEM, which were calculated after adjusting for medical center, age, body mass index, menopausal state, current smoking, alcohol use (alcohol intake ≥ 3 U/d), and regular outdoor exercise (exercise ≥ 30 min/d)

muscle weakness. The inverse correlation of 1-mg DST with ASM or ASMI might indicate that subtle cortisol excess per se seems to be an important determining factor. This finding is in accordance with other reports that a biological continuum with no clear separation between non-functioning adenomas and functioning adenomas is associated with some degree of cortisol excess [5, 6]. Therefore, recent guidelines also suggested interpreting the 1-mg DST level as a continuous rather than categorical variable [5]. To the best of our knowledge, these findings provide the first clinical evidence that even subtle cortisol excess in SH may contribute to the loss of skeletal muscle including ASM and ASMI, especially in women. While we were preparing this paper, another report published about impact of hypercortisolism on total abdominal muscle mass using abdominal CT [18]. In that study, total abdominal muscle mass was not decreased in patients with mild autonomous cortisol excess compared to patients with NFAI. But in our study, ASM and ASMI, which are used as an approximation of muscle mass in diagnostic criteria for sarcopenia [1], were significantly decreased in patients with mild cortisol excess compared to patients with NFAI. In addition, previous studies reported that increased mean

levels of cortisol in older adults [19] and upregulation of skeletal muscle 11 β -HSD1 with age in women [19]. Considering our present results, future studies are needed to evaluate the clinical usefulness of screening women with SH for lower skeletal muscle mass and of considering lower skeletal muscle mass as one of the factors indicating a need for adrenal surgery.

Another interesting feature of our study is that the deleterious effect of subtle cortisol excess on skeletal muscle mass showed sexual dimorphism and a site-specific difference between upper and lower limbs. Although this is a novel finding, it is consistent with clinical features of muscle weakness in CS such as a predominance of women [13] and lower limbs [14] being affected. Sexual dimorphism is of particular interest because sarcopenia was closely associated with menopause [20]. Therefore, it has been suggested that declining sex steroid (androgens and estrogens) to cortisol ratios could underpin the emergence of sarcopenia after the menopause [20]. We cannot determine the exact reason for sexual dimorphism in our study, although upregulation of skeletal muscle 11 β -HSD1, a local tissue amplifier of cortisol effects, was observed in older women, but not men [14]. The mechanism of the typical

