Bone Reports 19 (2023) 101700

Contents lists available at ScienceDirect

Bone Reports

journal homepage: www.elsevier.com/locate/bonr

Bone mineral density and its relationship with ground reaction force characteristics during gait in young adults with Prader-Willi Syndrome☆

Daniela A. Rubin[a,*], Skylar C. Holmes[b], Jacqueline Ramirez[a], Steven A. Garcia[c], Eric J. Shumski[d], Derek N. Pamukoff[e]

[a] California State University, Fullerton, 800 N. State College Blvd., Fullerton, CA 92831, United States
[b] University of Massachusetts Amherst, 30 Eastman Lane, Amherst, MA 01003, United States
[c] University of Michigan Dominos Farms, Lobby A. 24 Frank Lloyd Wright Drive, Ann Arbor, MI 48106, United States
[d] University of Georgia, Athens, 330 River Rd, Athens, GA 30602, United States
[e] Western University, 1151 Richmond Street, London, Ontario N6A3K7, Canada

ARTICLE INFO

Keywords:
Prader-Willi syndrome
Obesity
Bone density
Gait
Mechanical forces

ABSTRACT

Introduction: The incidence of osteopenia and osteoporosis is of concern in adults with Prader-Willi syndrome (PWS). Walking generates reaction forces that could stimulate bone mineralization and is popular in people with PWS. This study compared bone parameters and ground reaction forces (GRF) during gait between young adults with PWS and without PWS and explored associations between bone and GRFs during gait.

Methods: 10 adults with PWS, 10 controls with obesity (OB) and 10 with normal weight (NW) matched on sex participated. Segmental and full body dual-energy x-ray absorptiometry scans provided femoral neck, spine, total body minus the head bone mineral density (BMD), bone mineral content (BMC). Vertical GRF, vertical impulse, posterior force and negative impulse were measured during 5 walking trials at a self-selected speed along a 10 m runway.

Results: Multivariate analyses of variance showed that adults with PWS ($n = 7$–8) had hip and body BMD and BMC comparable ($p > .050$) to NW and lower ($p < .050$) than OB. Adults with PWS showed slower speed than NW ($p < .050$) but similar to OB ($p > .050$). Adults with PWS presented lower absolute vertical GRF, vertical impulse and negative impulse than OB ($p < .050$). Pearson r correlations ($p < .050$) in those with PWS (n = 7–8) indicated that femoral neck BMC was associated with vertical GRF ($r = 0.716$), vertical impulse ($r = 0.780$), posterior force ($r = -0.805$), and negative impulse ($r = -0.748$). Spine BMC was associated with speed ($r = 0.829$) and body BMD was associated with speed ($r = 0.893$), and posterior force ($r = -0.780$).

Conclusions: Increased BMC in the femoral neck and body were associated with larger breaking forces during walking, a phenomenon normally observed at greater gait speeds. Faster walking speed was associated with greater BMC in the spine and body. Our preliminary results suggest that young adults with PWS could potentially benefit from faster walking for bone health; however, larger prospective studies are needed to confirm this.

1. Introduction

Prader-Willi syndrome (PWS) is a rare genetic disorder that affects 1 in 15,000 live births resulting from abnormal expression of paternal genes from chromosome 15q11.2-q13 (Cassidy et al., 2012). PWS is a form of congenital obesity and is characterized by hypotonia, high adiposity, low lean mass, hyperphagia, growth hormone (GH) deficiency, hypogonadism, and developmental disabilities (Cassidy et al., 2012). GH replacement therapy (GHRT) coupled with nutrition and exercise have been standard of care for people with PWS in the last decades (Hoybye et al., 2021).

Bone mineral density (BMD) is one of the predictors for fracture risk in people with and without PWS (Brunetti et al., 2018; Marshall et al., 1996). Recent data in a large sample of adults with PWS (median age: 31

☆ Funded by the California State University Chancellor's Research, Scholarship and Creative Activities Program 2017-2018. The sponsor had no role in the study design; in the collection, analysis and interpretation of data; in the writing of the report; and in the decision to submit the article for publication.

* Corresponding author.
E-mail addresses: drubin@fullerton.edu (D.A. Rubin), scholmes@umass.edu (S.C. Holmes), stevenag@umich.edu (S.A. Garcia), Eric.Shumski@uga.edu (E.J. Shumski), dpamukof@uwo.ca (D.N. Pamukoff).

https://doi.org/10.1016/j.bonr.2023.101700
Received 25 June 2023; Received in revised form 11 July 2023; Accepted 14 July 2023

[interquartile range: 25–45]) from Europe and Australia show the incidence of osteoporosis at 14 % and of osteopenia at 54 % in people with PWS (van Abswoude, 2022), supporting earlier data (Brunetti et al., 2018; Butler et al., 2002). Environmental factors, health behaviors and genetic traits influence bone acquisition (Kralick and Zemel, 2020). Sex steroids and GH play an important part in the accrual of peak bone mass during childhood (Mauras et al., 1996) and in the maintenance of bone mass during adulthood (Amin et al., 2006; Finkelstein et al., 2008). Physical activity is also key during growth with weight bearing activities that generate ground reaction forces (GRF) >3.5 body weight per leg providing the most benefits (Weaver et al., 2016; Gunter et al., 2012).

In PWS, low BMD could be attributed to poor bone deposition during childhood-adolescence influencing peak BMD (Kralick and Zemel, 2020; Weaver et al., 2016). Lack of GH or sex hormones, short stature, poor lean mass coupled with low muscle activity all contribute to poor BMD (Butler et al., 2001; Vestergaard et al., 2004; Duran et al., 2016). While increased acceleration because of ambulation at the hip has been associated with better bone health parameters in children with PWS (Duran et al., 2016), insufficient physical activity is a risk factor for osteoporosis in this population (van Abswoude, 2022). To date only one resistance training pilot study in adults has examined the role of short (<10 weeks) exercise interventions in bone health showing no improvement likely because of insufficient study length (Shields et al., 2020).

