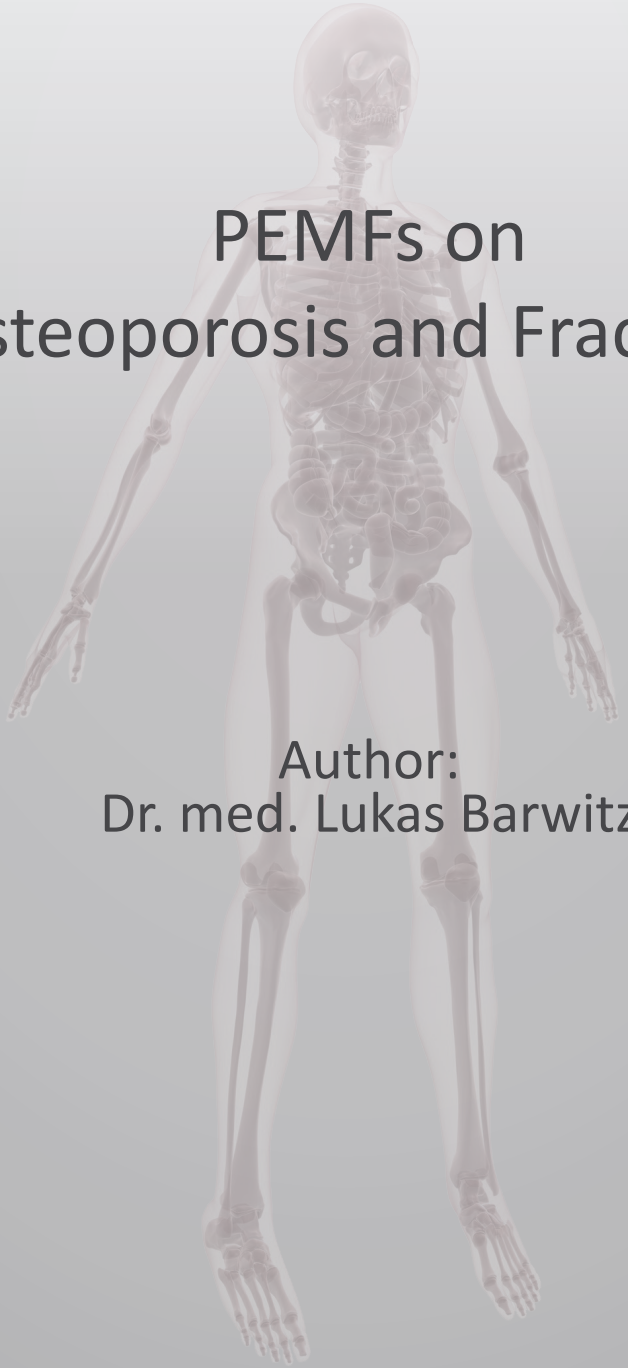
SWISSBIONIC SOLUTIONS

LIFESTYLE

Bone Metabolism

PEMFs on Osteoporosis and Fractures



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Bone metabolism is a continual cycle of bone growth and resorption that is carefully orchestrated by the dynamic relationship between osteoclasts, osteoblasts and an array of hormonal and regulatory influences. The relative levels of these signaling molecules dictate whether healthy, balanced bone metabolism ensues. Disturbances to this delicate equilibrium where resorption is greater than growth can weaken the skeletal architecture and put one at risk for the development of chronic and debilitating diseases such as Osteoporosis.

Osteoporosis

Osteoporosis is a disease where bone mass atrophy and the destruction of the microarchitecture of bone tissue increases the risk of fractures. It becomes more common with age and is the most common reason for a broken bone among the elderly. About 15% in their 50s and 70% of those over 80 are affected; it is more common in women than men.

Bones that commonly break include the vertebrae, the bones of the forearm, and the hip. Until a broken bone occurs there are typically no symptoms. Bones may weaken to such a degree that a fracture may occur with minor stress or even spontaneously. Chronic pain and a decreased ability to carry out normal activities may follow a fracture.