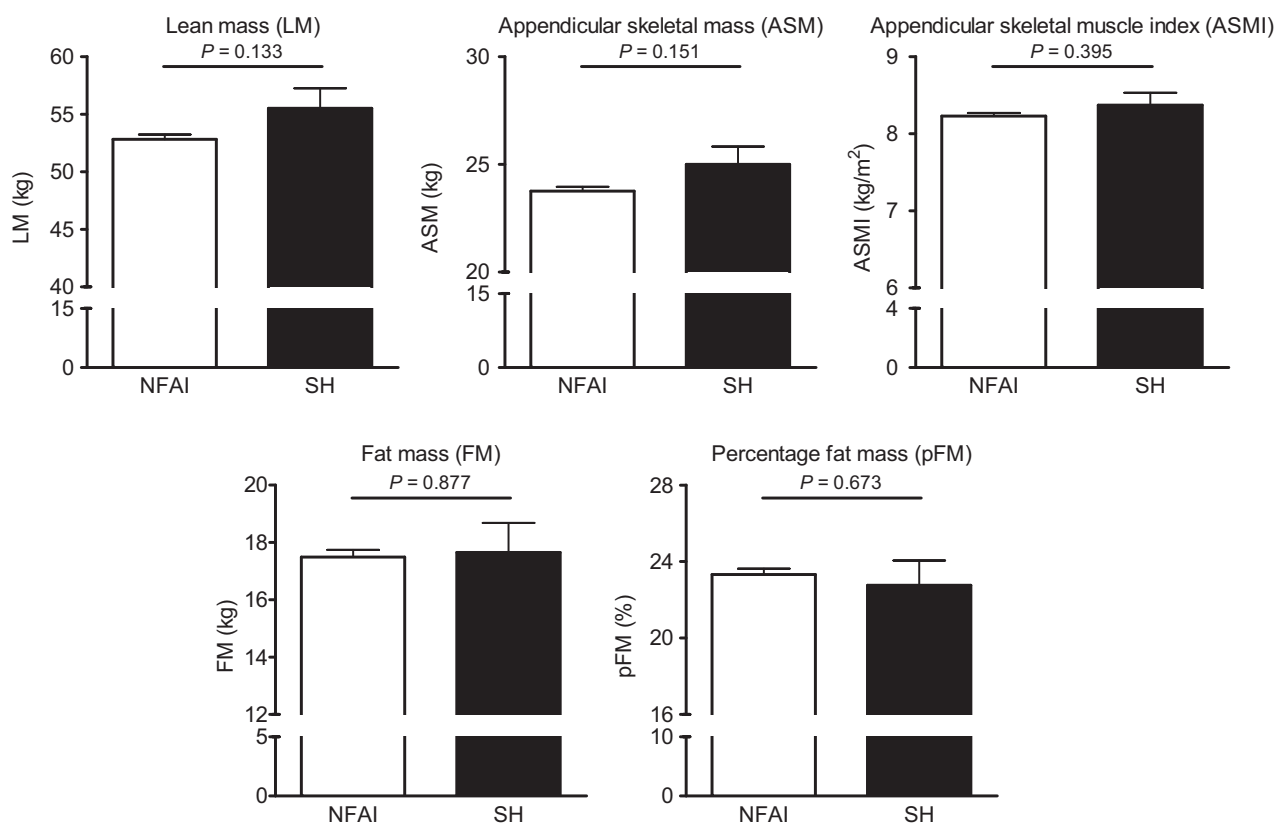


Fig. 3 Lean mass (LM), appendicular skeletal mass (ASM), appendicular skeletal muscle index (ASMI), fat mass (FM), and percent fat mass (pFM) in the men in the nonfunctional adrenal incidentaloma (NFAI) and subclinical hypercortisolism (SH) groups. Data are the

means \pm SEM, which were calculated after adjusting for medical center, age, body mass index, current smoking, alcohol use (alcohol intake \geq 3 U/d), and regular outdoor exercise (exercise \geq 30 min/d)

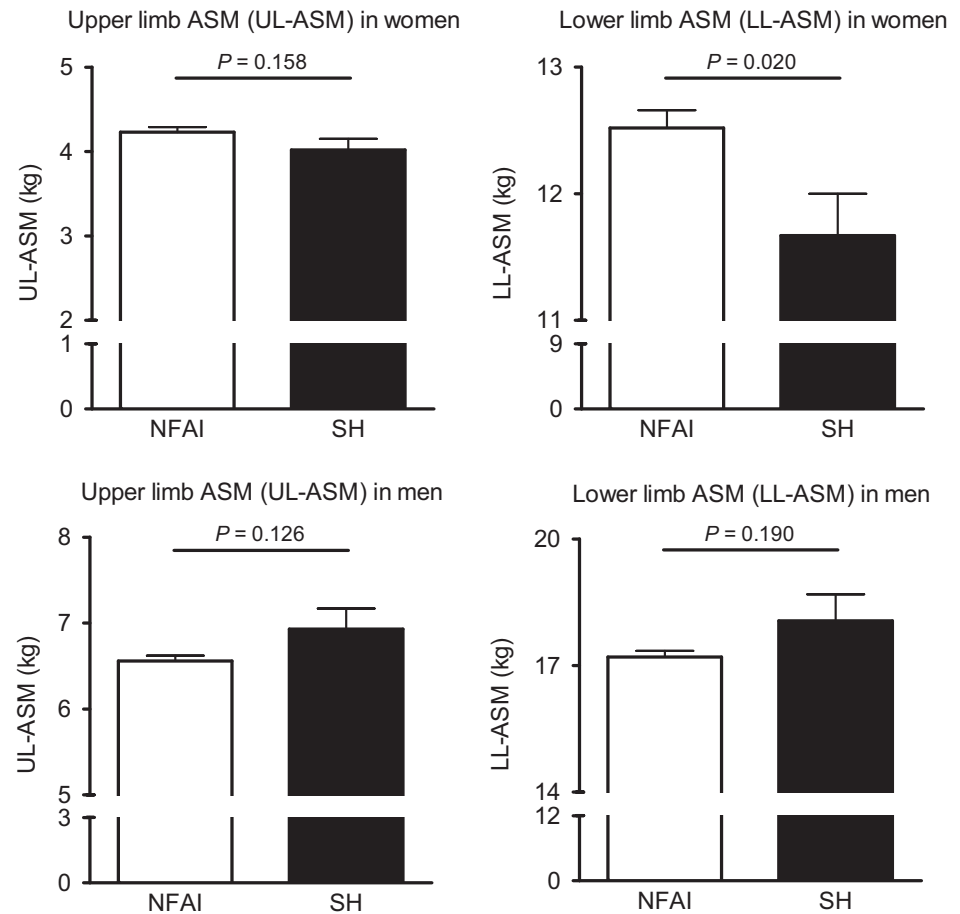
pattern of muscle weakness caused by cortisol affecting the lower limbs more than the upper ones has been an unsolved issue [14].

