# Relevance of Whole-Body Vibration Exercises on Muscle Strength/Power and Bone of Elderly Individuals

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#### Abstract

Beneficial effects are associated with whole-body vibration exercises (WBVEs). Increases in muscular strength/power, flexibility, and gait speed; improvements in bone mineral density, balance, and the quality of life; and decreased pain and risk of falls are reported. The aim is to present a review about the importance of WBVE for elderly individuals, considering clinical studies and meta-analyses, on bone and muscle strength/power. There is evidence supporting beneficial effect of WBVE in postmenopausal women (PW); however, effects in PW with osteoporosis are unclear. Age-related decrease in muscle mass and function contribute to undesirable health conditions, including death risk. The WBVEs improve muscle strength/power, functional independence measure, balance, and various fall risk factors, and mobility, measured by Timed Up and Go test, increased significantly after WBVE. An explanation for the absence of positive effects in some outcomes could be related to discrepancies in WBVE protocols as well as the populations tested. It is concluded that WBVE is effective for counteracting the loss of muscle strength associated with sarcopenia in elderly individuals. Balance and leg and plantar flexor strength improvements due to WBV indicate benefit to reduce risk and incidence of falls, frailty, and fracture risks. However, long-term feasibility of WBVE for musculoskeletal and bone health in elderly individuals needs further investigation.

#### **Keywords**

whole-body vibration, elderly, postmenopausal women, bone, muscle strength/power

### Introduction

Tissues and organs of our bodies naturally produce mechanical vibrations that are associated with various important functions, such as (1) heart rate, (2) lung movement, (3) peristaltic movement of the digestive tract, (4) contractions of the blood and lymphatic vessels, and (5) muscular contractions.<sup>1</sup> In addition, during our daily activities, we are exposed to mechanical vibrations from the external environment, which can aid our bodies to perform metabolic functions efficiently.<sup>2,3</sup>

### Transmission of Mechanical Vibration to a Body

In healthy individuals, it is expected that the transmission of mechanical vibrations from the environment to the body can occur in (1) simple activities, such as walking or running due to the impact of the feet on the ground and (2) sports, such as kicking a soccer ball or hitting/catching a ball with the hands (handball, basketball, kitesurfing, or volleyball), skiing, snow-boarding, cycling, and tennis (where a racquet is used to hit in a ball).<sup>2,4</sup> In some occupational activities, the vibration of the

equipment is also transmitted to the body of the individuals, as in the case of the drivers of buses, truckers, tractors, and other types of machinery in farms and/or cities and pilots of helicopters and airplanes.<sup>5-9</sup>

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When there is exposure to physical agents, such as mechanical vibration to a body, and energy is delivered to the biological systems, physiological stresses can be generated leading to expected and unexpected consequences. The relevance of mechanical vibration for human beings is well documented, and a healthy individual is usually exposed to it in normal daily activities. However, in individuals with disability and/or disease, this exposure does not occur naturally during their activities of daily living due to their limitations. In this case, a device that could generate mechanical vibration to be transmitted to the body of an individual would be desirable as the vibration platform.<sup>10,11</sup>

### Mechanical Vibration and Whole-Body Vibration Exercises

Russian researchers, in the half of the last century, observed that astronauts who were on a special mission had a decrease in muscle and bone mass due to the zero gravity, and a vibration platform was developed to treat these astronauts.<sup>12</sup> The possibility of using sources of mechanical vibration as a clinical intervention was initiated. These platforms generated oscillatory and sinusoidal mechanical vibrations with controlled frequencies and peak-to-peak displacement, resulting in whole-body vibration exercise (WBVE) in the individual who is in contact with the base of the platform.<sup>13</sup>

The mechanical vibration transmitted to the body continues through a variety of different biological structures, such as muscles, bones, cartilages, synovial fluids, and joints. Depending on the characteristics of each of these structures and the biomechanical parameters of the mechanical vibration, such as frequency, peak-to-peak displacement, and peak acceleration, the energy of the mechanical vibration is dampened.<sup>10,13</sup>

# Possible Mechanisms Associated With the Biological Effects of the WBVE and Applications

The observed biological effects of WBVE would be partly due to the stimulation of neuromuscular responses in conjunction with the hormonal and nonhormonal signaling pathways.<sup>14</sup> Moreover, a complex spinal and supraspinal neurophysiological mechanism, the tonic vibration reflex (TVR), could also be involved in the response to the mechanical vibration.<sup>10,15</sup>

