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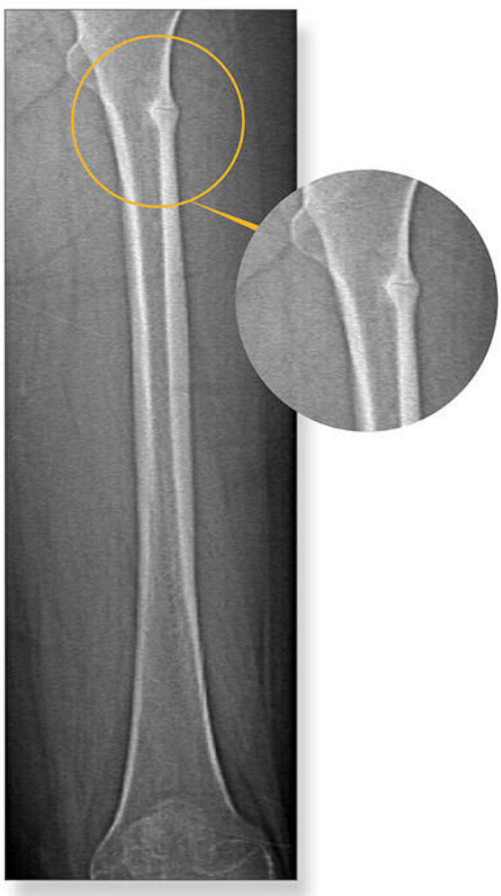

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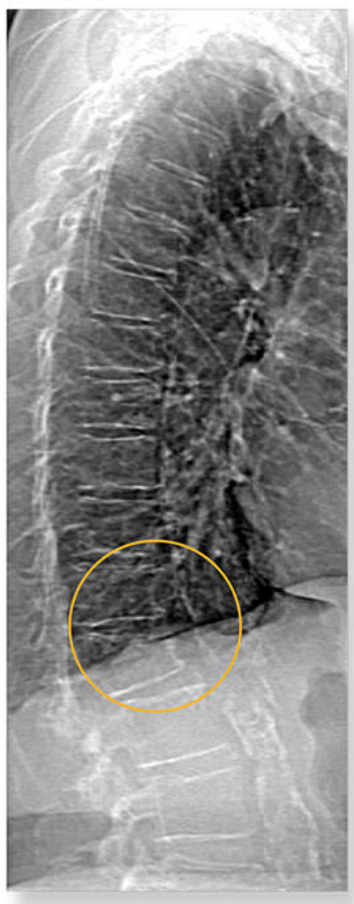

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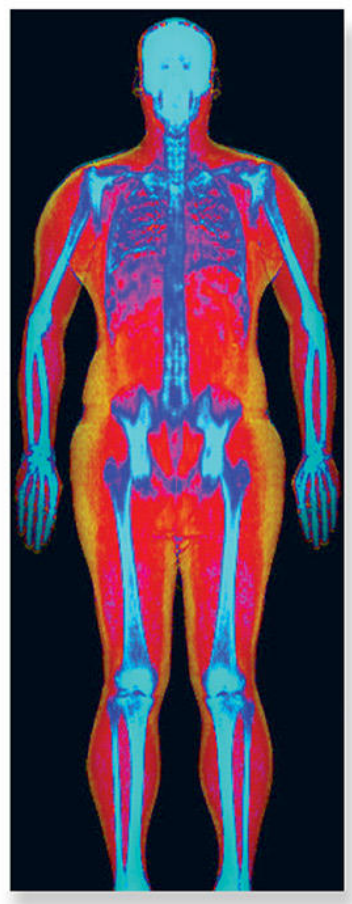

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Low Magnitude Mechanical Loading Is Osteogenic in Children With Disabling Conditions

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ABSTRACT: The osteogenic potential of short durations of low-level mechanical stimuli was examined in children with disabling conditions. The mean change in tibia vTBMD was +6.3% in the intervention group compared with −11.9% in the control group. This pilot randomized controlled trial provides preliminary evidence that low-level mechanical stimuli represent a noninvasive, non-pharmacological treatment of low BMD in children with disabling conditions.

Introduction: Recent animal studies have demonstrated the anabolic potential of low-magnitude, high-frequency mechanical stimuli to the trabecular bone of weight-bearing regions of the skeleton. The main aim of this prospective, double-blind, randomized placebo-controlled pilot trial (RCT) was to examine whether these signals could effectively increase tibial and spinal volumetric trabecular BMD (vTBMD; mg/ml) in children with disabling conditions.
Materials and Methods: Twenty pre-or postpubertal disabled, ambulant, children (14 males, 6 females; mean age, 9.1 ± 4.3 years; range, 4–19 years) were randomized to standing on active ($n = 10$; 0.3g, 90 Hz) or placebo ($n = 10$) devices for 10 minutes/day, 5 days/week for 6 months. The primary outcomes of the trial were proximal tibial and spinal (L_2) vTBMD (mg/ml), measured using 3-D QCT. Posthoc analyses were performed to determine whether the treatment had an effect on diaphyseal cortical bone and muscle parameters.
Results and Conclusions: Compliance was 44% (4.4 minutes per day), as determined by mean time on treatment (567.9 minutes) compared with expected time on treatment over the 6 months (1300 minutes). After 6 months, the mean change in proximal tibial vTBMD in children who stood on active devices was 6.27 mg/ml (+6.3%); in children who stood on placebo devices, vTBMD decreased by −9.45 mg/ml (−11.9%). Thus, the net benefit of treatment was +15.72 mg/ml (17.7%; $p = 0.0033$). In the spine, the net benefit of treatment, compared with placebo, was +6.72 mg/ml, ($p = 0.14$). Diaphyseal bone and muscle parameters did not show a response to treatment. The results of this pilot RCT have shown for the first time that low-magnitude, high-frequency mechanical stimuli are anabolic to trabecular bone in children, possibly by providing a surrogate for suppressed muscular activity in the disabled. Over the course of a longer treatment period, harnessing bone's sensitivity to these stimuli may provide a non-pharmacological treatment for bone fragility in children.
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Key words: clinical/pediatrics, mechanical loading, bone QCT, novel entities, tibia, osteoporosis