Walking is the most common activity for people with PWS (Rubin et al., 2012), and while it may be insufficient for some people for starting an osteogenic response (because the impact is not above the necessary threshold) it may still contribute to bone health (Weaver et al., 2016; Kelley et al., 2012). Previous studies evaluating gait in PWS have shown altered thigh-shank and shank-foot coordination that contributes to reduced propulsion and slower speed (Pamukoff et al., 2022). Additionally, difficulty with foot placement during early stance (Pamukoff et al., 2022) may contribute to a more rigid and cautious gait (Rubin et al., 2022). Under healthy conditions, walking provides a weight-bearing stimulus to initiate bone remodeling processes (Anderson and Madigan, 2013). As such, faster walking speed in older adults has been associated with greater BMD in the hip (Moradell et al., 2020; Du et al., 2021). Moreover, walking speeds >5 km/h have been associated with higher BMD in older women (Pellikaan et al., 2018). Gait characteristics associated with speed and weight-bearing may also contribute to markers of bone health in adults with PWS. To our knowledge, no study has evaluated these hypotheses despite reports of lower gait function and bone health in PWS.

Thus, this paper compared bone parameters in young adults with PWS to controls without PWS and with and without obesity. Additionally, we compared gait parameters related to mechanical loading and speed between groups. Last, we examined potential associations between bone parameters and mechanical loading during self-selected gait speeds. We expected that those with PWS will have similar bone parameters compared to those without when considering lean mass differences. We further hypothesized that there will also be a positive relationship between gait speed, ground reaction forces and bone parameters in young adults with PWS.

2. Materials and methods

2.1. Study design

This study had a cross sectional study design.

2.2. Participants

Ten participants with PWS were included if they were between the ages of 18 and 40 and had a genetic diagnosis that confirmed PWS. Participants without PWS were included if they had a body mass index (BMI) between 18.0 and 24.9 kg/m^2 (controls with normal weight [NW], $n = 10$) and 30.0 and 40.0 kg/m^2 (controls with obesity, n = 10) with comparable age (±5 years) and sex. Exclusion criteria were a history of cardiovascular or neurological conditions, or recent (within 6 months) musculoskeletal injury in the lower limbs. A sample size of ten participants per group was estimated based on a priori calculations for a one-way MANOVA. We expected differences between groups with large effect sizes for biomechanical and neuromuscular related outcomes as previously published ($f^2 = 0.65$, number of groups = 3, response variables = 13, $\alpha = 0.05$, $\beta = 0.020$) (Pamukoff et al., 2020a). The study was approved by the Institutional Review Board at California State University, Fullerton. Participants signed a consent form, but for adults with PWS who were unable to sign for themselves the consent form was signed by their legal guardian.

2.3. Anthropometrics

Body mass to the nearest 0.01 kg was obtained using a digital scale (ES200L; Ohaus, Pinewood, NJ, USA) with the subject wearing a t-shirt, shorts, and no shoes. Height was measured to the nearest 0.1 cm using a wall-mounted stadiometer (Seca, Ontario, CA, USA) at the end of inhalation. Body mass index (BMI) was derived from dividing body mass in kg by height in meters squared.

2.4. Bone density

Segmental and full body dual-energy x-ray absorptiometry (DXA) scans (Lunar Prodigy Advance Plus; GE Healthcare, Milwaukee, WI) were done for the hip, spine, and total body. Participants were positioned according to manufacturer's instructions. Bone measurements included areal BMD (g/cm^2), and BMC (grams), for bilateral femoral necks, for the lumbar spine (L1–L4), and for total body minus the head (total body) and their derived z-scores. Proportion of fat and fat-free mass were determined from the total body scan. For participants that did not fit the table area delimitation for the full body scan the mirroring technique was utilized. One technician completed all scans for all participants in the same scanner. The coefficient of variation values (minimum-maximum) for this technician were for bilateral hips = 0.33 % (0–1.24), for lumbar spine = 0.58 % (0–1.92), and for total body = 0.69 % (0.06–1.87) with least significance change (LSC) values within the accepted range of the 2019 statement by the International Society of Bone Densitometry.

2.5. Gait parameters

Three-dimensional walking biomechanics were sampled at 240 Hz using a 9-camera Qualisys Motion Capture System (Goteburn, Sweden) with dual AMTI force plates (AMTI, Watertown MA) sampling at 2400 Hz. Participants were outfitted with bilateral retroreflective markers placed on the first and fifth metatarsal, calcaneus, medial and lateral malleoli, medial and lateral femoral epicondyles, greater trochanter, anterior superior iliac spine, and iliac crest. Rigid clusters of four markers were placed on the sacrum and bilaterally on the foot, shank, and thigh segments. Individual markers were removed after the standing calibration trial. Participants were asked to walk at a self-selected speed over a 10-m walkway while striking consecutive force plates. They performed all activities in self-selected footwear and this is a limitation on the methodology; however this was chosen as people with PWS usually wear orthotics and the intent was to capture a habitual gait pattern without disrupting stability. Speed was monitored using timing gates placed 2-m surrounding the force plates. Five practice trials were completed to confirm that participants could strike force plates without altering their stride, and to determine self-selected gait speed. Afterward, five trials were recorded and accepted if participants made full contact with the plate without altering their stride and were within 5 % of their self-selected speed. Peak vertical ground reaction (VGRF), vertical impulse (VI), peak posterior force (PF), negative impulse (NI) and gait speed (m/s) were extracted for analyses. The peak VGRF and PF were extracted during the first 50 % of the stance phase. The vertical impulse was defined as the integral of the VGRF during the entire stance phase (Fig. 1.A.), and the negative impulse was defined as the negative integral of the PF (Fig. 1.B.). Ground reaction force waveforms were time normalized from 0 to 100 % of the stance phase for data visualization purposes (Fig. 2).

2.6. Data analysis

Mean and standard deviation values were computed for participant characteristics, bone parameters, and gait measurements. One-way analyses of variance compared participant characteristics among groups. Three separate multivariate analyses of variance compared bone parameters (model one), absolute values for gait parameters (model two) and body weight (BW) normalized (n) values for gait parameters (model three) among the groups. Post hoc pairwise comparisons were done using Tukey HSD tests for univariate models and Bonferroni tests for the multivariate. Pearson r correlations explored associations in participants with PWS between bone and gait parameters and their 95 % confidence intervals for 1-tailed tests. Only r values with positive or negative intervals were reported and interpreted. We were unable to collect bone measurements in all participants with PWS, and thus, the sample size for these correlations is 7–8.