Bone loss increases after menopause due to lower levels of estrogen. Osteoporosis may also occur due to a number of diseases or treatments, including alcoholism, anorexia hyperthyroidism or kidney disease.

Certain medications increase the rate of bone loss, including some antiseizure medications, chemotherapy, proton pump inhibitors, selective serotonin reuptake inhibitors, and glucocorticosteroids. Smoking and too little exercise are also risk factors. Osteoporosis is defined as a bone density of 2.5 standard deviations below that of a young adult. This is typically measured by dual-energy X-ray absorptiometry.

Efforts to prevent broken bones in those with osteoporosis include a good diet, exercise, and fall prevention. Lifestyle changes such as stopping smoking and not drinking alcohol may help. Studies showed that Biphosphonate medications are useful in those with previous broken bones due to osteoporosis. In those with osteoporosis but no previous broken bones, they are less effective. A number of other medications may also be useful.

History of PEMFs on bone repair

In 1892, Wolf indicated that mechanical stress determines bone growth and remodeling[i]. In 1953, Yasuda revealed that bending the long tubular bone is related with the development of electric currents and this instance is defined as piezoelectric phenomenon[ii]. Since then, the theory that electrical stimulation is the path for bone formation in response to applied load has been gradually recognized, and various devices have been developed to produce

electrical stimulation for promoting the healing of bone fracture. In 1978, Bassett first applied noninvasive PEMFs to treat delayed union or non-union fractures and have achieved good clinical effect[iii]. Shortly thereafter, PEMFs were approved as a safe and effective method for treating delayed union or non-union fractures by the US Food and Drug Administration (FDA) [iv,v].Inductive coupling is the rationale for the application of PEMFs[vi]. PEMF devices consist of a wire coil wherein a current passes and a pulsed magnetic field is generated. The pulsed magnetic field, in turn, induces a time-varying secondary electrical field within the bone. The secondary electrical field is dependent on the characteristics of the applied pulsed magnetic field and the tissue properties. Magnetic fields of 0.1–20 G are usually applied to produce electrical fields, ranging from 1 mV/cm to 100 mV/cm in the bone[vii]. Through the PEMF device, a time-varying electrical field is produced to simulate the normal response of bone cells physiologically to the applied mechanical stress[viii], and the subsequent enhanced growth and remodeling bioeffects on the bone are initiated by the time-varying electrical field.

PEMFs on Osteoporosis and Fractures

In numerous studies PEMF could demonstrate its positive effects on bone metabolism and in the healing of fractures by stimulating osteoblasts, calcium uptake and its mineralization in vitro and in vivo and is therefore a suitable addition for the treatment of osteoporosis and fractures[ix,x,xi,xii,xiii,xiv,xv,xvi,xvii,xviii,xix].

What exactly happens on a cellular level?

Recently, considerable research progresses have been made in exploring the underlying cellular and subcellular mechanisms of PEMFs promotion effect in bone repair. Several key signaling pathways during the osteogenesis and angiogenesis which are two essential aspects for bone repair, were revealed by various studies when the bone was exposed to PEMF including Ca^{2+}, Wnt/β-catenin, mitogen-activated protein kinase (MAPK), fibroblast growth factor (FGF) and vascular endothelial growth factor (VEGF), transforming growth factor (TGF)- β/bone morphogenetic proteins (BMP), insulin-like growth factor (IGF), Notch, and cAMP/protein kinase A (PKA)[xx].