INTRODUCTION

A CENTURY-OLD PREMISE, widely referred to as Wolff's law,[1] first described the strong influence of function on skeletal morphology. These strain signals, which arise in bone tissue during loading,[2] enhance the bone density of participants in intense exercise,[3] while the dearth of such signals is considered the key etiologic factor in the bone fragility that afflicts children with disabling conditions such as cerebral palsy.[4] This "form follows function" relationship has fueled the presumption that sporadic, large strain events (3000 microstrain) are more important in defining skeletal architecture than the persistent barrage of low-level mechanical signals that arise from passive activities such as standing.[5]

In contrast to a "bigger is better" premise for bone adaptation, recent experiments in animals have demonstrated that high-frequency (10–90 Hz), extremely low-magnitude ($\ll$100 microstrain) strain stimuli are strongly anabolic to trabecular bone.[6,7] Results show that there are increases in trabecular BMD, width, and number in the weight-bearing

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skeleton, and that brief exposure to these low-level signals can effectively inhibit disuse osteopenia.[8] These higher frequency mechanical strain signals in bone, although small, are physiological in nature, resulting from the contractions of adjacent musculature.[9] Over any given 24-h period, these low-level signals represent a dominant component of a bone's mechanical strain history.[10]

Taken together, these data indicate that the maintenance of skeletal health may depend as much on the persistent barrage of low-magnitude, high-frequency loads arising from long-term, relatively passive activities such as standing as it does on the relatively large, but far less frequent, low-frequency, high-amplitude loads associated with locomotion.[11]

Children with disabling conditions such as cerebral palsy (CP) and muscular dystrophy (MD) are prone to fractures of their long bones, which occur with minimal trauma.[4,12] BMD, a surrogate for bone strength, is reduced in children with CP and MD compared with their healthy peers.[13–18] In a previous cross-sectional study in children with CP,[19] we reported that the degree of reduction in calcaneal broadband ultrasound attenuation (related to bone mass and structure) and spinal volumetric trabecular BMD (vTBMD) were associated with the degree of immobility and non–weight-bearing of subjects.

Together, these findings indicate that a reduced level of activity in disabled children is reflected by a lower BMD, which predisposes them to fractures. Some means of inhibiting further bone loss, or improving bone density, should help to reduce the number of fractures in these children. Considering all of these factors, this pilot trial was designed to examine whether such low-level mechanical signals could effectively enhance trabecular BMD in this at-risk population. The primary hypothesis of this randomized, double-blind, placebo-controlled, pilot trial (RCT) was that short daily doses (10 minutes/day) of low-magnitude, high-frequency loading (vibration) intervention (0.3g, 90 Hz) would serve as an anabolic stimulus and cause an increase in the tibial and spinal vTBMD of ambulant children with disabling conditions. Posthoc analyses were performed to investigate the effects of the intervention on parameters related to diaphyseal bone strength (cortical BMD [vCBMD], mg/ml; cross-sectional bone area; periosteal bone circumference; and the polar moment of inertia) and muscle cross-sectional area.

MATERIALS AND METHODS

Study group

The pilot trial was approved by the North-West England Multi-Centre Research Ethics Committee and was performed in concordance with the Declaration of Helsinki. Informed written consent was obtained from the parents of each child. Consultant community pediatricians identified suitable children for the trial; the main criterion for recruitment was that the children had to be able to stand independently but have limited mobility associated with their disability. The parents of 45 children were approached; 23 agreed to participate in the trial (49%), of whom 20 (14 males, 6 females) fulfilled the inclusion criteria (mean age, 9.1 $\pm$ 4.3 years; range, 4–19 years) and took part in this pilot RCT.

Weight (kg), height (m), and calcium intake (mg; 3-day dietary recall; CompEat, Nutrition Systems, Grantham, UK) were estimated at the beginning and end of the trial. Each subject's muscle tone was classified into (1) low (reduced muscular activity resulting in a degree of floppiness in the limbs)/variable tone (predominantly low tone with occasional periods of high tone) or (2) spastic (sustained increased tone in the limb/limbs) categories. The pubertal status of each subject was determined by the grading system of Tanner.[20] Subjects were matched using approximately similar spinal vTBMD SD scores,[21] and each child within the pair was randomly allocated to either the intervention (active) or the placebo (control) group.