Various beneficial effects have been associated with WBVE, and neuromuscular responses have been described. Increases in muscular strength/power, flexibility, and gait speed; improvements in bone mineral density, balance, blood flow, cognition/executive functions, and quality of life; and decreased pain and risk of falls have been reported.<sup>10,16,17</sup> Moreover, different populations, from children to elderly individuals have been exposed to WBVE. Furthermore, individuals with specific clinical disturbances, such as cases of obesity, sarcopenia, diabetes, metabolic syndrome, chronic kidney disease, fibromyalgia, Parkinson disease, stroke, multiple sclerosis, cerebral palsy, Duchenne muscular dystrophy, osteogenesis imperfecta, osteoarthritis, attention-deficit hyperactivity

disorder, and chronic obstructive pulmonary disease, have been treated with WBVE.<sup>10,16,18,19</sup> Besides the beneficial effects, undesirable and unpleasant side effects due to WBVE have been documented.<sup>10,20</sup>

As life expectancy continues to increase, it is expected that the proportion of elderly populations in the world will continue to expand.<sup>21</sup> The WBVE also has been a suitable and efficient strategy with reduced cost for the management of several unhealthy age-related conditions.<sup>10</sup> A search performed in the database PubMed on June 25, 2018, with the key words whole-body vibration and elderly, has found 551 articles, which correspond to 28.15% of all the publications with the key word whole-body vibration. The aim of this work, therefore, is to present a narrative review about the importance of the WBVE for the elderly individuals, considering clinical studies and meta-analyses effects on bone and muscle strength/power.

### Effect of WBVE in Postmenopausal Women

It is well known that bone adapts to mechanical loading by increasing its density and altering its geometry and microarchitecture, resulting in increased strength.<sup>22</sup> Since WBVE induces mechanical oscillations that are sensed by bone cells,<sup>23</sup> there is growing interest in the potential for WBVE to be used as a nonpharmacologic therapy for improving bone health especially in postmenopausal women (PW) who are at increased risk for osteoporosis.

A PubMed search using the key words "whole body vibration" and "bone" and "postmenopausal women" resulted in 47 articles, including 21 clinical trials, 12 review articles, and 4 meta-analyses. The meta-analyses are recently published in 2016 and 2017, <sup>24-27</sup> and they analyze the bulk of the randomized clinical trials (RCTs) on this topic. Therefore, this section will focus on the meta-analyses results that are informative on the effects of WBVE characteristics (frequency, magnitude, type of platform, and body posture) on bone mineral density (BMD) in this population (Table 1). A 2018 RCT not included in the meta-analyses also will be discussed.

Ma et al<sup>24</sup> determined that WBVE groups had a significant positive effect on lumbar spine BMD (weighted mean difference 0.01 g/cm<sup>2</sup>) compared to the control groups; however, no benefit was found for the femoral neck BMD site. They also reported this significant lumbar spine effect occurred for lowmagnitude (< 1 g) WBVE protocols but not for high-magnitude ( $\geq$ 1 g) protocols. It should be noted that this meta-analysis included studies with anti-osteoporosis drugs/therapies such as vitamin D, calcium supplementation, and alendronate. Also, the control groups were a mix of conditions, including a sham, other types of exercise, or no treatment at all.

Luo et al<sup>27</sup> conducted a meta-analysis of WBVE RCT studies  $\geq 12$  weeks in duration that targeted PW diagnosed with osteoporosis. Based on their analysis, effect sizes for BMD outcomes (lumbar spine, femoral neck, and total hip) in WBVE groups did not differ from the control groups. They attributed the lack of effect on large methodological differences in the

Study	Inclusion Criteria	Target Population	Outcome Variables	Total Sample Size, n	Number of Articles Included
Ma et al (2016) <sup>24</sup>	$RCT \ge 6$ months (included drug treatments)	PW	BMD: Lumbar spine and femoral neck	1014	8
Luo et al (2017) <sup>27</sup>	$RCT \ge 12$ weeks (WBVE only, not combined with other types of exercise)	PW diagnosed with osteoporosis	BMD: Lumbar spine, femoral neck, trochanter, and total hip	625	9
Oliveira et al (2016) <sup>23</sup>	$RCT \ge 6 \ months$	PW	BMD: Lumbar spine, femoral neck, trochanter, total hip	1833	17 in qualitative review and 15 in meta-analysis
Fratini et al (2016) <sup>22</sup>	RCT and CCT (WBVE with or without other types of exercise, no drug treatments)	PW	BMD: Site-specific effect sizes not reported	527	9

Table I. Characteristics of Meta-Analyses on Whole-Body Vibration Exercise and Bone Mineral Density in Postmenopausal Women.