For those still interested in the very detailed signaling pathways during the osteogenesis and angiogenesis that PEMF takes influence on I recommend **Yuan J et al. 2018. Underlying Signaling Pathways and Therapeutic Applications of Pulsed Electromagnetic Fields in Bone Repair**[xx] and quote:

Ca2+ Signaling

Intracellular Ca^{2+} is generally considered as one of the main actors to translate the PEMF signal into a biological signal. Many studies revealed that PEMF signal passes through the cell membrane to set up a time-varying electrical field within the cytosol; this electrical field subsequently induces the release of intracellular Ca^{2+}, leading to increases in cytosolic calcium, activated calmodulin and the enhancement of bone cell viability[viii,xxi,xxii].
Voltage-gated Ca channels (VGCCs), especially the L type, play a pivotal role in intracellular Ca^{2+} release. PEMF exposure significantly elevated the expression levels of VGCCs in mesenchymal stem cells (MSCs) during osteogenesis[xxiii,xxiv]. PEMF-initiated Ca^{2+} signaling strikingly accelerates the osteogenic differentiation of MSCs as represented by the upregulated osteogenic markers, such as collagen I and ALP, and the increased deposition of extracellular calcium[xviii]. Accumulated studies indicated that increased intracellular Ca^{2+} caused by PEMF stimulation leads to increased nitric oxide levels, which in turn increases the synthesis level of cGMP and the subsequent activation of protein kinase G. Through the Ca^{2+}/nitric oxide/ cGMP/protein kinase G pathway, PEMFs promote osteoblast differentiation and maturation, exert their therapeutic effect on bone repair, and remarkably reduce the pain of patients by modulating the release of inflammatory cytokines, such as interleukin-1 beta (IL-1β)xvi[xvi,xxv,xxvi,xxvii,xxviii,xxix]. Moreover, the activated Ca^{2+}/nitric oxide/cGMP cascade is also closely related to the increased expression of FGF-2 and VEGF, two key regulators of angiogenesis[xxiii]. In addition, the crosstalk between Ca^{2+}, ERK, PKA, and PKG signaling under PEMF stimulation was also reported[xv,xviii]. All these findings show the prominent role of Ca^{2+} signaling in PEMFs-induced bone repair.

Wnt/β-catenin signaling pathway

Extracellular Wnt ligands bind to their seven-pass transmembrane Frizzled receptors simultaneously with a co-receptor of the arrow/Lrp family (e.g., LRP5 and LRP6), thus stabilizes β-catenin in the cytoplasm and initiates the canonical Wnt/β-catenin signaling pathway[xxx]. This signaling pathway is conserved throughout metazoans and is essential for cell proliferation, differentiation, development, self-renewal, and cell fate determination[xxxi,xxxii].

Much evidence has suggested that the Wnt/β-catenin signaling pathway acts as a key regulator in PEMF-induced osteogenic differentiation of mesenchymal progenitor cells, bone formation and repair. For instance, in vitro assay studies, gene and protein expressions of canonical Wnt/β-catenin signaling pathway, including Wnt1, LRP6, and β-catenin, were all significantly enhanced after PEMF exposure at both proliferation and differentiation stages of osteoblast-like MC3T3-E1 cells[xxxiii]. In addition, except the upregulation of mRNA expressions of Wnt1, Wnt3a, LRP5 and β-catenin in tissue derived mesenchymal stem cells (ADSCs), PEMFs intervention could also reduce the expression of dickkopf1 (DKK1) which usually acts as an inhibitor of Wnt signaling pathway[xxxiv]. Furthermore, the enhanced Wnt/β-catenin signaling induced by PEMFs notably elevated the expression of proliferation phase related target genes,

Ccnd 1 and Ccne 1, and differentiation phase related genes, ALP, OCN, COL1, and Runx2, in osteoblast cells, which accelerated the osteoblasts proliferation, differentiation, and mineralization, three pivotal processes of bone formation[xxvii,xxviii]. On the other hand, according to in vivo assay studies, PEMFs effectively reversed the bone mass loss and deterioration of bone microarchitecture analyzed by microCT and attenuated biomechanical strength deterioration evaluated by three-point bending test in hind limb-suspended ovariectomized rats through the Wnt/Lrp5/β-catenin signal pathway[xxxv,xxxvi], indicating that activating this pathway by PEMF exposure is beneficial for bone disorders[xxxvii].