Loading regimen

Loading intervention was provided through vertical ground-based vibration,[22] induced by a small plate oscillating at 90 Hz, designed to create peak–peak accelerations of 2.9 m/s^2, referred to as a fraction of earth's gravitational field, 0.3g (1g = 9.8 m/s^2). The placebo devices were identical in appearance, but when activated, did not vibrate; instead, they emitted a 500-Hz audible tone, identical to that produced by active devices. Subjects were instructed to stand on the active or placebo devices for 10 minutes each day, 5 days/week for 6 months (Fig. 1). The intervention was performed either in the home or at school. The displacement of the device, at 0.3g, 90 Hz, is less than 100 μm. Each device has a built-in electronic monitoring system that automatically detects and records the duration that the subject stood on the device (Fig. 1). For each child, the total duration that he/she stood on the device was used to assess length of treatment and compliance.

Outcome measures

QCT scan protocol: Despite subjects' underlying medical conditions and associated disabilities (autism, involuntary movements, limb deformity, and spasticity), all scans were performed without sedation.

3-D scans of the spine and proximal tibia were obtained using a Philips Medical Systems SR-4000 Tomoscan (Best, Netherlands) scanner. The CT scan parameters were 120 kV, 50 mA, 2-s slice scan time, field of view = 420, and voxel size 0.82 mm $\times$ 0.82 mm $\times$ 3 mm; a 3-D block of longitudinal length 90 mm was collected at each site; the volume scanned was limited by the X-ray cooling requirement of the CT scanner. A fluid di-potassium hydrogen phosphate (K_2HPO_4) bone equivalent calibration phantom (Mindways, San Francisco, CA, USA) was placed centrally on the scanner table and covered with gel bolus bags to eliminate air between phantom and patient, which may cause artifacts; the child was positioned appropriately over the phantom. The phantom contains differing concentrations of K_2HPO_4 (50, 100, 200 mg/ml) and is used for image quantitation, transforming CT Hounsfield Units into bone mineral equivalents (mg/ml). For spinal scans, the child laid supine on the scanner table with the lower thoracic and lumbar spine centered over the gel pads and the arms raised and placed on a pillow. A lateral scan projection radiograph

FIG. 1. A child standing on the vibrating platform. Note that the angulation of the knee, which was not controlled for, could influence the transmissibility of the mechanical signal to the axial skeleton. A desktop was incorporated into the platform system to allow the child to draw or read during the active or placebo interventions. The arrow indicates the electronic monitoring device for recording compliance.

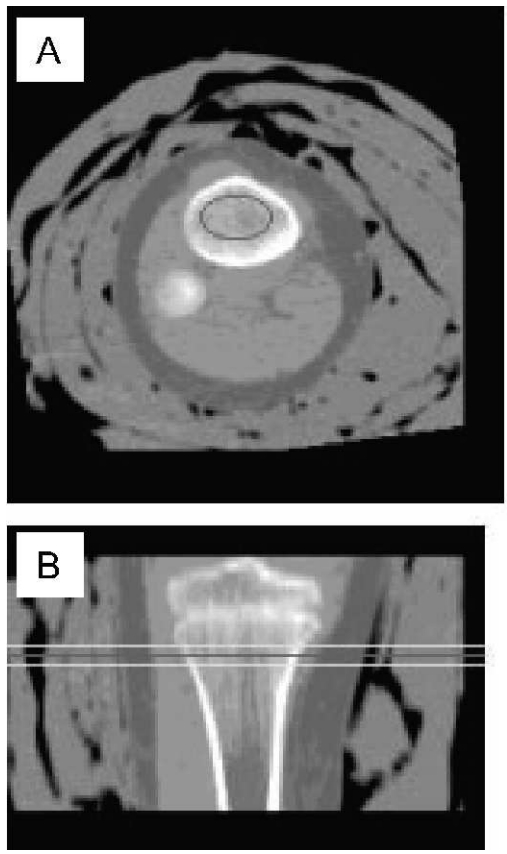

FIG. 2. vTBMD measurement. A reconstructed image of a proximal tibial scan showing (A) the axial view, the cross-sectional size and position of the ROI is shown, and (B) the coronal view, the region of analysis is annotated on the image. This was manually positioned just distal to the growth plate, away from zone of provisional calcification. These parameters were exactly the same in baseline and follow-up scans.

was taken from T_{10}–L_5, and scanning levels were prescribed from this localizer image. In the tibial scans, both tibias had to be positioned in the scan field, and measurements were made in the proximal segment of the nondominant proximal tibia in all children; if the child had hemiplegia, nondominant was defined as the affected side. A posterior–anterior (PA) scan projection radiograph was taken from the knee joint to the upper one-third of the proximal tibia, and sections were prescribed distally from the tibial plateau.

Total duration for both spinal and tibial examinations, including positioning, was 10 minutes each; actual scan time was 1 minute/site. The baseline projection radiograph for each site was used to aid section positioning in the follow-up examination.