Abbreviations: BMD, bone mineral density; CCT, controlled clinical trial; PW, postmenopausal women; RCT, randomized clinical trial; WBVE, whole-body vibration exercises.

WBVE protocols (eg, type of platform, frequency, amplitude, and magnitude).

Oliveira et al reported a significant WBVE BMD effect for the lumbar spine but not for the total hip, radius, or tibia sites in their primary analysis of 15 RCTs >6 months in duration. They also conducted interesting subgroup analyses for WBVE protocol characteristics, including magnitude, frequency, body position, and type of vibration platform. They found that high-magnitude (>1 g), low-frequency (<20 Hz) WBVE protocols were effective for both lumbar spine and trochanter BMD, whereas high-frequency (>20 Hz), low-magnitude (<1 g) protocols had positive effects only on the lumbar spine BMD. Significant effect sizes for lumbar spine, femoral neck, and trochanter BMD sites occurred for protocols implementing a semi-flexed knees (FK) posture but not for straight leg protocols. Also, side-alternating vibration (SV) platforms were effective for improving lumbar spine and trochanter BMD; however, synchronous platforms had no significant effects on BMD. They also commented that ranges of effect sizes in the RCT analyzed (lumbar spine = 0.004-0.016 g/cm<sup>2</sup>, femoral neck =  $0.040 \text{ g/cm}^2$ ), although small, are comparable to other types of treatments (eg, pharmacological, supplements, and traditional exercise).

Fratini et al<sup>25</sup> reported similar findings as Oliveira et al<sup>26</sup> in their meta-analysis, which included RCT and controlled clinical trials. They concluded that WBVE interventions significantly improved BMD from 2% to 5.5% at the hip and spine compared to placebo-controlled groups. The WBVE protocols that utilized high-magnitude (>3 g), low-frequency (12.5-20 Hz), and side-alternating platforms were more effective for improving BMD. They also reported that both full straight leg and FK postures on the platform were effective but that performing exercises while standing on the platform did not improve BMD variables.

The findings of these meta-analyses provide valuable guidance for investigators designing RCT in PW. Recently, de Oliveira et al<sup>28</sup> compared the effects of a 6-month WBVE protocol in PW. The vibration intervention utilized a lowfrequency (20 Hz), high-magnitude (3.2 g) protocol performed on an SV platform with a semi-FK posture 3 times per week. The WBVE group had significant increases in lumbar spine and trochanter BMD versus the control group, supporting the efficacy of these WBVE characteristics for enhancing bone health in this population.

# Effects of WBVEs on Muscle Strength/Power in Older People

Age-related changes in muscle function. Authors have reported that age-related decreases in muscle mass and function contribute to increased frailty, risk of falls and fractures, impaired mobility, balance, gait speed, and activities of daily living resulting in loss of independence and other age-related chronic disorders, as osteoporosis, sarcopenia, dynapenia, osteoarthritis, type 2 diabetes, insulin resistance, obesity, cardiovascular diseases, and risk of death.<sup>29,30</sup>

In addition, it is known that sarcopenia and central obesity are both age-related body composition changes, and they are often combined in older individuals. Many older individuals who exhibit frailty have sarcopenia, and many people with sarcopenia exhibit frailty syndrome.<sup>31</sup>

# Effects of WBVE on Clinical Conditions Associated With Age-Related Impairments in Muscle Strength/Power

Physical exercise is important for the management of undesirable clinical conditions due to age, and WBVE could be an important intervention due to its safety and efficacy. Moreover, WBVE is associated with reduced health-care costs.

