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Clinical Research Article

Impaired Muscle Parameters in Individuals With Premature Ovarian Insufficiency: A Pilot Study

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Abstract

Context: Although bone loss is a recognized consequence of premature ovarian insufficiency (POI), the impact on skeletal muscle health is less well-defined.

Objective: To compare muscle mass and function parameters between women with POI and controls.

Methods: Cross-sectional study from a tertiary health network and community between 2017 and 2023. Participants were women aged 20 to 40 years with POI associated with Turner syndrome (TS; n = 11) and spontaneous normal karyotype POI (s-POI; n = 7) compared with age- and body mass index (BMI)-matched controls (n = 45).

Results: All women with POI (mean age 28.70 ± 5.58) were using hormone therapy. Appendicular lean mass (ALM)/total fat mass and ALM/ BMI was lower in the POI group. Height-adjusted muscle mass parameters did not differ between groups. Compared with controls, women with TS and s-POI had lower muscle strength (TS 19.72 ± 4.89; s-POI 22.73 ± 5.35; controls 28.67 ± 5.65 kg; $P < .001$) and muscle quality (TS 11.09 ± 2.06; s-POI 10.89 ± 2.01; controls 14.10 ± 1.99 kg/kg; $P < .001$). Higher C-reactive protein levels, higher depression scores, and lower sex-steroid and physical activity levels were observed in women with POI ($P < .05$). Creatinine/cystatin C ratio, insulin-like growth factor-1, and transthyretin did not differ between groups.

Conclusion: Despite hormone therapy usage, women with POI exhibited compromised muscle parameters compared with age-matched controls. Potential contributory factors were identified. Further research is required to clarify pathophysiology and inform management strategies.

Key Words: premature ovarian insufficiency, muscle, body composition, appendicular lean mass, handgrip strength, Turner syndrome

Abbreviations: ALM, appendicular lean mass; BA, bone area; BDI, Beck Depression Inventory; BMAD, bone mineral apparent density; BMD, bone mineral density; BMI, body mass index; COCP, combined oral contraceptive pill; CRP, C-reactive protein; CSA, muscle cross-sectional area; CTX, C-terminal beta-crosslinked telopeptide of type 1 collagen β; DXA, dual-energy x-ray absorptiometry; FN, femoral neck; GOS, Geelong Osteoporosis Study; HT, hormone therapy; LS, lumbar spine; POI, premature ovarian insufficiency; MET, metabolic equivalent value; MHT, menopausal hormone therapy; P1NP, procollagen type 1 N-terminal propeptide; PAS, physical activity score; pQCT, peripheral quantitative computed tomography; s-POI, spontaneous POI; TFM, total fat mass; TS, Turner syndrome; TSH, thyroid-stimulating hormone.

The average age at natural menopause ranges from 48 to 52 years [1]; premature ovarian insufficiency (POI) can occur spontaneously or due to medical and surgical interventions [2]. Spontaneous POI (s-POI) is most commonly idiopathic but can be associated with an abnormal karyotype. Turner syndrome (TS), associated with the loss of all or part of an X chromosome, is characterized by short stature and POI [3].

POI adversely impacts women physically and psychologically, leading to increased morbidity and mortality. Although accelerated bone loss and increased risk of osteoporosis are well recognized associations with POI [4],

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consistent data regarding muscle health are lacking. A recent systematic review including 6 studies (n = 18 291; mean age of participants 63.8 years) concluded that women with POI had lower muscle strength than controls [5]. A cross-sectional study by Soucek et al involving 60 adolescent girls with TS reported similar muscle force (Fmax) but diminished power (Pmax) relative to healthy controls [6]. Conversely, Milde and colleagues demonstrated that girls with TS aged 10 to 18 years exhibited superior handgrip strength than healthy peers ($P < .001$) [7].

Skeletal muscle has multiple roles in the human body, including enabling movement, maintaining body posture and balance, generating energy, and as a complex endocrine organ, actively releasing a diverse array of myokines [4, 8]. Consequently, disruptions in muscle health can lead to an increased susceptibility to falls and frailty, exacerbating bone density loss and metabolic disturbances [9]. Clinical assessment of muscle health encompasses several important components including muscle quantity, strength (the muscle's ability to exert force [10]), and quality (incorporating both functional and morphological domains [11]). In older postmenopausal women, the term "sarcopenia" refers to low muscle mass, strength, and physical performance [12]. However, muscle health assessment in women with POI is challenging as there are no established age-specific thresholds for defining low muscle mass or muscle strength in women <40 years compared with older postmenopausal women [4].

We hypothesized that POI is associated with abnormal skeletal muscle mass, strength, and quality. This pilot study aimed to investigate skeletal muscle health and associated clinical, biochemical, and imaging variables in women with POI (including TS and normal karyotype s-POI women) and compare these findings with age-matched healthy controls.