All baseline and follow-up scans were performed and analyzed by a radiographer (CA) who was blinded to treatment allocation and had much experience in performing and analyzing QCT scans. To ensure consistency, an experienced radiologist (JA) checked all baseline and follow-up scans for quality and location of regions of interest (ROIs).

vTBMD measurements of spine and tibia: The primary prespecified outcome measures of this pilot RCT were vTBMD derived from a 9-mm transverse section in the mid-plane of the vertebrae (L_1–L_3 or L_2–L_3; the vertebrae included depended on the size of the child) and proximal tibia; data were analyzed using QCT-Pro software (Mindways). Vertebral BMD analyses were performed in a section in the midplane of the lumbar vertebral body using visualization of the basi-vertebral vein to confirm positioning; this is the conventional site for spinal scan analysis.[23]

The QCT-Pro software automatically transforms the vertebral vTBMD values into SD scores using the data collected in healthy 2- to 19-year-old North American white subjects.[21] These normative data were collected on a different make of scanner, but the software normalizes for differences during quality assurance procedures and is appropriate in subjects with a body area below 600 cm^2.[24]

The proximal tibial analyses were made in the plane distal to the tibio-fibular junction, avoiding the growth plate and the metaphyseal zone of provisional calcification, thus ensuring that purely vTBMD was measured (Fig. 2). Despite short scans times, the difficulties in scanning these children meant that some scan sections were degraded by movement artifact and had to be excluded from analysis. In 4 of 20 subjects, the thickness of the volume analyzed was reduced from 9 mm to either 5 ($n = 2$ subjects) or 7 mm ($n = 2$

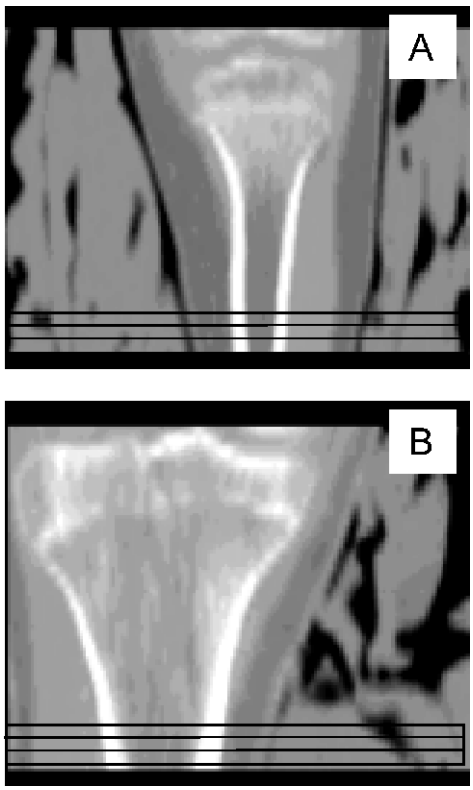

FIG. 3. Mid-diaphyseal bone analysis. Examples of coronal reconstructions from (A) a postpubertal and (B) a prepubertal child. Shown on the images are the locations of ROIs for the posthoc analyses. This varied between 25% and 50% of the total tibial length, depending on the size of the child.

subjects). However, pre- and post-trial sections in the same individual were always consistent in their anatomical positioning and volume thickness. To ensure the accurate relocation of the ROI in the follow-up scan, the digitally stored baseline scan was restored on a computer workstation and used for comparison. The cross-sectional area and volume thickness of the ROI analyzed in the baseline and the follow-up scans were identical (Fig. 2).

Measurements of diaphyseal cross-sectional bone area, periosteal bone circumference, vCBMD, polar moment of inertia, cortical thickness, and muscle area: Digitally stored 90-mm blocks of tibial data provided an opportunity to explore changes in the diaphyseal portion of the tibia, that is, parameters related to diaphyseal bone strength and muscle area. In each child, the diaphyseal measurements were made at the same site in baseline and follow-up scans. To optimize the amount of diaphyseal bone analyzed measurements were always taken at the most distal sections of the scan, and therefore, the location of analysis was dependent on the length of the tibia; in younger children, these measurements were made at approximately 50% tibial length, whereas in older children, they were made at around 25% tibial length (Fig. 3). Three adjacent slices per scan were used to maximize the volume of data analyzed (9 mm); a mean of the results from these three sections was taken and used for data analysis. BonAlyse software (version 1.3; BonAlyse Ltd., Jyväskylä, Finland) was used for analyses; a contour threshold algorithm was used to automatically separate trabecular and cortical bone by user-defined thresholds, which were determined by studying histogram profiles of the images using full width at half maximum to select the threshold. The thresholds selected for bone were pixels with values between 100 and 1500 mg/ml (a threshold of 438 mg/ml was used to separate cortical from trabecular bone), and for muscle analysis, pixels with values between −52 and 54 mg/ml. These thresholds were used for all subjects and all scans. Outcome measures were vCBMD (mg/ml) parameters of diaphyseal bone geometry and muscle area.

The total radiation dose (effective dose equivalent) for the scans was 85 μSv (55 μSv lumbar spine, 30 μSv tibia).[25] In a group of children with CP, root mean square precision (CV%)[26] of *repeated analysis* of pre- and post-trial QCT scans ($n = 48$) of tibia vTBMD was 2.1%.[27] Precision of *reanalysis* in the spine was 0.9%; this is similar to the CV reported by other centers (0.9–1.3%).[28,29] Precision after *repositioning* was not determined in children for ethical and radiation dose reasons. However, in our unit the precision for *repositioning* in adults was 0.9% (spine) and 1.8% (tibia).