Considering the key words whole body vibration and muscle strength and power and elderly in the PubMed on June 27, 2018, eleven articles were included in this review. From these studies, those that used WBVE as the only intervention were

Study	Total Sample Size, n/Years Old	Sex	Duration of the Intervention, Weeks	Frequency/PPD or A	Number of session/Week
Santin-Medeiros et al (2017) <sup>32</sup>	37/ 82.4 ± 5.7	Women	32	20 Hz/2 mm (PPD)	2
Smith et al (2016) <sup>35</sup>	60/ 82.2 ± 4.9	36 women/24 men	12	30 Hz/1-2 mm	2
Corrie et al (2015) <sup>34</sup>	61/80.2 ± 6.5	37 women/24 men	12	30 Hz/1.3 mm (PPD)	3
Perchthaler et al (2015) <sup>36</sup>	51/55 ± 8	Men	Acute	6-30 Hz/1.3-3.9 mm (A)	Acute
Cristi et al (2014) <sup>38</sup>	16/older(ANR)	7women/9 men	9	30-45/2 mm (PPD)	3
Giombini et al (2013) <sup>37</sup>	9/71.0 ± 3.0	Women	Acute exposition	20-50Hz	Acute
Mikhael et al (2010) <sup>39</sup>	19/50-80	Women/men	13	12 Hz/1 mm(A)	3
Machado et al (2010) <sup>40</sup>	26/76-79	Women	10	20-40 Hz/2-4 mm(A)	3-5
Rees et al (2008) <sup>41</sup>	30/73.7 $\pm$ 4.6	14 women/16 men	8	26 Hz/5-8 mm(A)	3

Table 2. Some Characteristics of the Analyzed Publications Involving Whole-Body Vibration Exercises on Muscle Strength/Power.

Abbreviations: A, Amplitude; ANR, age not reported; PPD, peak to peak displacement.

selected. One of these articles did not report any improvement in the population that was tested.<sup>32</sup>

Table 2 shows some characteristics of the analyzed publications. The number of the individuals in the studies varied from 6 to 60 and the age ranged from 47 to 88.1 years old. There were studies with individuals of both sexes. The duration of the interventions with WBVE varied from acute exposure (1) to 13 weeks.

# Studies With Improvements in Some Outcomes due to WBVE in Elderly Individuals

Findings of the WBVE studies that reported some positive results in the elderly individuals are discussed. Smith et al<sup>35</sup> allocated the individuals into bioDensity (bD), Power Plate (PP), whole-body vibration (WBV), or bD+PP groups. The bD groups performed once weekly 5-second maximal isometric contraction of 4 muscle groups. The PP group involved two 5-minute WBV sessions. Primary outcomes were strength, balance, and a Functional Independence Measure (FIM). Balance significantly improved in PP and bD+PP but not in control or bD. The bD, PP, and bD+PP differentially improved FIM selfcare and mobility. Corrie et al<sup>34</sup> evaluated the influence of vertical vibration (VV) and SV on musculoskeletal health in older people at risk of falls in a WBV training to sham vibration (Sham). Falls risk factors and neuromuscular tests were assessed and blood samples collected for determination of bone turnover markers at baseline. There were significantly greater gains in leg power in the VV than in the Sham group and in bone formation in SV and VV compared to the Sham group. Perchthaler et al<sup>36</sup> documented that after a WBV intervention. jump height increased by 18.55% in the WBV group, whereas values of the control group remained unchanged. The subjective perceived exertion of the WBV exercises and respective training parameters ranged between moderate rating levels of 7 and 13 of Borg scale. Giombini et al<sup>37</sup> measured the maximal power output during a double leg press on an isoinertial dynamometer at 1 and 5 minutes after WBV on a platform at 3 different frequencies in a random order: 20 Hz, 50 Hz, and optimal vibration frequency (OVF). The mean (standard deviation) OVF was 33 (2.5) Hz. The 25.9% increase in maximal

power output after 1 minute of WBV at OVF was significantly higher than the 14.3% increase after 1 minute of WBV at 20 Hz. Similarly, the 32.1% increase in maximal power output after 5 minutes of WBV at OVF was significantly higher than the 16.1% and 16.3% increase after 5 minutes of WBV at 20 Hz and 50 Hz, respectively.