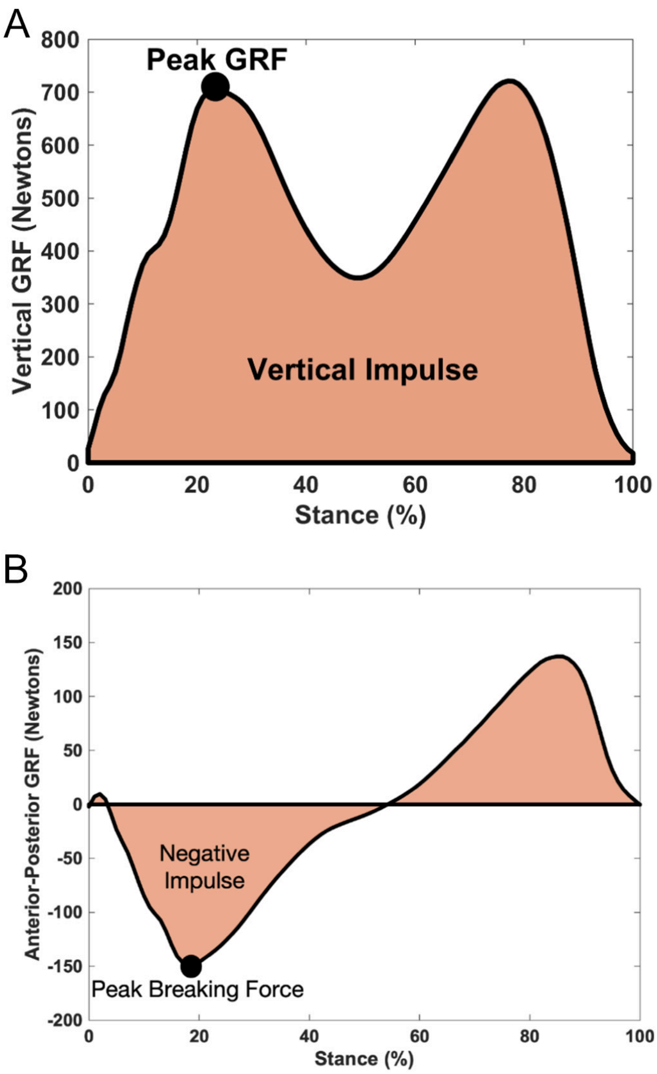

Fig. 1. A. Illustration of peak vertical ground reaction forces (VGRF) extracted during the first 50 % of the stance phase; vertical impulse was defined as the integral of the VGRF during the entire stance phase (Fig. 1 A).
B. Illustration of anterior and posterior ground reaction forces (PF) during the stance phase; the negative impulse was defined as the negative integral of the PF (Fig 1B).

3. Results

Participants with PWS presented the following documented genetic diagnoses: paternal deletion ($n = 7$) and uniparental disomy ($n = 3$). Seven participants with PWS (2 female/5 male) were in growth hormone replacement therapy (GHRT) at 0.4–3 mg/dl daily doses. Seven participants were receiving testosterone, with five of them receiving jointly GHRT and testosterone. One female participant received estrogen jointly with GHRT. Six participants with PWS presented with mild-to-moderate scoliosis and two participants presented past fractures that were older than the past six months, one in the elbow and one in the foot. Currently, participants were enrolled in physical therapy ($n = 1$), horse therapy (n = 1), dietary management ($n = 3$), structured physical activity ($n = 4$). In the past participants received physical therapy ($n = 6$), aquatic therapy ($n = 2$), horse therapy (n = 6), occupational therapy ($n = 7$) dietary management (n = 4), structured physical activity ($n = 5$). Participants with normal weight reported exercising 2 days a week (n = 2) and 3–6 days a week (n = 7) for 40–90 min/session. Participants with obesity reported exercising once a week (n = 2), 2 days a week (n = 4) and 3–6 days a week (n = 4) for 40–120 min/session. Participants with PWS reported exercising 3 days a week (n = 2) and 6–7 days a week ($n = 8$) for 40–60 min/session.

Participant characteristics are shown in Table 1. Estimation of total body fat, lean mass and bone variables was done for ten participants who exceeded the DXA table dimensions. Adults with PWS had comparable age and height to controls with NW and controls with obesity. Adults with PWS had lower body mass, BMI, and lean mass than controls with obesity but comparable to controls with NW. Adults with PWS had greater body fat than controls with NW but comparable to controls with obesity. Pairwise comparisons indicated lower gait speed in those with PWS compared to controls with NW ($p = .021$), but not compared to controls with obesity ($p = .514$).

A one-way MANOVA comparing bone parameters was significant (Pillai's Trace =1.127, $F_{18,\ 32} = 2.293$, $p = .020$, Table 2). Group effects were found for femoral neck BMD, BMD z-score and BMC, total body BMD, BMD z-score and BMC ($p < .048$ for all). Adults with PWS had lower femoral neck BMD, BMD z-score and BMC, and total body BMD, BMD z-score and BMC than controls with obesity ($p < .049$ for all) but similar to controls with NW ($p > .645$ for all) (Please see Table 2). Spine BMD was not statistically different between groups ($p = .054$), but a large effect size was observed (partial eta square = 0.225). However, no significant differences or large effect sizes were found when comparing those with PWS to controls with obesity or to NW ($p > .061$ for both). There were no significant differences among the groups for spine BMD z-score or BMC ($p > .225$ for both). There were two participants with PWS (one male and one female) who presented a femoral neck BMD lower than expected for age (z-score < -2.0). There were one male participant with PWS and one male participant with obesity with spine BMD z-scores lower than expected for age.

A one-way MANOVA comparing gait absolute mechanical loading parameters was significant (Pillai's Trace = 0.965, $F_{8,50} = 5.829$, $p < .001$, Table 3A). Group effects were found for VGRF, vertical impulse, posterior force and negative impulse ($p < .041$ for all). The group with PWS had lower absolute VGRF, vertical impulse, and negative impulse compared to controls with obesity ($p = .003$, $p = .001$ and $p = .009$). The group with obesity had a greater posterior force than controls with NW ($p = .038$) All other variables were not significantly different between the groups ($p > .114$).