Statistical analysis

Independent sample *t*-tests were used to determine the effect of treatment on vTBMD before adjustment for the prespecified covariates. For adjusted analyses, a multiple regression model was used to determine the effect of treatment on spinal (L_2 vertebral body) and tibial vTBMD (mg/ml). The baseline covariates of age, weight, muscle tone category, puberty, calcium intake, corresponding baseline vTBMD, and time on treatment were included in the model. To determine whether the BMD response altered with compliance, the interaction between treatment group and trial duration was entered into the model. All data were tested for normality and are presented as mean changes with 95% CIs. All analysis was by intention-to-treat.

For posthoc analyses of diaphyseal bone parameters and muscle area the same multiple regression model was used as previously adjusting for the same covariates as for vTBMD changes.

RESULTS

Over the course of the 6-month trial, three children dropped out (two intervention, one control); one child's behavioral problems worsened after the trial began, another child began an intensive physiotherapy program and was too tired to stand for the 10 minutes required for this trial, and the third child got bored with participation. Regardless, each of these children had follow-up BMD scans at the end of the study for inclusion in final intention-to-treat analysis. No adverse effects of the low-magnitude, high-frequency loading treatment were reported or observed during the trial.

All children ($n = 20$) had baseline and follow-up BMD scans at the end of the 6-month study period, and mean time between baseline and follow-up scans was 8 months; the time between start of intervention and follow-up scan was no more than 7 months. Nineteen scans were successfully

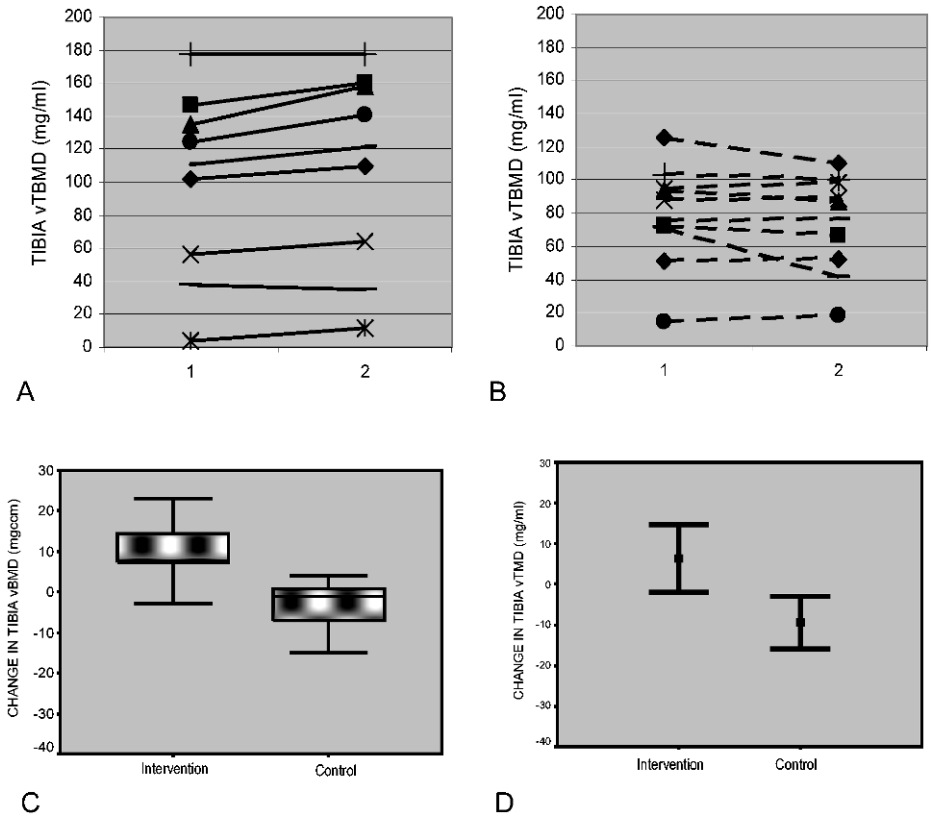

FIG. 4. Graphs illustrating the absolute values at baseline and at 6 months for tibial vTBMD (mg/ml) for each individual subject in (A) intervention and (B) placebo groups. (C) The median unadjusted change in vTBMD is shown with interquartile ranges; lines represent high and low values, excluding outliers. (D) The adjusted mean change in vTBMD is shown with 95% CIs.

analyzed; one patient was excluded from spine and tibia analysis because of degradation of scan quality caused by movement artifacts (this child had dropped out of the study). As good quality pre- and post-trial scans were obtained in all study subjects in L_2 the vTBMD of this vertebra (L_2) was used for statistical analysis. Posthoc analyses on tibial diaphyseal strength parameters and cross-sectional muscle area were performed in 19 subjects.

Compliance

Ten subjects participated in the trial at home and 10 at school (7 of these subjects were in a residential school). The median duration of treatment actually received by subjects ($n = 20$) in the active and placebo groups was 35.5 days (range, 15–117 days), with the median standing time of 481 minutes (range, 88–1206 minutes). The compliance of the trial, in relation to mean standing time on the devices (567.9 minutes) compared with prescribed standing time (1300 minutes), was 44% (4.4 minutes/day). There were no significant differences in compliance between the home and school group or between those in the active and control group.