Cristi et al<sup>38</sup> observed that WBV significantly improved several tests that evaluate physical fitness, such as 30-second chair stand, arm curl, or chair sit and reach test. There was a significant increase in maximal voluntary isometric contraction (MVIC) between pre- and posttraining conditions. Muscle power values, reached at 20%, 40%, and 60% MVIC, were also significantly greater after training. However, messenger RNA or protein levels for C-reactive protein, interleukin 6, interleukin 1 $\beta$ , tumor necrosis factor- $\alpha$ , and interleukin 10 did not significantly differ from basal values.

Mikhael et al<sup>39</sup> examined the effects of standing posture during low-magnitude WBV training on muscle function and muscle morphology in individuals exposed to WBV with FK, WBV with locked knees (LK), or sham WBV with FK (Control-CON). Relative (%) upper body contraction velocity significantly improved after WBV with FK compared to LK (FK 16.0%, LK 7.6%, CON 4.7, P = .01). Relative upper body strength (LK 15.1%, P = .02; FK 12.1%, P = .04; CON 4.7%) increased significantly following WBV compared to control. Potentially clinically meaningful but statistically nonsignificant improvements in lower leg muscle cross-sectional area (LK 3.7 cm<sup>2</sup>, FK 2.4 cm<sup>2</sup>, CON 2.2 cm<sup>2</sup>, P = .13) were observed after WBV with LK compared to the other groups.

Machado et al<sup>40</sup> observed that MVIC increased 38.8% in the WBV group, without changes in the control group. Electromyographic activity of the *vastus medialis* (VM), the *vastus lateralis*, and the *biceps femoris* (BF) did not change in either group. Thigh muscle cross-sectional area increased significantly after training in VM (8.7%) and BF (15.5%). Muscle power at 20%, 40%, and 60% MVIC decreased from pretest to posttest in the CON group; however, WBV training prevented the decrease in the WBV group. Consequently, mobility, measured by the Timed Up and Go test, increased significantly after training (9.0%) only in the WBV group. Ten weeks of lower limb WBV training in older women produced a significant increase in muscle strength induced by thigh muscle hypertrophy, with no change in muscle power. Rees et al<sup>41</sup> reported a significant improvement in ankle plantar flexor strength and power in the WBVE group compared to the exercise group.

# Studies With No Improvements in Outcomes due to WBVE in the Elderly Individuals

In a study by Santin-Medeiros et al<sup>32</sup> in which elderly individuals were exposed to WBVE, positive findings were not reported. These authors aimed to determine the effects of WBV training on health-related quality of life (HRQoL), nonsignificant changes in HRQoL, and additional health-related outcomes (fall risk, life satisfaction, or cognitive status) were found. In addition, Corrie et al<sup>34</sup> found that WBV training did not promote changes in fall risk factors, and the intervention did not provide any additional benefits for balance or fall risk factors beyond a fall prevention program in older people at risk of falls. Pertchtaler et al<sup>36</sup> also observed no statistically significant differences in isokinetic maximal strength, mean power, or work values in knee extension or flexion in elderly individuals exposed to WBV exercise. Mikhael et al<sup>39</sup> reported no significant effects of WBV on any functional performance tests (muscle morphology, balance, habitual and maximal gait velocity, stair climb power, and chair stand performance) in a population of elderly individuals.

A possible explanation for the absence of positive effects due to WBV exposure in some studies and in some outcomes could be related to discrepancies in WBV protocols as well the populations tested.

### Discussion

Due to the increase in life expectancy, some public health conditions related to the impairment of the bone and mass muscle have been also documented, such as osteoporosis, sar-copenia, and dynapenia.<sup>29,30,33,42,43</sup> In general, the different biological/clinical effects of the WBVE<sup>10,15,16</sup> are highly relevant to elderly individuals.

## Some Thoughts About Fractures, Bones, and Muscles in Older Age

It is commonly stated that most fractures occur late in life due to low bone density.<sup>44-46</sup> As aging is associated with bone loss, it has been reasoned that people with fractures and low bone density have lost more bone during life than people who do not have fractures.<sup>45,47,48</sup> However, it could also be discussed that people with fractures in older age may have achieved a lower peak bone mass in young adulthood and lost bone at the same rate and for the same length of time as other people.<sup>49</sup> If low peak bone density was an important contributor to reduced bone density of elderly people with fractures, then the development of optimal peak bone mass during childhood and adolescence would be of crucial importance for the prevention of