Materials and Methods

Study Design

This was a cross-sectional cohort study.

Participants

Participants included women aged 20 to 40 years with (1) a diagnosis of s-POI with normal puberty and spontaneous menarche aged <16 years (n = 7); (2) a diagnosis of TS-associated POI (n = 11); or (3) healthy controls (n = 45) with regular menstrual cycles, no history of exogenous estrogen therapy within the last 3 months, and no evidence of clinical hyperandrogenism. Women with impaired renal function, estimated glomerular filtration rate <60 mL/min/1.73 m^2, diagnosed with malignant disease within 2 years, intestinal malabsorption, alcohol abuse, medical condition known to affect bone including drugs (with effects on calcium homeostasis and bone metabolism), or those who were unable to give consent were excluded.

Participants with POI were recruited via hospital clinics in a large multisite health service network and professional networks. Recruitment of healthy controls was achieved via a targeted outreach strategy involving widespread community advertising.

Assessments were completed between 2017 and 2023. Informed consent was obtained from each participant, and all aspects of the study complied with the World Medical Association Declaration of Helsinki—Ethical Principles for Medical Research Involving Human Subjects. The study was approved by the Monash Health Human Research Ethics Committee (reference number 16-0000-575A).

Demographic and Clinical Data

Participants' clinical and demographic data were collected via structured interviews encompassing age, POI causes, comorbidities, hormone therapy (HT), and medication history. For individuals with primary amenorrhea, the duration of amenorrhea was calculated from the age of 13 until the initiation of HT. Depression levels were assessed using the Beck Depression Inventory (BDI) II [13].

Women completed study questionnaires based on the international physical activity questionnaire-short version [14] documenting the type, duration, and frequency of physical activities per week. The physical activity score (PAS) was derived by summing the products of weekly minutes spent in 3 activity categories and their respective metabolic equivalent values (METs): [(light-intensity minutes × 3.0 METs) + (moderate-intensity activity minutes × 4.0 METs) + (vigorous-intensity activity minutes × 7.5 METs)] [15].

Anthropometry

On the day of the dual-energy x-ray absorptiometry (DXA) scan, participants' height (measured in meters) and weight (measured in kilograms) were recorded using a wall-mounted stadiometer and an electronic scale, respectively. Participants were lightly clothed and without shoes during these measurements. These measurements were converted to body mass index (BMI) using the formula: BMI = weight (kg)/height (m^2).

Body Composition and Bone Mineral Density

Muscle quantity/mass was assessed using DXA and peripheral quantitative computed tomography (pQCT) to determine appendicular lean mass (ALM), and muscle cross-sectional area (CSA) of specific muscle groups or body locations, respectively. ALM was normalized to body size, by adjusting for BMI, weight, or height [16, 17].

Dual-Energy X-ray Bone Densitometry

Body composition and bone mineral density (BMD) were assessed using the Hologic Discovery W (Hologic, Bedford MA, USA). Measurements from a whole-body scan included ALM (kg), total fat mass (TFM) (kg), and android/gynoid ratio. ALM and TFM were height-adjusted to generate ALM index = ALM/h^2; kg/m^2 and TFM index = TFM/h^2; kg/m^2. The coefficient of variation for percentage body fat, measured weekly using a total body phantom, was 3.11%.

The prevalence of diminished muscle mass (ALM index) in the 3 groups was determined using established normative data from the Geelong Osteoporosis Study (GOS) for Australian young adult women [18]. Values falling below 1 SD, and 2 SD were grouped together as "low muscle mass."

The DXA scan measured bone area (BA in cm^2), bone mineral content (in g), and areal BMD (aBMD in g/cm^2) at both the lumbar spine (LS) (L1-L4), and femoral neck (FN) regions. We calculated bone mineral apparent density (BMAD) using a previously published formula [19, 20]. The LS aBMD was divided by the square root of the BA to obtain LS BMAD. The FN BMAD was calculated by dividing the total bone mineral content by FN volume, where FN volume = $\pi(W/2)^2 \times h$ and the

average FN width (W) was determined by dividing the FN BA by the height (h) of the scanned region (1.5 cm) [21]. The coefficient of variation for BMD using a LS phantom measured daily on the Hologic Discovery W was 1%.

Peripheral Quantitative Computed Tomography

Participants were seated with their lower leg positioned inside the pQCT (Stratec XCT 3000, Stratec, Germany) gantry and the assessment was conducted on the nondominant lower leg. Standard 2.5-mm transverse scans were taken at 4% and 66% of the tibial length. Muscle CSA (in cm^2), fat area (in cm^2), and trabecular and cortical volumetric BMD (vBMD) in mg/cm^3 were determined using specific density thresholds: a density value of 15 mg/mm^3 separated fat from muscle tissue, and 180 mg/mm^3 separated muscle from bone tissue [22].