Influence of mechanical stimulation on proximal tibia vTBMD

In children who stood on active devices, a 6.27 mg/ml increase in tibial vTBMD ($n = 9$; 95% CI, −2.07, 14.06), representing a 6.3% increase over baseline was measured (Fig. 4A). This is in contrast to the response observed in children who stood on placebo devices, with the mean change in proximal tibial vTBMD being a decrease of 9.45 mg/ml ($n = 10$; 95% CI, −15.89, −3.02), which represents an 11.9% decrease from baseline measurements (Fig. 4B). Unadjusted analyses, performed without the prespecified baseline covariates, also showed a significant effect of treatment compared with the controls ($p = 0.004$, mean difference 14.35 mg/ml; 95% CI, 5.32, 23.4); these data are presented in Fig. 4C.

Compared with placebo, the mean net difference in proximal tibial vTBMD of the active group was +15.72 mg/ml (95% CI, 6.57. 24.87; $p = 0.0033$), reflecting a +17.7% difference between the two groups (Fig. 4D). There was no evidence of an interaction between efficacy of intervention and compliance ($p = 0.27$), indicating little influence of duration of intervention on the change in proximal tibial vTBMD.

Influence of mechanical stimulation on L_2 vTBMD

In children who stood on active devices, a +7.29 mg/ml increase in spinal vTBMD was found ($n = 9$; 95% CI, −0.88, 15.46), representing a 5.5% increase over baseline. In children who stood on placebo devices, the mean change of spinal vTBMD was +0.56 mg/ml ($n = 10$; 95% CI, −5.93, 7.06), representing a 0.3% increase from baseline measurements. The mean change in spinal vTBMD was 6.72 mg/ml higher for the active treatment group compared with the control (95% CI, −2.60, 16.05; $p = 0.14$), representing a 4.7% difference between the two groups. Unad-

TABLE 1. CHARACTERISTICS OF SUBJECTS, MEASURED AT THE BEGINNING OF THE TRIAL (MEAN ± SD)

Covariate		*Active*	*Placebo*
Age (years)		6.9 ± 2.4	11.2 ± 4.7
Weight (kg)		25.8 ± 7.0	40.8 ± 19.9
Disability category (N)	Spasticity	8	6
	Variable/low tone	2	4
Pubertal stage (N)	Prepubertal	10	5
	Postpubertal	0	5
Calcium intake (MG)		892.8 ± 326.5	858.6 ± 411.9
Spinal vTBMD (L2)		133.2 ± 31.9	151.1 ± 29
Spinal BMD Z-score		−1.2 (1.2)	−1.0 (1.3)
Tibial vTBMD (mg/ml)		99.3 ± 56.2	79.1 ± 30.5

TABLE 2. UNADJUSTED BASELINE AND FOLLOW-UP TIBIAL (FIG. 4) AND VERTEBRAL VTBMD VALUES FOR ACTIVE AND PLACEBO GROUPS

		Tibial vTBMD (mg/ml)			*Spinal vTBMD (mg/ml)*		
Group	*Child*	*Baseline*	*Follow-up*	*Change*	*Baseline*	*Follow-up*	*Change*
Active	1	101.6	109.3	7.7	150.1	149.6	−0.5
	2	146.2	160.5	14.3	105.3	116.5	11.2
	3	135.4	158.5	23.1	142.1	152.3	10.2
	4	56.5	63.8	7.3	133.1	128.4	−4.7
	5	3.7	11.2	7.5	67	60	−7
	6*	124.3	140.3	16	166	154.4	−11.6
	7	177.9	177.3	−0.6	141.1	155.4	14.3
	8	110.4	121.7	11.3	144.1	153.7	9.6
	9	37.5	34.5	−3.0	108	118.9	10.9
Placebo	10	50.9	51.8	0.9	153.2	156.2	3
	11	72.4	66.6	−5.8	102.4	98.8	−3.6
	12	94	86.9	−7.1	197.8	214.2	16.4
	13	88.4	89.1	0.7	153.7	146.3	−7.4
	14	94.9	98.2	3.3	139.8	143.6	3.8
	15	14.5	18.5	4.0	152.8	152.8	0
	16	103.4	100.4	−3.0	178.1	179.5	1.4
	17*	75.4	76.1	0.7	115.3	114.4	−0.9
	18	71.4	42	−29.4	178.2	172.6	−5.6
	19	125.5	110.5	−15.0	139.5	126.9	−12.6

* These children dropped out of the study and were scanned at the end of the 6-month period for inclusion into the intention-to-treat analysis.

justed analyses also showed a nonsignificant effect of treatment compared with the controls (mean difference, 4.15 mg/ml; 95% CI, −4.27, 12.58; $p = 0.31$).

Influence of mechanical stimulation on diaphyseal cross-sectional bone area, periosteal bone circumference, vCBMD, polar moment of inertia, cortical thickness, and muscle area

There were no significant changes in diaphyseal bone area, circumference, vCBMD, polar moment or inertia, cortical thickness, or muscle area, respectively. These data are presented in Table 3.