In this study, we used the BIA for evaluating the muscle mass. Because the BIA necessitates the assumption of normal hydration of the fat-free mass [1, 21, 22], its sensitivity (and responsiveness to body composition changes) in Cushing's syndrome (and/or other conditions related to changes in the hydration of the fat-free mass) seems poor. Likewise, one study reported no differences in the BIA-derived estimation of muscle mass between patients with Cushing's syndrome who exhibited the loss of muscle strength and performance and body mass index-matched obese controls [23], despite another study demonstrating the lower abdominal muscle mass in patients with Cushing's syndrome than patients with NFAI using CT, presently considered the gold standard for the assessment of body composition [18]. Regarding the assessment of the hydration status, we verified the ratio of the extracellular fluid to the total body water (ECF/TBW) [24]. In all subjects, ECF/TBW ratio was within the reference range (0.36–0.39) and similar between subjects with SH and those with NFAI (0.38 ± 0.00 vs. 0.38 ± 0.01 , $P = 0.912$). These findings

implied that the BIA was acquired under the normal hydration status.

We observed no significant gender differences in the frequency of AI in the pooled autopsy data of 71,206 cases [25]. In this study, AIs were more frequent in men (67.7%, 166 of 245 patients with AI). Although a Study Group on Adrenal Tumors of the Italian Society of Endocrinology reported that AIs are slightly more frequent in women (58.0%, 584 of 1004) [26], another Korean study revealed that AIs are slightly more frequent in men (54.9%, 631 of 1149) similar to the results of our study [27]. Although the exact reason is unclear, this phenomenon is attributed to referral bias. Furthermore, despite the significant incidence of AI in men (67.7%, 166 of 245), the proportion of SH was significantly lower in men (5.4%, 9 of 166) compared with women (15.2%, 12 of 79; $P = 0.021$). Although little data are available about the proportion of gender in patients with SH, one study reported no significant gender differences in the frequency of SH between men (28.8%, 32 of 111) and women (30.1%, 53 of 176; $P = 0.864$) despite the large number of AI in women (61.3%, 176 of 248) [28]. Finally, the prevalence of SH (9.4%, 21 of 245) in this study was similar with those (7.1%, 82 of 1149) in another

Fig. 4 Upper limb (UL) appendicular skeletal mass (ASM), lower limb (LL) ASM according to the two groups such as nonfunctional adrenal incidentaloma (NFAI) and subclinical hypercortisolism (SH) in women and men, respectively. Data are the means \pm SEM, which were calculated after adjusting for medical center, age, body mass index, menopausal state, current smoking, alcohol use (alcohol intake \geq 3 U/d), and regular outdoor exercise (exercise \geq 30 min/d)



study in Korea [27]; however, it was lower than those (median, 12%; range: 1.0–29%) in the guideline of the European Society of Endocrinology in collaboration with the European Network for the Study of Adrenal Tumors [5]. Although the exact reason remains unclear, perhaps, the used criteria for diagnosing SH and ethnic differences between Caucasian and East Asian populations according to ethnicity-associated polymorphisms of the glucocorticoid receptor [29] and the 11β -HSD1 enzyme [30, 31] might affect these results.

The major strengths of this study were that we enrolled consecutive patients with newly diagnosed AI to minimize selection bias and used the diagnostic criteria for SH to predict the occurrence of both postsurgical hypocortisolism, indicating a pre-surgical inappropriate cortisol elevation, and chronic complications (components of the metabolic syndrome and low bone mass) [16]. Despite these strengths, several potential limitations should be considered while interpreting our results. First, we undertook our study with Korean population including the small number of 21 patients with SH. Thus, further extensive studies comprising a significant number of other ethnic groups are needed for the replication of our results. Second, the precision of BIA

in measuring muscle mass is controversial [21]. The accuracy of the BIA is highly dependent on the conditions of assessments, for example, body position, temperature, and hydration status [1, 22]. However, the BIA is inexpensive, easy to use, and readily reproducible, and its accuracy has been validated in the sarcopenia diagnosis [1, 22]; the Asian Working Group for Sarcopenia therefore considers BIA to be acceptable for muscle measurements. Third, we did not measure muscle strength and physical performance. In the diagnosis of sarcopenia, the confirmation of the reduction in the muscle mass and muscle function (defined by muscle strength or physical performance) is imperative [1, 22]. Owing to the absence of an approach to the muscle strength and physical performance, we could only present the lower skeletal muscle mass in patients with SH. Thus, further studies need to include the approach to the muscle strength and physical performance to ascertain the risk of sarcopenia in patients with SH.

In summary, women with SH had a lower skeletal muscle mass, especially in the lower limbs, than those with NFAI. These results suggest that SH can be useful as a clinical model for further research in sarcopenia to study the metabolism and physiology of muscle.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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