A one-way MANOVA comparing gait normalized mechanical loading parameters was significant (Pillai's Trace = 0.525, $F_{8,50} = 2.225$, $p = .041$, Table 3B). A group effect was only found for normalized VGRF ($p = .019$). The group with PWS had lower normalized VGRF ($p = .020$)

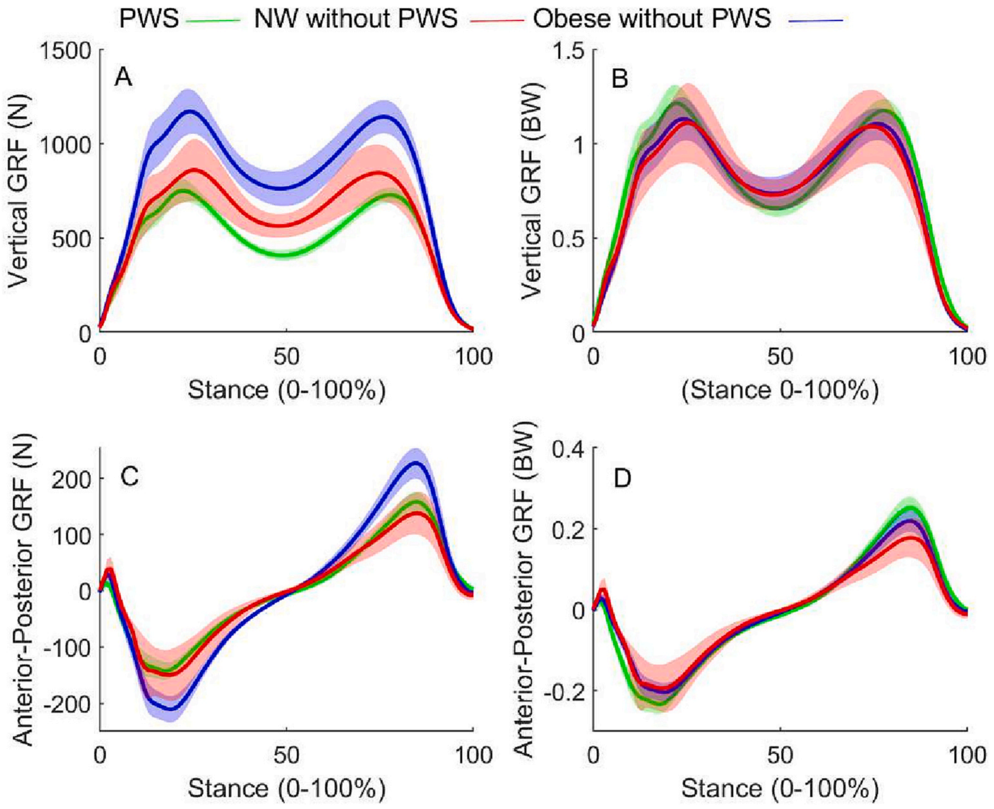

Fig. 2. Ensemble average and 95 % confidence interval for (A) absolute vertical ground reaction force; (B) normalized vertical ground reaction force; (C) absolute anterior-posterior ground reaction force; (D) normalized anterior-posterior ground reaction force. Red represents adults with PWS; green represents adults with normal weight without PWS; blue represents adults with obesity without PWS.

Table 1

Participant characteristics by groups: with PWS, with normal weight and with obesity. Values shown as mean (standard deviation).

	Adults with PWS (*n* = 10)	Adults with normal weight without PWS (*n* = 10)	Adults with obesity without PWS (n = 10)	Group *p*-value
Age (years)	22 (5)	23 (3)	22 (2)	0.978
Sex (Female/ Male)	3/7	3/7	3/7	
Height (cm)	166.53 (14.56)	166.69 (6.67)	174.08 (8.99)	0.210
Body Mass (kg)	79.09 (21.3)^{b}	63.57 (5.04)	105.46 (15.46)	<0.001
Body Mass Index (kg/ m^{2})	28.12 (5.43)^{b}	22.86 (1.19)	34.63 (3.01)	<0.001
Body fat %	40.6 (7.8)^{a}	23.4 (7.8)	42.4 (5.6)	<0.001
Lean mass (kg)	44.7 (11.0)^{b}	46.7 (7.3)	58.7 (12.2)	0.012
Gait speed (m/s)	1.240 (0.184)^{a}	1.466 (0.140)	1.328 (0.198)	0.026

[a] Different from Adults with normal weight without PWS, $p < .050$.

[b] Different from Adults with obesity without PWS, $p < .050$.

than controls with NW. All other variables were not significantly different between the groups ($p > .052$).

The most salient associations are presented in Fig. 3. Femoral neck BMD ($n = 8$) was associated with normalized posterior force ($r = -0.748$, 95 % Confidence Interval: −0.951, −0.091). Femoral neck BMC ($n = 8$) was associated with VGRF ($r = 0.716$; 95 % Confidence Interval:

Table 2

Bone parameters in adults with PWS, with obesity and with normal weight without PWS. Values are shown as mean ± SD.

	Adults with PWS (*n* = 7)	Adults with normal weight without PWS (*n* = 10)	Adults with obesity without PWS (*n* = 9)	Group *p*-value	Eta^{2}
Femoral neck BMD (g/ cm^{2})	0.947 (0.169)^{a}	1.016 (0.103)	1.303 (0.206)	<0.001	0.500
Femoral neck BMD z-score	−1.029 (1.432)	−0.343 (0.769)	1.200 (1.625)	0.006	0.357
Femoral neck BMC (g)	4.77 (0.92)^{a}	4.99 (0.76)	6.67 (1.01)	<0.001	0.503
Spine BMD (g/cm^{2})	1.127 (0.102)^{a}	1.197 (0.168)	1.330 (0.189)	0.054	0.225
Spine BMD z-score	−0.800	0.187	0.383	0.226	0.121
Spine BMC (g)	65.13 (10.14)	60.06 (18.51)	72.80 (19.40)	0.285	0.103
Total body BMD (g/ cm^{2})	1.144 (0.089)^{a}	1.159 (0.086)	1.327 (0.108)	<0.001	0.500
Total body BMD z-score	−0.714 (1.025)	0.922 (1.500)	−0.040 (1.128)	0.047	0.234
Total body BMC (g)	2880.96 (571.26)^{a}	2580.80 (313.92)	3523.60 (578.96)	<0.001	0.495

[a] Different from Adults with obesity without PWS, $p < .050$.