Muscle Strength

Muscle strength was evaluated as handgrip strength of the dominant hand, measured with a Jamar plus digital hydraulic handgrip dynamometer (Patterson Medical, Bolingbrook, IL, USA). Three trials were conducted with a 30-second rest between each, and the highest recorded value was used for analysis. In this study, we assessed the functional domain of muscle quality by estimating specific force, calculated as the ratio of handgrip strength (kg) in the dominant hand to ALM of the dominant arm (kg).

The occurrence of low handgrip strength in the 3 groups was assessed based on established normative data from the GOS for Australian young adult women [23]. Similar to the categorization of muscle mass, values falling below 1 SD, and 2 SD were grouped together as "low muscle strength."

Muscle quality has been previously described using 2 different domains [11]: (1) functional, which quantifies muscle function delivered per unit of muscle mass (also known as "specific force") such as the ratio between muscle strength and total, appendicular, or regional muscle mass [24]; and (2) morphological, which evaluates changes in muscle architecture and composition at both microscopic and macroscopic levels [25] such as muscle density and intramuscular adipose tissue [26, 27]. In this study, we assessed the functional domain of muscle quality by estimating specific force, calculated as the ratio of handgrip strength (kg) in the dominant hand to ALM of the dominant arm (kg).

Biochemistry

Blood samples were collected following a 10-hour fast and assessed for 25 hydroxyvitamin D (Cat# 310600, RRID:AB_2811287) levels, renal and liver function tests, electrolytes, reproductive profile (follicle-stimulating hormone [FSH], Beckman Coulter Cat# 33520, RRID:AB_2750983; luteinizing hormone, Beckman Coulter Cat# 33510, RRID: AB_2750984), thyroid-stimulating hormone (TSH) (Beckman Coulter Cat# B63284, (Beckman Coulter Cat# B63284, RRID:AB_3099411), serum calcium (Ca), magnesium (Mg) and phosphate (PO4), transthyretin, insulin-like growth factor-1 (IGF-1) (Cat# 313231, RRID:AB_2928957), procollagen type 1 N-terminal propeptide (P1NP) (Cat# 03141071190, RRID:AB_2782967), C-terminal beta-crosslinked telopeptide of type 1 collagen β (CTX) (Cat# 11972308122, RRID:AB_2905599), and an inflammatory marker—C-reactive protein (CRP) (Beckman Coulter Cat# OSR6299, RRID:AB_3073653). We used serum creatinine (μmol/L)/cystatin C (mg/L) as a surrogate marker for muscle mass; a higher creatinine/cystatin C ratio indicates a greater relative muscle mass [28].

The serum creatinine levels were analyzed using an isotope dilution mass spectrometry traceable Jaffe kinetic assay using an AU5800 Chemistry analyzer (Beckman Coulter, Inc. Brea, CA, USA). The CTX and P1NP were analyzed using a cobas e411 immunoassay analyzer (Roche Diagnostics GmbH, Mannheim, Germany) [29], 25-hydroxyvitamin D and IGF-1 on a Liaison XL immunoassay analyzer (DiaSorin Inc., Stillwater, MN, USA), hormones (FSH and TSH) by a Dx I 800 immunoassay analyzer (Beckman Coulter), and cystatin using a particle-enhanced turbidimetric immunoassay on an Abbott Alinity analyzer (Abbott Laboratories, Abbott Park, IL, USA). Estradiol and testosterone were analyzed by a liquid chromatography-tandem mass spectrometry method, using a Shimadzu Nexeria ultra performance liquid chromatography system for separation and a SCIEX Triple Quad 5500 system for detection (SCIEX, Concord, ON, Canada), CRP by an immune-turbidimetric method and all other parameters were analyzed on a Beckman AU5800 Chemistry analyzer.

Statistical Analysis

We conducted statistical analyses using IBM SPSS Statistics v23 (IBM Corp. USA). Categorical data were presented as percentages, while continuous data were expressed as either mean ± SD or median and 25th to 75th interquartile range (IQR). Independent samples t-tests for 2 groups and ANOVA for multiple groups were used to determine differences between groups for normally distributed continuous variables with Bonferroni's test for post hoc analyses. The Mann–Whitney U test and the Kruskal–Wallis tests were used to analyze differences for nonparametric variables between 2 and 3 groups, respectively. Group differences for categorical variables were assessed using chi-square or Fisher's exact test. A P value of $<.05$ was considered to be statistically significant. We adjusted skeletal, muscle, and body composition parameters to account for height differences using the univariate general linear model, with a significance level set at $P < .05$. Furthermore, muscle parameters, including ALM, ALM index, handgrip strength, and muscle quality (specific force), were additionally adjusted for PAS.