MAPK pathway

The MAPK pathway is important in the transduction of extracellular signals to various cellular compartments and is involved in cell proliferation, differentiation, migration, and death[xxxviii]. Conventional MAPKs include Erk1/2, JNK, and p38. The MAPK pathway plays a critical role in PEMF-induced osteogenic differentiation and osteoblasts' viability and function. For example, extremely low-frequency pulsed electromagnetic field (ELF-PEMF) treatment could significantly increase the total protein content, mitochondrial activity, and ALP activity and enhance the formation of mineralized matrix of human osteoblasts with a poor initial osteoblast function through triggering the ERK1/2 signaling pathway. When the cells were treated with U0126, an inhibitor of the ERK1/2 signaling cascade, the positive effects of the ELF-PEMF treatment on osteoblast function were abolished[xxxix]. Other studies also revealed that the MEK/ERK signaling pathway regulated the promoting effects of PEMF on bone marrow mesenchymal stem cell (BMSC) proliferation, expression of osteogenic genes (RUNX2, BSP, OPN), ALP activity, and calcium deposition[xviii,xxviii,xl,xli]. Additionally, one study reported that the p38 MAPK pathway is involved in the increased production of collagen synthesis in osteoblast-like cells stimulated by ELF-EMF exposure[xlii]. Interestingly, a recent research suggested that a 45 Hz EMF promoted the osteogenic differentiation of adipose-derived stem cells, whereas a 7.5 Hz EMF directly augmented the expression of osteoclastogenic markers and regulated the osteoclast differentiation through ERK and p38 MAPK activation[xliii]. This finding indicated that PEMFs can simultaneously influence osteoblastic and osteoclastic activities under defined electromagnetic conditions.

FGF and VEGF pathways

Osteogenesis and angiogenesis, including cell–cell communication between blood vessel cells and bone cells, are essential for bone repair. Many studies suggested that PEMFs play a promotion effect not only in osteogenesis but also in angiogenesis[xliv,xlv,xlvi,xlvii]. PEMFs may facilitate bone repair by augmenting the interaction between osteogenesis and blood vessel growth. During this complex process, FGF and VEGF, two key angiogenesis-related cytokines, may play critical regulatory roles. The FGF signaling pathway has been demonstrated to contribute in the regulation of proliferation and differentiation of osteoblasts and

in angiogenesis[xlvii] and the VEGF signaling pathway has also been reported to be involved in a reciprocal, functional, and regulatory relationship between osteoblasts and endothelial cells during osteogenesis[xlix,l,li]. A study indicated that a 150% increase in FGF-2 mRNA and a fivefold elevation of FGF-2 proteins in human umbilical vein endothelial cells (HUVECs) exposed to PEMF were monitored and the release of functional FGF-2 from PEMF-stimulated HUVECs specially increased endothelial cell proliferation and tubulization, processes that are important for vessel formation[lii]. KDR/Flk-1, a tyrosine kinase receptor of VEGF, is autophosphorylated in response to VEGF stimulation and is capable of transducing VEGF signals. One research has revealed that PEMF stimulation significantly increased the expression and phosphorylated levels of KDR/Flk-1 and promoted proliferation, migration, and tube formation of HUVECs[xxxix]. The pro angiogenesis effect through the FGF and VEGF signaling pathways of PEMFs provide another explanation for the therapeutic function of PEMFs in bone repair. Many studies are still required to further clarify the efficacy of FGF and VEGF in PEMF-induced bone repair.