Table 3A
Absolute gait biomechanical factors presented by group as mean (standard deviation).

	Adults with PWS (n = 10)	Adults with normal weight without PWS (n = 10)	Adults with obesity without PWS (n = 10)	Group *p*-value
VGRF (N)	873.474 (295.397)[a]	765.499 (92.33)	1183.180 (183.457)	<0.001
Vertical Impulse (N*s)	423.657 (113.83)[a]	336.433 (34.965)	594.473 (111.167)	<0.001
Posterior Force (N)	−169.183 (79.882)	−152.763 (26.335)	−214.886 (39.139)	0.040
Negative Impulse (N*s)	−25.099 (11.950)[a]	−23.726 (3.561)	−36.601 (5.969)	0.002

[a] Different from Adults with obesity without PWS, $p < .050$.

Table 3B
Normalized gait biomechanical factors presented by group as mean (standard deviation).

	Adults with PWS (n = 10)	Adults with normal weight without PWS (n = 10)	Adults with obesity without PWS (n = 10)	Group *p*-value
VGRF normalized (N*kg^{-1})	1.122 (0.072)[a]	1.225 (0.083)	1.144 (0.085)	0.019
Vertical Impulse normalized (N*s*kg^{-1})	0.547 (0.048)	0.539 (0.033)	0.572 (0.047)	0.225
Posterior Force normalized (N*kg^{-1})	−0.211 (0.049)	−0.244 (0.033)	−0.209 (0.041)	0.124
Negative impulse normalized (N*s*kg^{-1})	−0.031 (0.008)	−0.038 (0.005)	−0.035 (0.005)	0.063

[a] Different from Adults with normal weight without PWS, $p < .050$.

0.112, 1.000), vertical impulse ($r = 0.780$; 95 % Confidence Interval: 0.249,1.000), posterior force ($r = -0.805$, 95 % Confidence Interval: −1.000, −0.309), and negative impulse (r = −0.748, 95 % Confidence Interval: −1.000, −0.178). Spine BMC ($n = 7$) was associated only with speed ($r = 0.829$; 95 % Confidence Interval: 0.284, 1.000). Total body BMD (n = 7) was associated with speed ($r = 0.893$; 95 % Confidence Interval: 0.428, 0.984), posterior force ($r = -0.780$; 95 % Confidence Interval: − 0.966, −0.066), and normalized posterior force ($r = -0.795$; 95 % Confidence Interval: − 0.968, −0.106). Total body BMC (n = 7) was associated with speed ($r = 0.871$; 95 % Confidence Interval: 494, 1.000).

4. Discussion

Advances in understanding and management of PWS has led to an increase in the life expectancy of adults with PWS (Whittington et al., 2015; Proffitt et al., 2019) (Whittington et al., 2001) prompting interest in issues related to healthy aging. One such issue is bone health because 44–54 % and 14–16 % of adults with PWS present with osteopenia and osteoporosis, respectively (van Abswoude, 2022; Butler et al., 2001; Sinnema et al., 2012). As such, history of fractures is high (~50 %) in adults older than 50 years (Sinnema et al., 2012) and is in young adults (29–60 %) (Butler et al., 2002; Kroonen et al., 2006). And, as in other populations, greater risk of fractures is associated with mortality in PWS (Marshall et al., 1996; Proffitt et al., 2019). Thus, understanding factors that positively contribute to BMD and prevent the risk of fracture and morbidity are of utmost importance.

This study shows that young adults with PWS present comparable bone parameters and lean mass but greater fat than controls with NW. As expected, adults with PWS had lower bone parameters for the hip and total body than controls with obesity. Adults with PWS had lower gait speed and normalized VGRF than controls with NW and lower VGRF, vertical impulse and negative impulse than controls with obesity. In PWS, BMC in the femoral neck was associated to vertical and anterior-posterior mechanical forces during gait, while BMC in the spine and total body BMD and BMC were associated with gait speed.

Young adults with PWS had similar BMD and BMC compared to controls with NW. Earlier studies found lower bone BMC and BMD for the spine and the total body in adults with PWS who were overweight when compared to obese controls (Butler et al., 2001). Additionally, adults with PWS only had lower BMD for the spine, the femoral neck and total body compared to controls with NW when height was not accounted for, and no difference was found when controlling for height in analyses (Jorgensen et al., 2013; Longhi et al., 2015). In the present study, adults with PWS had similar body mass, lean mass and height (determinants of bone parameters) as controls with NW. Thus, similarities in bone parameters between the group with PWS and the control group with NW may be explained by similar height, lean mass, and body mass (Viardot et al., 2018). For instance, previous research found that children with PWS who were not GH naïve had comparable bone parameters to normal weight controls when matched by height and sex (Rubin et al., 2013) and in children with PWS not treated with GH a similar result was found when matched to controls by height (Edouard et al., 2012). Specifically, most participants ($n = 7$) in this sample had received or were receiving GHRT, which contributes to normalization of height and increases lean mass (Hoybye et al., 2021; Davies et al., 1998; Lindgren and Ritzen, 1999). Additionally, one participant had received estrogen and six participants received testosterone which also contribute to BMD when in conjunction with GHRT (Donze et al., 2018). The differences between the group PWS and controls with obesity at this age were expected. Non-syndromal obesity may not negatively influence bone parameters until later in life due to chronic inflammatory factors that are detrimental to bone development (Zhu et al., 2015). However, in young adults, additional mass may provide additional mechanical stimuli that elevates BMD.