Results

Clinical Characteristics of the Controls and POI Participants

Clinical characteristics of the participants are presented in Table 1. The study included 45 participants in the control group, 11 in the TS group and 7 women in the s-POI group. The median age of all 3 groups was similar. However, participants in the TS group were significantly shorter with a higher BMI.

Nine women with TS had primary amenorrhea; the average duration of amenorrhea prior to HT initiation was 24 months in the TS group and 25 months in the s-POI group. All-POI participants received HT, with 10 women in the TS group and 6 women in the s-POI group utilizing menopausal hormone therapy (MHT), while 1 woman in each of the s-POI and TS groups was prescribed the combined oral

Table 1. Clinical characteristics of the study participants

Characteristic		Control group (n = 45)	TS group (n = 11)	s-POI group (n = 7)	P^a value	All-POI group (n = 18)	P^b value
Age (years)		28.43 ± 8.29	28.18 ± 6.33	29.46 ± 5.03	.35	28.70 ± 5.58	.67
Age at menarche (years)		12.00 (12.00-13.00)	12.50[c] (12.00-16.00)	12.00 (11.50-13.50)	**.03**	12.00 (12.00-15.00)	.07
Fractures		10/44	3/11	2/7	.91	5/18	.69
Hormone therapy	COCP	0/44	1/11	1/7	NA	2/18	NA
	MHT	NA	10/11	6/7	NA	16/18	NA
Physical activity	No. of sessions/week	Light (25) Moderate (32) Vigorous (15)	Light (9) Moderate (6) Vigorous (1)	Light (2) Moderate (7) Vigorous (0)	—	Light (11) Moderate (15) Vigorous (1)	—
	PAS	1350 (840-2250)	450 (375-630)	680 (580-4080)	**.006**	560 (375-780)	**.008**
	Total minutes/week	332 (210-440)	145 (90-210)	230 (90-1320)	**.02**	150 (90-235)	**.012**
Lifestyle factors	Smoking	1/44	0/11	1/7	.24	1/18	.5
	Alcohol	0/34	0/11	0/4	NA	0/14	NA
Supplement use	Calcium	1/44	1/11	2/7	**.04**	3/18	.07
	Vitamin D	6/44	5/11	5/7	**.001**	10/18	**.001**
Psychosocial factors	BDI score	2.50 (0.50-5.50)	7.00 (3.50-11.00)	10.00 (3.50-14.00)	**<.001**	7.00 (2.50-11.00)	**.02**
TSH mIU/L (normal range 0.4-4.8)		1.54 (1.16, 2.75)	2.74 (1.84, 3.00)	1.69 (1.56, 2.56)	.06	2.19 (1.61, 2.99)	.06
eGFR mL/min/1.73 m^2(CKD-Epi)		120 (110, 127)	129 (121, 132)	120 (111, 124)	**.04**	124 (118, 132)	.11
Sex steroid profile	FSH IU/L (premenopausal normal range 1.8-8.8)	5.80 (4.30, 7.10)	15.10 (4.40, 46.9)	8.40 (7.10, 44.05)	.07	8.35 (4.90, 46.9)	**.03**
	Estradiol pmol/L (premenopausal normal range 40-1500)	391.20 (172.10, 671.00)	158.30 (39.60, 232.70)	277.20 (212.20, 310.10)	**.03**	232.10 131.60, 288.70)	**.01**
	Testosterone nmol/L (premenopausal normal range 0.1-1.7)	1.20 (0.90, 1.53)	0.55 (0.33, 0.63)	0.46 (0.42, 0.55)	**<.001**	0.54 (0.40, 0.61)	**<.001**

Bold values denote statistical significance at the $P < .05$ level.
Abbreviations: BDI, Beck Depression Inventory; COCP, combined oral contraceptive pill; eGFR, estimated glomerular filtration rate; FSH, follicle stimulating hormone; MHT, menopausal hormone therapy; PAS, physical activity score; POI, premature ovarian insufficiency; s-POI, spontaneous POI; TS, Turner syndrome; TSH, thyroid-stimulating hormone.
[a]*P* value level of significance between control, TS and s-POI group.
[b]*P* value level of significance between control, and all POI group.
[c]9/11 women with TS had amenorrhea.

contraceptive pill (COCP). The MHT formulations included transdermal (TS n = 7, s-POI n = 5) or oral (TS n = 3, s-POI n = 2) preparations. Most women received doses of 2 mg of oral estradiol daily or transdermal 50 µg/day 17β estradiol (TS n = 10; s-POI n = 5), or doses of transdermal estradiol 75 µg/day or above (TS n = 1; s-POI n = 2) HT, except for 1 woman in the s-POI group who received 1 mg of oral estradiol daily. Growth HT was administered to 10 out of 11 participants during childhood/adolescence in the TS group. Among the recognized TS-associated comorbidities, 1 participant in the TS group had horseshoe kidney, another had hypothyroidism, and 1 had portal hypertension, while no such conditions were reported in the control or s-POI groups.