TGF-β/BMP pathway

TGF-βs and BMPs, as multifunctional growth factors, belong to the TGF-β super family. The interaction of TGF-βs/BMPs with TGF-β specific type 1 and type 2 or BMP serine/threonine kinase receptors initiates the signaling cascade via canonical (or Smad-dependent pathways) and non-canonical pathways (or Smad-independent signaling pathways)[liii]. The TGF-β/BMP signaling pathway plays an important regulatory role in bone repair[liv,lv,lvi,lvii,lviii,lix]. It is also confirmed to be involved in PEMF-induced osteogenesis. Several studies demonstrated that PEMF stimulation could significantly increase the expression of TGF-β in both osteoblast-like cells and cells from atrophic or hypertrophic non-unions[xiii,lx,lxi,lxii,lxiii]. Moreover, a recent research suggested that PEMFs activated the TGF-β signaling via Smad2 in differentiated and mineralizing osteoblasts and augmented the expression of osteoblast differentiation marker genes, such as ALP and type I collagen, and exerted its osteogenesis promotional function . The expression of BMPs in osteogenesis was also enhanced by PEMFs according to in vitro and clinical studies[lxv,lxvi.lxvii]. Furthermore, another recent study revealed that PEMFs stimulate osteogenic differentiation and maturation of osteoblasts by primary cilium-mediated upregulated expression of BMPRII, one of the receptors of BMPs, and subsequently activation of BMP–Smad1/5/8 signaling[lxviii]. Given the separate promotional effects on the differentiation and maturation of osteoblasts of BMPs and PEMFs, many studies found that combined BMP and PEMF stimulation would augment bone formation to a greater degree than treatment with either stimulus[lxix,lxx,lxxi,lxxii].

Other pathways

IGF signaling pathway is also an important signaling implicating in osteoblast differentiation and bone formation[lxxiii,lxxiv]. It was reported that PEMFs significantly increase the level of mRNA expression of IGF-1 and promote bone formation in rat femoral tissues in vitro[lxxv]. In addition, IGF-1 in combination with PEMFs augmented cartilage explant anabolic activities, increased PG synthesis, restricted the catabolic effect of IL-1b, and showed a synergistic chondroprotective effect on human articular cartilage[lxxvi]. Another study showed that dexamethasone combined with PEMF upregulated the mRNA expression of IGF-1 and improved dexamethasone-induced bone loss and osteoporosis[lxxvii]. Notch signaling is a highly conserved pathway that regulates cell fate decisions and skeletal development. A recent research advocated that the expression levels of Notch receptor (Notch4) and its ligand DLL4 and nuclear target genes (Hey1, Hes1, and Hes5) were upregulated during the PEMF-induced osteogenic differentiation of hMSCs. Moreover, the Notch pathway inhibitors effectively inhibited the expression of osteogenic markers, including Runx2, Dlx5, Osterix, as well as Hes1 and Hes5, indicating that the Notch signaling plays an important regulatory role in PEMF-induced osteogenic differentiation of hMSCs[lxxviii]. The cAMP/PKA signaling pathway is another signaling involved in the PEMF-induced bone repair. Recent studies have demonstrated that PEMFs notably increased the cAMP level and PKA activity and accelerated the osteogenic differentiation of MSCs[xxvii,xxxiv,lxxix].

Therapeutic applications of PEMF in bone repair

The promotional effects of PEMFs on osteogenesis and angiogenesis in bone repair have been well established in either vitro or in vivo animal studies. Several key-signaling pathways involved in PEMF-induced bone repair were elaborate above. Moreover, several decades of PEMF applications in the treatment of skeletal diseases have clearly proved its potential benefit in augmenting bone repair.