The group with PWS had slower self-selected speed than controls with NW. Previous reports have found slower (Vismara et al., 2007) or comparable (Cimolin et al., 2021) gait speed in those with PWS compared to healthy controls. Lower absolute VGRF in PWS compared to controls with obesity are similar to other reports (Pamukoff et al., 2020a) and so are lower normalized VGRF in controls with obesity when compared to controls with normal weight (Pamukoff et al., 2020b). We also found that some ground reaction forces characteristics (e.g. VGRF) remained lower in those with PWS compared to controls with NW after normalizing to body mass. As such, there are additional factors besides body mass that may contribute to lower ground reaction forces in PWS. For example, we have previously hypothesized that lower plantarflexor function contributes to impaired propulsion and slower speed in PWS (Pamukoff et al., 2022). Adults with PWS show altered lower limb coordination compared with controls which may reduce propulsion during terminal stance and contribute to slower walking speed. Adults with PWS also walk with altered spatiotemporal gait features that may reduce single limb loading, and further contribute to lower VGRF. These aspects are important for mobility and for bone health because faster walking speed was associated with greater BMC, particularly for the spine and total body. We and others have shown that adults with PWS self-select slower walking speeds when compared to adults without PWS (Pamukoff et al., 2022; Vismara et al., 2007). The shorter, wider steps and longer double limb support in those with PWS likely reduces the load placed in single limb (Cimolin et al., 2010). With faster walking, there will be less time spent in double stance, thus increasing the load on single limbs during the gait cycle potentially generating greater vertical forces that may translate to the spine. Previous work by our group in children has shown that greater acceleration counts at the hip during

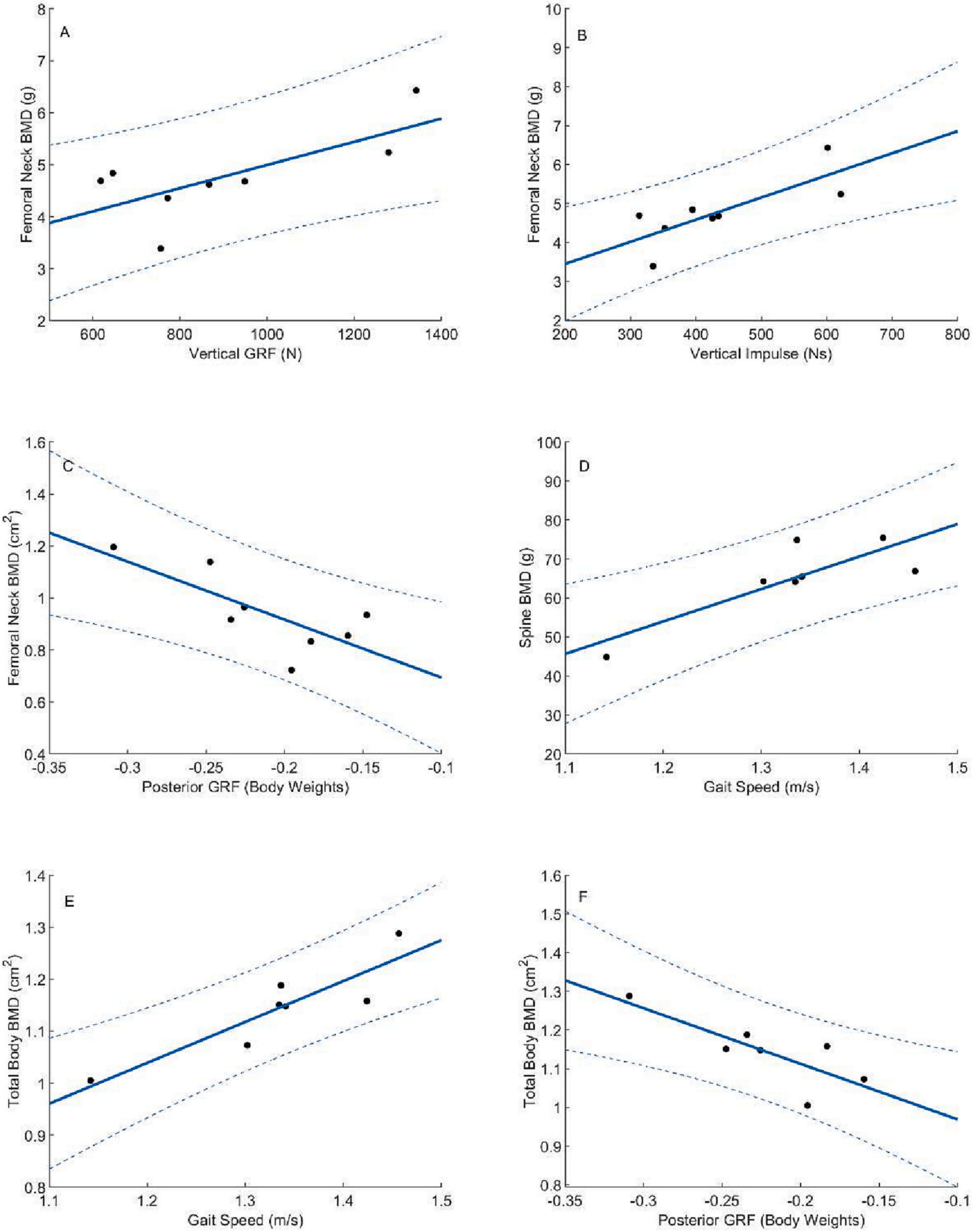

Fig. 3. Scatterplots presenting associations between gait mechanical loading characteristics and bone parameters in the group with PWS (n = 7–8). Fig. A presents vertical ground reaction forces (GRF) and femoral neck bone mineral density (BMD); Fig. B presents vertical impulse and femoral neck BMD; Fig. C presents posterior GRF normalized by body weights; Fig. D presents gait speed and spine BMD; Fig. E presents gait speed and total body BMD; Fig. E presents posterior GRF normalized by body weights and total body bone mineral content (BMC).

habitual physical activity were positively related to BMD in children with PWS (Duran et al., 2016). Thus, while a faster walk could be emphasized to increase VGRF, it should be considered that perhaps those with PWS adjust their gait to shorter and wider steps to prevent falling.

Vertical ground reaction forces during walking were related to femoral neck BMC. Weight-bearing exercise applies mechanical stress to bones through the ground reaction forces and the contractile activity of the muscles (Klein-Nulend et al., 2012). Therefore, these associations between BMC in the lower extremities and VGRF were expected. The VGRF may contribute to larger compressive stress on lower limb weight-bearing bones, which may be beneficial for bone mineralization. Moreover, larger negative braking forces were associated with BMC in the femoral neck. A larger posterior ground reaction force contributes to shear stress along and transverse to the long axis of weight bearing bones. The posterior ground reaction force has previously been linked to lower extremity stress fracture in runners (Napier et al., 2018), and bones are generally less able to tolerate large horizontal forces. However, in the context of walking in those with PWS, we posit that larger posterior forces from faster walking may contribute to bone development, and future studies are needed to confirm these hypotheses. Adults with PWS have altered foot-ankle coordination that manipulate the foot position at ground contact during early stance (Pamukoff et al., 2022). As such, those with PWS may not generate sufficient braking force to decelerate the body during each step contributing to gait instability. Alternatively, larger braking forces have been associated with greater energy absorption at the knee and hip during running gait (Heiderscheit et al., 2011). As such, future studies are needed to determine optimal loading parameters during gait or other weight-bearing activities for bone development in individuals with PWS.