The control group reported a higher level of physical activity and engaged more frequently in vigorous physical activities than women in both the TS and s-POI groups. Women with POI reported a higher frequency of calcium and vitamin D supplements use compared with controls. The median depression scores on BDI-II scores for each group were within the normal range and below the cutoff score of 10 required for mild depression classification, but, overall, women in the POI groups scored higher on the BDI ($P = .024$).

Muscle, Body Composition, and Skeletal Parameters

Among the 63 participants, pQCT data from 51 women were included in the analysis, comprising controls (n = 38), TS (n = 7), and s-POI (n = 6). Unfortunately, scans were unavailable for 6 women: 5 from the control group and 1 from the TS group. Additionally, 6 scans had to be excluded due to poor image quality, including 2 from controls, 3 from TS, and 1 from the s-POI group. The DXA data were available for 58 participants. In terms of muscle mass parameters, DXA-derived ALM showed significant differences across the 3 groups that persisted after adjusting for PAS. Additionally, ALM relative to TFM and BMI was lower in the POI group ($P < .005$). However, no significant differences were observed for ALM index among the 3 groups and the results did not change after adjusting for PAS.

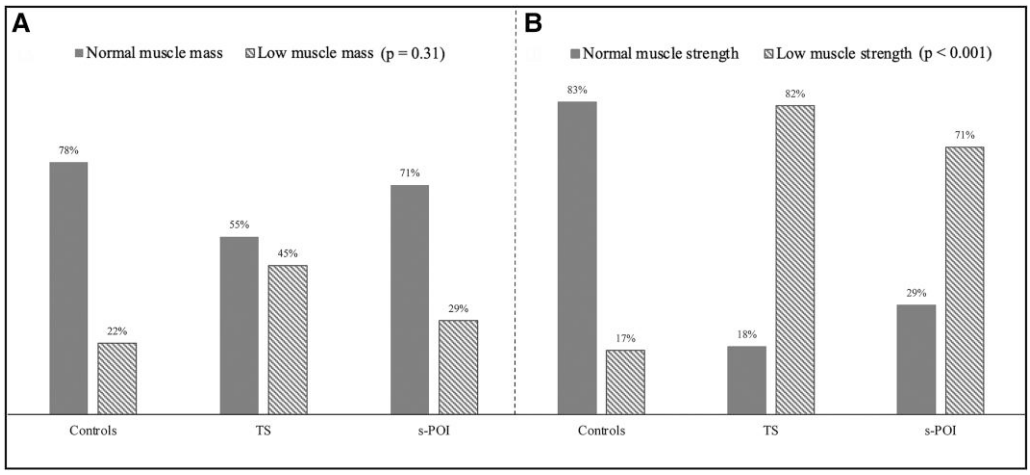

Figure 1. (A) Prevalence of low muscle mass and (B) prevalence of low muscle strength in control, TS, and s-POI groups. The prevalence of low muscle strength differed significantly between the 3 groups ($P < .001$). Low muscle mass and strength was defined as values falling below 1 SD and 2 SD below the GOS-established threshold for normative data Australian young adult women [18, 23].

The prevalence of low muscle mass (ALM index) did not differ significantly among the 3 groups ($P = .31$) (Fig. 1A). Similarly, unadjusted, height-adjusted, and PAS-adjusted pQCT-assessed muscle CSA did not differ between the groups (Table 2). Muscle strength and quality (muscle strength relative to regional muscle mass and BMI assessing the functional domain or specific force) showed significant differences across the 3 groups (Table 2). Both TS and s-POI participants exhibited lower muscle strength and muscle quality (specific force) than controls that persisted after adjusting for height and PAS ($P < .05$); however, no significant differences in these parameters were observed between the 2 POI groups. Additionally, both TS and s-POI participants showed a significantly higher prevalence of low muscle strength than controls ($P < .001$) (Fig. 1B).

Body composition parameters, including pQCT-derived ratio of fat to muscle area and DXA-derived index, varied among the groups. The android/gynoid ratio, also differed significantly between the groups, with the subgroup analyses detecting a difference between the controls and s-POI participants only (Table 2).

Cortical and trabecular vBMD were significantly lower in both the TS and s-POI groups compared with controls, although only cortical vBMD remained significant after height adjustment (Table 2). The post hoc analysis for cortical vBMD detected a difference between the control and TS groups only. Cortical area, but not total area, was significantly lower in participants in the POI groups than in controls, although this did not persist after adjustment for height (Table 2). The DXA parameters showed no significant difference in BMD and BMAD at the LS and FN between the 3 groups.

Biochemistry

Serum creatinine/cystatin C ratio was significantly lower in the TS and s-POI groups (Table 2); however, this finding did not persist after height adjustment. Although within the laboratory normal range, CRP was significantly higher in those with POI than in controls (Table 2). No significant differences were found in IGF-1 or transthyretin levels between groups (see Table 2).