osteoporosis and consecutively fractures. However, a study on soccer players showed that exercise in youth results in high and clinically relevant peak BMD, but cessation of exercise led to accelerated bone loss.<sup>50</sup> Thus, those who stopped playing for more than 35 years and who were aged 60 or older had no significant residual benefit of bone density and no reduced fracture rate than expected. Therefore, short-term effects on bone mass (regardless if positive or negative) do not seem to have any long-term effects. The skeletal system seems to adapt to the current requirements. Therefore, strong bones in children do not automatically lead to fracture-free old age. It might be possible that strong bones keep strong if an individual maintains the healthy lifestyle that made the bones strong in the first place. Improving bone parameters should be derived from the knowledge on the interaction of muscles and bone: A positive influence on bone can be expected through functional muscle activation. It is expected that mechanical vibration can aid in achieving this goal.

## Effects of WBVE on BMD in PW

Based on recent meta-analyses, high-magnitude (>1 g), lowfrequency (<20 Hz) WBVEs performed on SV platforms are effective for improving lumbar spine and hip BMD outcomes in PW. The lumbar spine is the BMD site most consistently responsive to WBVE protocols, possibly related to its higher relative trabecular bone composition compared to the hip BMD sites.<sup>51</sup> Regarding body posture, both semi-FK and straight leg stances had positive BMD effects, but BMD outcomes were not affected by performing simultaneous exercises while standing on the vibration platform.<sup>25</sup> There is evidence in the literature supporting a role for WBVE as a nonpharmacological intervention for osteoporosis prevention in PW; however, the benefits for those diagnosed with osteoporosis is not yet clear. The WBVE research to date provides guidance for designing effective protocols for bone health; there is a need for more RCT targeting the effects of WBVE protocols in individuals clinically diagnosed with osteopenia/osteoporosis.

# Consequences of WBVE for Muscle Strength/Power in Older People

Considering the studies selected related to the effects of WBVE on the impairment of muscle strength/power in older people, important variability in the parameters used in the works was identified. The number of individuals varied from<sup>37</sup> 9 up to  $61.^{34}$  Moreover, in 3 papers,<sup>32,37,40</sup> the individuals were only women, while in one<sup>36</sup> there were only men, and in another study there were both men and women. The duration of the intervention varied from 1 session per week<sup>36,37</sup> to 32 weeks,<sup>32</sup> while the frequency was from 6 Hz<sup>36</sup> up to 50 Hz<sup>37</sup> and the amplitude from 0.65 mm<sup>34</sup> up to 8 mm.<sup>41</sup> Indeed, relevant and significant findings were shown. Cristi et al<sup>38</sup>concluded that WBV training was useful for counteracting the loss of muscle strength associated with sarcopenia in older adults. This is in agreement with Smith et al<sup>35</sup> who suggested that balance and leg strength improvements with WBVE indicate that this physical intervention beneficially impacts risk and incidence of falls. Machado et al<sup>40</sup> suggested that the adaptations to the WBVE may be of use in counteracting the loss of muscle strength and mobility associated with age-induced sarcopenia. Rees et al<sup>41</sup> concluded that vibration training contributed to an increase in plantar flexor strength and power in a population of elderly individuals.

In a meta-analysis, Rogan et al<sup>52</sup> pointed out that results suggest that WBV can be used as a skilling up exercise in participants not able to perform standard exercises. However, it is suggested that further studies with the various types of WBV in different subpopulations of elderly persons are needed to determine the most effective vibration modes.

Although the ages of the recruited individuals of the selected studies are similar, some limitations should be mentioned in this review. The number of individuals as well as sex in the groups of the studies varied. Moreover, the biomechanical parameters, such as frequency and amplitude of the mechanical vibration, had an important variability. The number of the WBV sessions and how long were the interventions also have differences among the studies selected.

### Conclusion

Although some limitations must be considered, it is possible to conclude that WBV training is effective for counteracting the loss of muscle strength associated with sarcopenia in older adults. Balance and leg and plantar flexor strength improvements due to WBV indicate that this physical intervention beneficially impacts risk and incidence of falls, frailty, and fracture risks. There is evidence in the literature supporting a beneficial effect of high-magnitude, low-frequency WBVE performed on side-alternating platforms in PW. However, the efficacy, mechanism of action, and long-term feasibility of WBVE for musculoskeletal and bone health in older adults warrants continued investigation in robustly designed and sufficiently powered future studies.

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