We used very stringent criteria to evaluate associations between bone and mechanical loading factors of gait, and we did not interpret moderate associations between gait speed and femoral neck BMC ($r = 0.442$) due to the limited sample size. Thus, we cannot exclude the link between faster walking and BMC in the lower extremities. However, as our results are based on a cross-sectional design, future studies with a longitudinal are needed to test the possible role of faster walking in bone health in PWS. Clearly, there is much room and need for randomized control trials in this population to determine the potential effect of resistance training, walking, impact exercises, and/or vibration platforms in bone parameters.

4.1. Study limitations

This study used a cross-sectional design and does not indicate a causal relationship or directionality of the association between gait and bone parameters. We had a small sample size, but PWS is a rare disease with a very low prevalence. Despite the small sample, we still identified meaningful group differences in bone parameters with large effect sizes, and some hypothesized associations between gait kinetics and bone parameters. As the sample size was small, comparisons between the sexes were not statistically feasible; this is a limitation of the findings. Some participants with PWS had metal inserts ($n = 2$) or did not agree to have a DXA scan ($n = 1$), which resulted in a lower sample size than anticipated ($n = 7$–8). We did not control for GHRT in our analyses as a covariate as the sample size was too small. As such, future studies would benefit from larger samples that can better control for confounders (e.g. GHRT, vitamin D or calcium supplementation, history of smoking, and habitual physical activity). Additionally, we used ground reaction forces as surrogates of external bone loading, and these outcomes may not represent the internal stress applied to bones during weight-bearing tasks. Nonetheless, we identified meaningful and expected associations between ground reaction force outcomes and BMD that suggest that additional weightbearing activity and gait modification may be beneficial for bone health in adults with PWS.

5. Conclusion

The results of this study suggest that adults with PWS have lower BMD compared with controls with obesity. This study also showed associations between BMD and BMC and gait parameters in PWS suggesting walking may be related to bone health. A focus on strengthening and balancing exercises, as well as walking interventions are needed to facilitate faster walking speeds that may increase ground reaction forces. Efficient ambulation along with a reduction in double limb stance may positively influence bone parameters in PWS; however, larger prospective studies are needed to confirm this.

CRediT authorship contribution statement

Daniela A. Rubin: Writing – original draft, Methodology, Investigation, Funding acquisition, Formal analysis, Conceptualization. **Skylar C. Holmes:** Writing – review & editing, Investigation, Data curation. **Jacqueline Ramirez:** Writing – original draft. **Steven A. Garcia:** Writing – review & editing, Data curation. **Eric J. Shumski:** Writing – review & editing, Data curation. **Derek N. Pamukoff:** Writing – review & editing, Supervision, Software, Methodology, Investigation, Funding acquisition, Conceptualization.

Declaration of competing interest

None.

Data availability

Data will be made available on request.

Acknowledgement

The authors would like to thank the study participants for their time, the Prader-Willi California Foundation for advertising of the study, and Brett Post and Derrick L. Escano for their help during data collection.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bonr.2023.101700.

References

Amin, S., et al., 2006. Estradiol, testosterone, and the risk for hip fractures in elderly men from the Framingham study. Am. J. Med. 119 (5), 426–433.

Anderson, D.E., Madigan, M.L., 2013. Effects of age-related differences in femoral loading and bone mineral density on strains in the proximal femur during controlled walking. J. Appl. Biomech. 29 (5), 505–516.

Brunetti, G., et al., 2018. Analysis of circulating mediators of bone remodeling in Prader-Willi syndrome. Calcif. Tissue Int. 102 (6), 635–643.

Butler, M.G., et al., 2001. Decreased bone mineral density in Prader-Willi syndrome: comparison with obese subjects. Am. J. Med. Genet. 103 (3), 216–222.

Butler, J.V., et al., 2002. Prevalence of, and risk factors for, physical ill-health in people with Prader-Willi syndrome: a population-based study. Dev. Med. Child Neurol. 44 (4), 248–255.

Cassidy, S.B., et al., 2012. Prader-Willi syndrome. Genet. Med. 14 (1), 10–26.

Cimolin, V., et al., 2010. Gait patterns in Prader-Willi and Down syndrome patients. J. Neuroeng. Rehabil. 7, 28.

Cimolin, V., et al., 2021. Gait strategy and body composition in patients with Prader-Willi syndrome. Eat. Weight Disord. 26 (1), 115–124.

Davies, P.S., et al., 1998. Effect of growth hormone on height, weight, and body composition in Prader-Willi syndrome. Arch. Dis. Child. 78 (5), 474–476.

Donze, S.H., et al., 2018. Bone mineral density in young adults with Prader-Willi syndrome: a randomized, placebo-controlled, crossover GH trial. Clin. Endocrinol. 88 (6), 806–812.

Du, X., et al., 2021. The impact of nordic walking on bone properties in postmenopausal women with pre-diabetes and non-alcohol fatty liver disease. Int. J. Environ. Res. Public Health 18 (14).

Duran, A.T., et al., 2016. Association between physical activity and bone in children with Prader-Willi syndrome. J. Pediatr. Endocrinol. Metab. 29 (7), 819–826.

Edouard, T., et al., 2012. Muscle-bone characteristics in children with Prader-Willi syndrome. J. Clin. Endocrinol. Metab. 97 (2), E275–E281.

Finkelstein, J.S., et al., 2008. Bone mineral density changes during the menopause transition in a multiethnic cohort of women. J. Clin. Endocrinol. Metab. 93 (3), 861–868.

Gunter, K.B., Almstedt, H.C., Janz, K.F., 2012. Physical activity in childhood may be the key to optimizing lifespan skeletal health. Exerc. Sport Sci. Rev. 40 (1), 13–21.