Participants in each group demonstrated normal TSH values and renal function. The reproductive endocrine profile showed significantly higher FSH levels (Table 1) and lower estradiol and total testosterone levels in women with TS and s-POI. The prevalence of vitamin D insufficiency (serum 25-hydroxyvitamin D < 50 nmol/L) was highest in women with TS group (73%), followed by the control group (59%), and lowest in the s-POI group (14%) ($P = .04$) (Fig. 2). The levels of serum-corrected Ca, Mg, and PO_4 were similar across all 3 groups. No statistically significant difference was observed for CTX and P1NP levels among the 3 groups (Table 2).

Discussion

This pilot study explores muscle health in women with POI observing no difference in muscle mass, but lower muscle strength and muscle quality indices, coupled with an increased fat to muscle ratio at the distal tibia in women with POI. These parameters remained significantly lower in women with POI after adjusting for physical activity. The prevalence of low muscle strength was also greater in the POI cohort vs population controls. Exploration of potential contributing factors showed differences in sex steroids, inflammation biomarkers, and physical activity between women with POI and controls.

Our study observed a prevalence of low muscle mass and strength in 40% and 70% respectively of women with POI compared with similar age GOS-established normative data. Consistent with our findings, Li et al reported a 32% prevalence of low muscle mass defined according to Asian Working Group for Sarcopenia criteria for older women [30]. In addition, a recent study assessing sarcopenia risk using the SARC-F tool in women (mean age 61 years) reported a greater risk of sarcopenia in women with surgical (but not spontaneous) premature menopause compared with those undergoing menopause at age 45 years or older [31]. There are currently no established criteria for defining low muscle mass and strength in younger women, including women with POI.

In accordance with previous studies, our research found reduced handgrip strength in women with POI [5], including those with TS [32], compared with controls. Inconsistent

Table 2. Comparative analyses for the muscle, body composition and skeletal parameters between groups

Modality	Parameter	Control group	TS group	s-POI group	*P* value	*P* value (height-adjusted)
Muscle parameters and body composition						
Anthropometry n = 63 Controls = 45 TS = 11 s-POI = 7	Height	163.00 (157-168)	146.50 (144.00-145.50)	164.00 (154-164.5)	**<.001**	—
	Weight	58.54 ± 8.46	57.02 ± 10.86	61.36 ± 7.45	.19	—
	BMI	21.76 ± 2.11	24.86 ± 4.24	23.32 ± 3.69	**.006**	—
pQCT (lower limb) n-51 Controls = 38 TS = 7 s-POI = 6	T66 muscle CSA[a] (mm^2)	6192.88 (5678.00-6897.25)	5623.88 (4542.50-5835.50)	5763.13 (5189.25-6669.25)	.06	.24
	T66 fat to muscle area[b] ratio (%)	42.31 (34.28-46.39)	55.93 (48.66-59.06)	48.54 (43.94-51.57)	**.006**	**.03**
DXA n = 58 Controls = 41 TS = 11 s-POI = 6	ALM (kg)	17.63 ± 2.80	14.11 ± 2.16	16.92 ± 2.01	**<.001**	—
	ALM index (kg/m^2)	6.52 ± 0.70	6.15 ± 0.72	6.41 ± 0.88	.32	—
	TFM (kg)	19.39 ± 4.42	22.59 ± 7.36	22.24 ± 6.32	.14	—
	TFM index (kg/m^2)	7.17 ± 1.43	9.85 ± 3.07	8.29 ± 2.53	**<.001**	—
	Android/gynoid ratio	0.77 ± .09	0.82 ± 0.11	0.90 ± 0.19	**.01**	**.005**
Muscle strength n = 59 Controls = 41 TS = 11 s-POI = 7	Handgrip strength (kg)	28.67 ± 5.65	19.72 ± 4.89	22.73 ± 5.35	**<.001**	**.01**
Muscle quality[c] (specific force) n = 59 Controls = 41 TS = 11 s-POI = 7	HGS/Arm lean mas (kg/kg)	14.10 ± 1.99	11.09 ± 2.06	10.89 ± 2.01	**<.001**	—
	HGS/BMI ($kg/kg/m^2$)	1.32 ± 0.26	0.82 ± 0.25	0.52 ± 0.23	**.006**	—
Biomarkers n = 63 Controls = 45 TS = 11 s-POI = 7	Creatinine/cystatin C (μmol/L/mg/L)	85.82 ± 14.97	69.36 ± 16.45	81.23 ± 6.36	**.006**	—
	CRP (mg/L) (normal 0-5)	0.60 (0.40, 1.10)	2.20 (0.95, 5.65)	2.70 (1.50, 19.75)	**<0.001**	—
	IGF-1 (nmol/L) (normal 15.3-43.1)	31.82 (26.76, 40.28)	34.06 (21.46, 38.79)	31.68 (22.49, 32.92)	0.81	.68
	Transthyretin (g/L) (normal 170-340)	250.00 (235.00, 274.00)	268.00 (226.50, 297.00)	258.00 (234.50, 269.50)	.76	.89
Skeletal parameters						
pQCT n = 51 Controls = 38 TS = 7 s-POI = 6	T4 total area (mm^2)	1051.56 ± 155.98	1115.92 ± 60.02	926.83 ± 63.72	.18	.67
	T4 trabecular density (mg/cm^3)	240.58 ± 34.06	198.34 ± 28.55	236.21± 29.23	**.01**	.06
	T66 cortical area (mm^2)	262.6250 (250.25– 290.75)	208.00 (196.00-252.75)	248.63 (220.75-266.00)	**.007**	.06
	T66 cortical density (mg/cm^3)	1135.63 ± 21.70	1098.44± 31.36	1122.35 ± 32.66	**.01**	**.04**
DXA n = 59 Controls = 41 TS = 11 s-POI = 7	LS-BMD (gm/cm^2)	1.02 ± .11	0.93 ± 0.12	1.03 ± 0.12	.051	—
	LS-BMAD (gm/cm^2)	0.13 ± 0.01	0.13 ± 0.01	0.14 ± 0.02	.37	—
	FN-BMD (gm/cm^2)	0.80 ± 0.10	0.73 ± 0.08	0.79 ± 0.10	.10	.99
	FN-BMAD (gm/cm^2)	0.31 ± 0.04	0.28 ± 0.03	0.31 ± 0.04	.06	—
Biomarkers n = 63 Controls = 45 TS = 11 s-POI = 7	C-telopeptide (ng/L) (normal premenopausal females[d]150-800)	422.00 (352.00, 546.00)	322.00 (218.50, 693.00)	561.00 (437.00, 716.50)	.54	—
	P1NP (μg/L) (normal premenopausal females[d] l5-70)	57.00 (46.00, 74.00)	40.00 (30.50, 77.50)	60.0 (50.50, 74.50)	.29	—
	Vitamin D (nmol/L) (normal 50-75)	44.00 (35.70, 58.70)	44.70 (36.70, 51.60)	68.70 (61.10, 70.50)	**.02**	—