DISCUSSION

To the best of our knowledge, this is the first RCT to investigate the effects of low-magnitude, high-frequency loading treatment on low TBMD in children with disabling conditions. The results of this pilot trial in children with disabling conditions indicate that extremely low-magnitude, high-frequency mechanical stimuli can be strongly anabolic to trabecular bone in humans, directly contrasting with the perception that functional signals need be large to be influential in skeletal morphology.[5] In vivo evidence in animals indicates that the 0.3*g* accelerations, similar to those used in this RCT, will induce a mechanical signal well below 5 microstrain.[7] Considering this in relation to the peak strains (>3000 microstrain) experienced during intense activities,[30] these data suggest that bone modeling and remodeling are influenced more by a biological benefit of loading[31] rather than mediated by the repair of microdamage.[32]

The 6.3% increase in tibial vTBMD of the active group compared with the placebo group (−11.9%) was achieved in the relatively short period of 6 months after only 4.4 minutes of daily treatment, implying that rather than "accumulating" adaptive signals in the bone tissue, the bone's

Table 3. Summary of Posthoc Analysis: Unadjusted Changes of Diaphyseal Bone Parameters

	Intervention			*Control*		
Outcome	*Baseline*	*Follow-up*	*Change*	*Baseline*	*Follow-up*	*Change*
WBA (mm^2)	281.8	305.0	23.2	513.6	525.4	11.8
WBD (mg cc)	456.8	458.6	1.8	417.3	416.1	−1.1
WBCIRC (mm)	58.8	61.1	2.4	78.4	79.3	0.9
PMI (mg mm)	8,059.3	9,516.6	1,457.3	29,100.8	30,566.4	1,465.6
CTA (mm^2)	134.9	145.3	10.3	208.9	210.6	1.8
CTD (mg cc)	699.7	709.5	9.8	703.9	715.7	11.8
CT THK (mm)	3.6	3.6	0.06	3.7	3.7	0.006
MA (mm^2)	1,496.5	1,593.3	129.8	2,225.2	2,510.5	141.5
MD (mg cc)	24.8	23.8	−1.0	26.4	26.8	0.39
B:M RATIO	0.2	0.2	0.003	0.2	0.2	−0.004

WBA, whole bone area; WBD, whole bone density; WBCIRC, whole bone circumference; PMI, polar moment of inertia; CTA, cortical area; CTD, cortical density; CT THK, cortical thickness; MA, muscle area; MD, muscle density; B:M ratio, bone:muscle ratio.

response is elicited after only a brief exposure to an anabolic stimulus. The triggering of this adaptive response by even brief exposure to the mechanical stimulus is consistent with findings in animal studies, where the bone tissue's response rapidly (72 s) reaches a threshold, and additional mechanical input has no added benefit to the anabolic response.[33] The relatively low compliance (44%) is likely to be because of a variety of reasons, including (1) these were a challenging group of disabled children in whom to conduct a research RCT, many of whom had behavioral problems; (2) one control child began a physiotherapy program and became too tired to comply with the RCT and therefore withdrew; (3) some children lost the motivation to stand on the platforms; and (4) some parents found it difficult to supervise the prescribed standing treatment, especially if there were other siblings in the house.

While the lack of an observed effect of the intervention on spinal vTBMD is disappointing, it could well be a result of inefficient transmission of these low-level mechanical stimuli to the spine, because of the subjects' abnormal stance (Fig. 1) dampening the transmission of high-frequency signals to the axial skeleton.[34] Furthermore, there is the possibility that these low-magnitude, high-frequency signals are anabolic only where there is low bone mass, as shown in animal models,[35] or that postural actions of spinal musculature[9] supersede any mechanical signals that the platform delivers. There may also be the possibility that there is a site-specific sensitivity to mechanical stimuli, just as there is a differential sensitivity in responsiveness of the spine, versus the appendicular skeleton, to some pharmaceutical agents.[36] Nevertheless, the large increase in tibial vTBMD in the active subjects compared with the much lower response seen in this RCT group at the spine is further evidence that these low-level signals are anabolic in the lower appendicular skeleton and that adaptation in bone is locally, rather than systemically, controlled.

The sensitivity of the skeletal system to perceive and respond to signals in the order of tens—rather than thousands—of microstrain is remarkable; however, it is not clear how such exceedingly small mechanical signals influence bone tissue. Recent work shows[37–39] that by-products of deformation, such as fluid flow[40] and intramedullary pressure,[41] may amplify the signal as dependent on frequency (e.g., an increase from 0.1 to 10 Hz will elevate pressure by *an order of magnitude*). Considering that accelerations at this magnitude and frequency are barely perceptible, it is also possible that the anabolic response is regulated indirectly through a system such as neuromuscular feedback perturbed by exceeding a stochastic threshold.[42]

Evidence from animal studies suggests that the expression of several genes critical to bone formation are better influenced by low-level than high-level signals,[43] but of course, this does not dismiss large signals as having no osteogenic potential.[44] Although bone architecture can be influenced by very few large strain signals,[33] such signals happen only rarely,[11] even under the severe conditions of military training.[45] Therefore, signals that arise from more typical activities (e.g., walking) would serve as a more dependable means of defining bone morphology. Ten minutes of this vibration induces 54,000 cycles of a stimulus that is 10 times as large as the 90-Hz signal that arises during quiet standing[9] and represents an order of magnitude increase in the strain energy induced at that frequency over a 12-h period.[10] Whether this stimulates adaptation because of some preferential sensitivity of bone cells to higher frequency biophysical signals,[46,47] a threshold of stochastic noise that has been exceeded,[48,49] or by an intrinsic sensory system within the musculoskeletal system tuned to a specific "window" of frequency, such as that achieved by Pacinian or Meisner corpuscles,[50] is not yet known. Alternatively, this "increase" may disrupt the $1/f$ power:law relationship of bone strain history,[51] stimulating adaptation in a self-organized system, or through some other, as yet unidentified physical mechanism. Certainly, an "other than peak" perspective is used in several biological systems subject to exogenous stimuli, such as vision, touch, and hearing.[52]