Osteoporosis

Osteoporosis is a worldwide health problem with high morbidity, especially in postmenopausal women[lxxx,lxxxi,lxxxii]. It is generally defined as a systemic skeletal disease characterized by low bone mineral density (BMD) and compromised bone strength, leading to enhanced bone fragility, increased fracture risk, and resultant disability, which strikingly affects patients' quality of life[lxxxiii,lxxxiv]. As PEMFs were verified to be equally effective with mechanical stimulation in maintaining or improving bone mass according to experiments of NASA between 1976 and 1979, many clinical studies have gradually achieved positive therapeutic effects for osteoporosis by PEMF exposure[lxxxv,lxxxvi,lxxxvii,lxxxviii,lxxxix,xc,xci]. Chronic pain is a common symptom of

people with osteoporosis. Many randomized controlled trials indicated that PEMF exposure could relieve chronic pain caused by osteoporosis[lxxxii,lxxxiii]. Moreover, in a study of 126 patients with primary osteoporosis, PEMF provided a faster and significant effect in relieving pain for patients with type I osteoporosis than those with type II[84]. BMD is the gold standard for diagnosing osteoporosis and the best quantitative indicator for forecasting the risk of osteoporotic fracture, monitoring the natural course of osteoporosis, and evaluating the effect of osteoporosis. Tabrah indicated that BMD of the treated radii was elevated notably in the sixth week in a clinical study of 20 women with PMOP treated with PEMFs[lxxvi]. In Garland's research, which evaluated the effect of PEMFs on knee osteoporosis in individuals with spinal cord injury, BMD was also elevated. At three months, BMD was increased by 5.1% in the stimulated knees but declined to 6.6% in the control knees[lxxvii]. PEMFs as a noninvasive physical therapy method avoids the defects of pharmacotherapy for osteoporosis, including the multiple side effects, the more cost and the low persistence. More importantly, a randomized, active-controlled clinical trial on postmenopausal osteoporosis (PMO) in Southwest China revealed that PEMFs had the same effect as alendronate, which is, currently, the most commonly prescribed medication for treating PMO within 24 weeks[xciii]. Furthermore, the hemorheological safety of PEMFs for treating osteoporosis was also observed by a randomized, placebo-controlled clinical study[xciv]. All these results support the efficiency and safety of PEMFs for osteoporosis treatment and as an advantageous treatment strategy in the future.

Fractures, delayed unions, and non-unions

Fractures, particularly those that had developed into delayed unions or even non-unions, have a substantial clinical, economic, and quality of life impact . Apart from traditional surgical management and rigid fixation (either internal or external), noninvasive PEMFs have already been used effectively in clinics as physical therapy to accelerate and finalize the healing process of a fresh fracture and reactivate the healing process of delayed unions and non-unions for nearly forty years since they were first approved by the US Food and Drug Administration[iv,v]. A recent systematic review and meta-analysis of randomized controlled trials showed that PEMFs significantly shortened the time to radiological union for acute fractures undergoing non-operative treatment and acute fractures of the upper limb and accelerated the time to clinical union for acute diaphyseal fractures[xcvi]. Moreover, a prospective study that evaluated the treatment effect of PEMFs on 64 patients undergoing hindfoot arthrodesis (144 joints) revealed that the adjunctive use of a PEMF in elective hindfoot arthrodesis may increase the rate and speed of radiographic union of these joints[xcvii]. Despite the relative scarcity of well-organized randomized controlled trials, many studies highlight the practice usefulness of PEMFs in treating tibial delayed unions or non-unions, with efficacy up to 87%[vi,lxxv,xcviii,xcix]. Furthermore, in a broad literature review comparing PEMF treatment of non-unions with surgical therapy, Gossling noted that 81% of reported cases healed with PEMF versus 82% with surgery. Obvious therapeutic advantages of PEMFs were showed compared with surgery in treatment for infected non-unions (81% versus 69%)[c] and closed injury caused non-unions (85% versus 79%) . In addition, a recent double-blind randomized study advocated that the adjunctive use of PEMF for fifth metatarsal fracture non-unions significantly shortened the average time to complete radiographic union from 14.7 weeks to 8.9 weeks compared with

the control group without PEMF exposure; the elevated expression levels of PIGF, BMP-5, and BMP-7, key regulators of angiogenesis and osteogenesis, were first detected in the non-union environment before and after the application of PEMFs[lviii]. These studies strikingly support PEMFs as an optional and effective method to accelerate fracture healing.

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