Bold values denote statistical significance at the $P < .05$ level.
Abbreviations: ALM, appendicular lean mass; BMAD, bone mineral apparent density; BMD, bone mineral density; BMI, body mass index; CRP, C-reactive protein; CSA, cross-sectional area; DXA, dual-energy X-ray absorptiometry; HGS, handgrip strength; IGF 1, insulin-like growth factor 1; POI, premature ovarian insufficiency; s-POI, spontaneous POI; P1NP, Procollagen type 1 N propeptide; pQCT, peripheral quantitative computed tomography; TFM, Total fat mass; TS, Turner syndrome.
[a]n = 48.
[b]n = 47.
[c]Muscle quality was assessed as the functional domain.
[d]Aged 20-49 years.

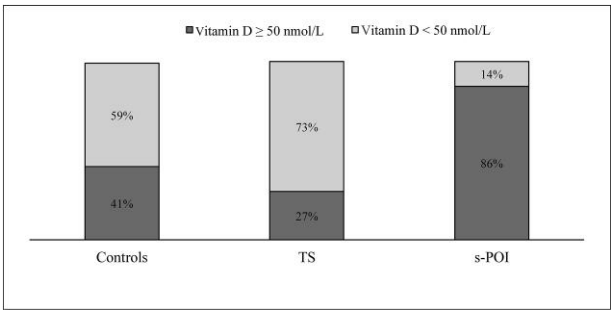

Figure 2. Prevalence of low vitamin D levels in controls, TS, and s-POI women. The prevalence of vitamin D was significantly different between the 3 groups ($P < .001$). Fifty-six percent of women in the POI group were taking vitamin D supplements compared with 14% in the control group.

findings have been reported previously regarding DXA-derived measures of muscle mass in women with POI [30, 33-36], reflecting differences in methodology, ethnicity, cause of POI, and HT use. Despite using the more accurate technique of pQCT-derived CSA [37], we did not observe any difference in muscle mass between the POI and control groups after height adjustment. This may reflect the small sample size; however, no difference in CSA was reported in another TS cohort (n = 21) vs controls [38]. Our study was the first to assess creatinine/cystatin C in women with POI. The creatinine/cystatin C was lower in the TS group; however, after height adjustment, the levels were insignificant, suggesting that this measurement reflects the differences in body size rather than absolute muscle mass.