The results of this RCT also indicate that these low-level signals are perhaps more important than the large, albeit infrequent, signals that the bones of these children are subjected to during limited ambulation, and perhaps serve as a surrogate for dysfunctional musculature. Indeed, that these very low-level signals influence bone mass and morphology also indicates the important role of long-term activities, such as standing, in defining skeletal architecture. Until

recently, only high-intensity exercise[53,54] and parathyroid hormone treatment[36] have been shown to have an anabolic effect on the skeleton; neither has been investigated in children with clinical conditions. The long-term effects of current pharmaceutical treatments on the skeletal health of children are unknown; for example, bisphosphonates have the potential of producing iatrogenic osteopetrosis,[55] and therefore, an alternative non-pharmacological treatment might be preferential. Clearly, in disabled children, implementing high-intensity exercise programs is not viable, and therefore, the tolerance of the vibrating devices offers a unique and non-pharmacological way of improving bone health. Preliminary results in a postmenopausal population, using similar but lower magnitude mechanical signals, also showed efficacy in preventing bone loss.[56]

This pilot RCT was primarily designed to investigate the effects of mechanical stimulation on vTBMD of the spine and tibia. Trabecular BMD was chosen as the primary outcome measure because it has faster turnover than cortical bone[57] and would therefore be more likely to respond to intervention in the short duration of this RCT. However, in our experience, most fractures in children with neurodevelopmental disabilities occur at diaphyseal sites. We therefore undertook posthoc analysis to investigate whether the short period of intervention resulted in increases in tibial diaphyseal bone parameters, which are related to the bending strength of a long bone (area, circumference, polar moment of inertia, cortical thickness) and muscle area. None of the diaphyseal parameters measured showed a response to intervention. There are several possible explanations for this: (1) the heterogeneity in age (pubertal stage) of the subjects within the intervention and control groups will have resulted in different sites of measurement between subjects (consistent in individuals) for these posthoc analyses, which might have masked any effects of intervention; (2) the low compliance; and (3) response to these stimuli may be site-specific to trabecular bone. The site specificity of the response has already been shown in animal studies, where trabecular bone responded to the mechanical stimuli, whereas the cortical bone did not.[58] Finally, a longer period of mechanical stimulation might be required to induce changes in diaphyseal cortical bone, and muscle geometry parameters and scans might more appropriately have been performed consistently at the 50% length mid-diaphyseal site (10-mm section).

The success and completion of this pilot RCT in a challenging group of disabled children depended on using 3-D–QCT as opposed to conventional bone densitometry techniques, which were inappropriate or impossible because of the nature of the patient group. DXA has a number of limitations. First, it provides "areal" BMD (g/cm^2), which does not fully account for changes in bone size in growing children.[59] Furthermore, in this group of children, inability to flatten the limbs because of contractures would have caused projectional inaccuracies in area measurement. Peripheral QCT (pQCT) only permits the acquisition of narrow slice widths ($\leq$2 mm), which would have posed difficulties in the precise relocation of the ROI in follow-up; a small difference in the slice location significantly alters the values of measured parameters.[60] 3-D–QCT enabled rapid acquisition of a block of data, allowing the scan sections at follow-up to be matched to those at baseline, and separate analysis of cortical and trabecular vBMD to be performed.[61] Therefore, while the effective radiation dose associated with 3-D–QCT scans (85 μSv, approximately the same as four chest radiographs[62]) was higher than that associated with the other techniques, we believe that the benefits of QCT far outweighed the potential risks associated with higher effective radiation dose.

There are a number of shortcomings of this pilot trial, which include (1) the subjects were a very heterogeneous group with respect to the medical/genetic conditions from which they suffered; and 2) after randomization, there was an imbalance between the pubertal stages in the intervention and placebo groups. Randomization to intervention/control was blinded, and groups were matched using spinal BMD z-scores to control for age; matching the groups for tibial z-scores might have been preferable, but because this is a novel site for application of QCT, there are no reference data available for calculation of tibia BMD z-scores. The imbalance between the two groups was taken into account by performing unadjusted analyses, which again showed a significant effect of treatment. Additionally, none of the covariates in the analysis of covariance (ANCOVA) model were found to have a strong association with change in vTBMD at either the tibia or spine. Despite these limitations, we have successfully carried out the trial in a challenging group of disabled children. Our results show a magnitude of change in trabecular bone that is in agreement to that reported in animal studies and that was achieved in controlled conditions.

In summary, this pilot RCT in children with disabling conditions provides clear evidence that short durations of extremely low-magnitude, high-frequency mechanical loading can significantly increase vTBMD of the proximal tibia, with a positive trend observed in the spine. A longer and more adequately powered trial in a more homogenous group of children with outcome measures that include diaphyseal cortical bone and muscle geometry parameters of a weight-bearing long bone need to be performed to fully evaluate the efficacy of this intervention. Nevertheless, these data are indicative of the potential of this unique, biomechanically based intervention to offer a non-pharmacological, noninvasive method to increase low trabecular BMD in humans.

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