Heiderscheit, B.C., et al., 2011. Effects of step rate manipulation on joint mechanics during running. Med. Sci. Sports Exerc. 43 (2), 296–302.

Hoybye, C., et al., 2021. Time for a general approval of growth hormone treatment in adults with Prader-Willi syndrome. Orphanet J. Rare Dis. 16 (1), 69.

Jorgensen, A.P., et al., 2013. Two years of growth hormone treatment in adults with Prader-Willi syndrome do not improve the low BMD. J. Clin. Endocrinol. Metab. 98 (4), E753–E760.

Kelley, G.A., Kelley, K.S., Kohrt, W.M., 2012. Effects of ground and joint reaction force exercise on lumbar spine and femoral neck bone mineral density in postmenopausal women: a meta-analysis of randomized controlled trials. BMC Musculoskelet. Disord. 13, 177.

Klein-Nulend, J., Bacabac, R.G., Bakker, A.D., 2012. Mechanical loading and how it affects bone cells: the role of the osteocyte cytoskeleton in maintaining our skeleton. Eur. Cell Mater. 24, 278–291.

Kralick, A.E., Zemel, B.S., 2020. Evolutionary perspectives on the developing skeleton and implications for lifelong health. Front. Endocrinol. 11, 99.

Kroonen, L.T., et al., 2006. Prader-Willi Syndrome: clinical concerns for the orthopaedic surgeon. J. Pediatr. Orthop. 26 (5), 673–679.

Lindgren, A.C., Ritzen, E.M., 1999. Five years of growth hormone treatment in children with Prader-Willi syndrome. Swedish National Growth Hormone Advisory Group. Acta Paediatr. Suppl. 88 (433), 109–111.

Longhi, S., et al., 2015. Adults with Prader-Willi syndrome have weaker bones: effect of treatment with GH and sex steroids. Calcif. Tissue Int. 96 (2), 160–166.

Marshall, D., Johnell, O., Wedel, H., 1996. Meta-analysis of how well measures of bone mineral density predict occurrence of osteoporotic fractures. BMJ 312 (7041), 1254–1259.

Mauras, N., et al., 1996. Sex steroids, growth hormone, insulin-like growth factor-1: neuroendocrine and metabolic regulation in puberty. Horm. Res. 45 (1–2), 74–80.

Moradell, A., et al., 2020. Associations between physical fitness, bone mass, and structure in older people. Biomed. Res. Int. 2020, 6930682.

Napier, C., et al., 2018. Kinetic risk factors of running-related injuries in female recreational runners. Scand. J. Med. Sci. Sports 28 (10), 2164–2172.

Pamukoff, D.N., et al., 2020a. Plantar flexor function in adults with and without Prader-Willi Syndrome. Med. Sci. Sports Exerc. 52 (10), 2189–2197.

Pamukoff, D.N., et al., 2020b. Body mass index moderates the association between gait kinetics, body composition, and femoral knee cartilage characteristics. J. Orthop. Res. 38 (12), 2685–2695.

Pamukoff, D.N., et al., 2022. Lower extremity coordination and joint kinetic distribution during gait in adults with and without Prader-Willi Syndrome. J. Biomech. 141, 111213.

Pellikaan, P., et al., 2018. Ranking of osteogenic potential of physical exercises in postmenopausal women based on femoral neck strains. PLoS One 13 (4), e0195463.

Proffitt, J., et al., 2019. Contributing factors of mortality in Prader-Willi syndrome. Am. J. Med. Genet. A 179 (2), 196–205.

Rubin, D.A., et al., 2012. Physical activity in children with Prader-Willi Syndrome: a parents' perspective. Californian J. Health Promot. 10, 57–66.

Rubin, D.A., et al., 2013. Update on body composition and bone density in children with Prader-Willi syndrome. Horm. Res. Paediatr. 79 (5), 271–276.

Rubin, D.A., et al., 2022. Body Composition in Adults with Prader-Willi Syndrome and Its Relationship with Gait Spatiotemporal Parameters in American College of Sports Medicine 2022 Annual Meeting and World Congress. Lippincot Williams, San Diego, CA.

Shields, N., et al., 2020. Is strength training feasible for young people with Prader-Willi syndrome? A phase I randomised controlled trial. Physiotherapy 106, 136–144.

Sinnema, M., et al., 2012. Aging in Prader-Willi syndrome: twelve persons over the age of 50 years. Am. J. Med. Genet. A 158A (6), 1326–1336.

van Abswoude, D.H., et al., 2022. Bone health in adults with Prader-Willi syndrome: clinical recommendations based on a multicenter cohort study. J. Clin. Endocrinol. Metab. 108 (1), 59–84. https://doi.org/10.1210/clinem/dgac556.

Vestergaard, P., et al., 2004. Reduced bone mineral density and increased bone turnover in Prader-Willi syndrome compared with controls matched for sex and body mass index–a cross-sectional study. J. Pediatr. 144 (5), 614–619.

Viardot, A., et al., 2018. Relative contributions of lean and fat mass to bone mineral density: insight from Prader-Willi Syndrome. Front. Endocrinol. 9, 480.

Vismara, L., et al., 2007. Clinical implications of gait analysis in the rehabilitation of adult patients with "Prader-Willi" Syndrome: a cross-sectional comparative study ("Prader-Willi" Syndrome vs matched obese patients and healthy subjects). J. Neuroeng. Rehabil. 4, 14.

Weaver, C.M., et al., 2016. The National Osteoporosis Foundation's position statement on peak bone mass development and lifestyle factors: a systematic review and implementation recommendations. Osteoporos. Int. 27 (4), 1281–1386.

Whittington, J.E., et al., 2001. Population prevalence and estimated birth incidence and mortality rate for people with Prader-Willi syndrome in one UK Health Region. J. Med. Genet. 38 (11), 792–798.

Whittington, J.E., Holland, A.J., Webb, T., 2015. Ageing in people with Prader-Willi syndrome: mortality in the UK population cohort and morbidity in an older sample of adults. Psychol. Med. 45 (3), 615–621.

Zhu, K., et al., 2015. Associations between body mass index, lean and fat body mass and bone mineral density in middle-aged Australians: the Busselton Healthy Ageing Study. Bone 74, 146–152.