Various factors interact to regulate skeletal muscle [4] contributing to reduced muscle mass/function including ageing, hormonal (androgens [39] and estrogens [40]), inflammation [41], metabolism, nutrition, and exercise [42, 43]. Consistent with this we observed increased inflammatory biomarkers concentrations, reduced sex steroid levels, and the novel finding of pQCT-derived increased fat to muscle area in women with POI. Myosteatosis is associated with aging and this finding in young women is consistent with the hypothesis that POI is a condition of accelerated aging [44, 45].

We and others have previously identified bone abnormalities such as low BMD and abnormal TBS in women with POI and TS [32, 35, 46, 47]. Similarly, this study demonstrated that women with POI had worse skeletal parameters; however, apart from low cortical BMD in the TS group, none of the results attained statistical significance, possibly due to the small sample size. Muscle and bone health are closely interconnected via myokines and osteokines [48], with the coexistence of osteoporosis and sarcopenia in older adults designated "osteosarcopenia" [49] and associated with worse outcomes than either condition alone. In addition, the factors described above that influence muscle also impact bone. Further investigation is required regarding the roles of osteokines, myokines, and other factors in the pathophysiology underlying musculoskeletal deterioration to improve diagnostic and management strategies.

A 2009 meta-analysis, including 23 studies (n = 9956 postmenopausal women aged 51-77 years), reported increased muscle strength in HT users compared with nonusers despite heterogeneity in HT regimens [50]. Effect sizes varied between different muscle groups with greater effect size for thumb adductors and forearm flexors vs hip adductors or knee flexors. All women with POI received HT in the current study; however, serum estradiol levels were lower, and impaired musculoskeletal parameters were observed in women with POI compared to controls. Li and coworkers observed no difference in DXA-derived muscle parameters between HT users and nonusers in Chinese women with idiopathic POI [30]. Differences between studies may relate to sample size, cause of POI, muscle group assessed and/or HT regimen. A randomized controlled trial of 30 women with TS demonstrated an increase in total body lean mass with a higher dose estrogen replacement (4 mg of oral estradiol per day) compared with a conventional dose of 2 mg of oral estradiol per day [34]. A recent systematic review reported that HT use in women with POI was associated with increased bone density compared with control groups and a cohort study in women with POI indicated that higher doses of estrogen (≥2 mg oral estradiol/day or equivalent) were associated with higher bone density than a lower dose HT or noncontinuous use of the COCP [51, 52]. Thus, many HT regimens commonly used in clinical practice currently may be inadequate for optimal musculoskeletal health in POI and more research is needed.

In our study, participation in physical activity among POI women was notably lower than in the control group, potentially influencing their muscle variables. This is supported by recent findings from Vallejo et al, demonstrating a lower risk of sarcopenia associated with physical activity in women, including those with premature menopause [31] . Our study observation of lower physical activity levels in women with POI aligns with prior research findings showing only 23% of women with s-POI engaged in regular physical activity and that 65% of women with POI were not meeting national exercise recommendations [46, 53]. The higher depression scores in women with TS and s-POI observed in the current study could be an additional barrier to engaging in physical activity. Furthermore, studies focusing on women with TS highlighted short stature, fatigue, anxiety, and visual–spatial deficits as factors contributing to lower physical activity [45].

This pilot study has provided valuable insights into the muscle health of women with POI. However, the limitations in our research include (1) our limited sample size, which may have caused type II error resulting in some of the null findings and precluded robust associations between muscle variables and clinical risk factors; (2) variations in self-reported PAS (subject to recall bias, individual interpretation, and physical function) may have significantly contributed to the differences observed in muscle parameters between the 2 groups; (3) inclusion of knee extension and other functional assessments in addition to hand grip strength can provide a more comprehensive evaluation of global muscle function, and (4) age-matched GOS normative data used to assess the prevalence of low muscle mass were determined using Lunar DPXL and Lunar Prodigy, whereas our study employed Hologic for ALM assessment, Lunar known to yield higher ALM values than Hologic devices [54]. While it can be argued that the ALM prevalence in individual groups might have been lower due to the different devices, our findings of no significant difference in the prevalence of low muscle mass among the 3 groups would have not altered. Future studies with larger cohorts using comprehensive assessment tools are essential to further elucidate the associations between POI and muscle parameters, with rigorous control for critical covariates such as PAS.

Conclusion

This pilot study provides novel insights into the musculoskeletal health of women diagnosed with POI (including TS and s-POI), having identified lower strength, diminished quality, and altered body composition among women with POI compared with their peers. We also identified potential associations including lower estrogen and testosterone levels, increased inflammatory markers, increased depression scores, and decreased physical activity. Notably, our findings may indicate that current HT was insufficient to maintain muscle health in women with POI. This study highlights the need for further research and informs the design of studies to identify underlying mechanisms, diagnostic criteria, and strategies to address the poorer muscle health in this population.

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Disclosures

The authors have nothing to disclose.

Data Availability

The data that support the findings of this study are available from the corresponding author (A.V.) upon reasonable request.

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