ECRONICON OPEN ACCESS

EC DENTAL SCIENCE
Opinion

Novel Insights on Cellular Effects of Pulsed Electromagnetic Fields on Osteoblasts are Key to Define their Clinical Applications

C Galli* and S Guizzardi

Department of Medicine and Surgery, University of Parma, Italy

***Corresponding Author:** Carlo Galli, Department of Medicine and Surgery, University of Parma, Italy.

Received: August 28, 2017; **Published:** September 22, 2017

Keywords: *Electromagnetic Fields; Osteoblasts, Primary Cilium*

Advances in bone regeneration have opened up new possibilities for the treatment of localized bone defects (LBDs), conditions that can affect any segment of the skeleton, including the maxillofacial area, as a consequence of trauma, disease or atrophy. As LBDs may heavily impact organ function and thus take a heavy toll on many aspects of patients' lives, better performing and more predictable approaches to promote the regeneration of lost tissues are sorely needed. Of particular interest are non-surgical approaches, which can spare patients from the additional morbidity of undergoing surgery to harvest or implant grafts or tissue substitutes into their LBDs. Electromagnetic Fields (EMF) are known to affect cell function and to promote osteoblast maturation and differentiation *in vitro* and *in vivo* and are therefore candidates for such therapies. Several papers have been published reporting the effects of EMF stimulation, mostly by Pulsed EMF (PEMF), on osteoblastic cell lines, e.g. MC3T3 (Li K 2014) or MG63 [1], or primary osteoblasts [2-5] and PEMFs have been proposed to prevent bone loss in animal models of tail suspension or ovariectomy-induced osteoporosis [6]. Although reports suggest that PEMFs can be used to treat non- or delayed unions or osteotomies [7,8], their clinical application has been significantly hindered and delayed by our poor understanding of its underlying mechanisms and therefore by difficulties in defining consistent sets of parameters to be applied across a number of patients (Li K 2014). A systematic review by Griffin., *et al.* [9] showed that evidence suggested a beneficial use of EMFs for the treatment of non-unions but there was a high degree of heterogeneity in the studies they examined, preventing more definitive conclusions. A vast range of frequencies, intensities, durations of exposure, pulses [10-12] or even waveforms [13] have been investigated and no consensus has been thus far reached. Studies have investigated frequencies ranging from few Hz to over 100 Hz and intensities of up to 4.5 mT or more, and the range of tested parameters appear to be more often associated to the constraints of the equipment used in the studies than to scientific evidence. A few important papers have however been published in the last two years, generating evidence that supports the idea that the effects of PEMFs require the existence of primary cilia. These manuscripts have brought specific cell substructures to the limelight, which could prove of immense value for a better understanding of how EMFs work. Primary cilia are solitary organelles that most cell types display depending on their differentiation state [14]. Their structure comprises an axoneme of 9 doublet microtubules (a 9+0 structure) that differentiates them from motile cilia, which possess a 9+2 structure, i.e. they possess a central doublet of microtubules, which, together with their associated proteins, is responsible for motility [15]. The group of Yan., *et al.* determined that 0.6 mT, 50 Hz PEMFs yielded the most effects in primary rat calvaria cells, in terms of cell proliferation, colony formation, mineralization nodules, and expression of differentiation markers. Cellular effects of PEMF however were abrogated by siRNA inhibition of intraflagellar transport protein IFT88, which is necessary for the formation of primary cilia [16].

Long believed to be inert structures, the idea has been gradually developed that primary cilia play a role in controlling cell division, possibly by sequestering centrioles (which is consistent with the observation that primary cilia are mostly possessed by quiescent cells), and that they are important sensory structures. With their antenna-like morphology they are indeed suited to sense mechanical stimuli, such as in osteocyte-mediated mechanoception [17] and mechanical forces were believed to be transduced through Ca^{2+} influx into the

cilium upon its deformation. Delling., *et al.* have recently proven that primary cilia do not require calcium signaling, by using a transgenic mouse model expressing a calcium indicator in cilia [18]. Cilia however have long been known to preferentially harbour several other receptor structures, such as PDGFR [19]. More recently, primary cilia have been shown to play a regulatory role in clathrin-dependent endocytosis (CDE), which occurs mostly in the pocket region at the base of cilia [20,21]. Interestingly, cilia-associated CDE has been shown to be relevant for TGF-β signaling, as TGF- β receptors have been localized at the tip of primary cilia in fibroblasts, and receptor stimulation activated SMAD2/3 and ERK1/2 at the cilium base. Conversely, inhibition of CDE or stunning of primary cilia decreased TGF-β signalling [22]. It has been furthermore recently shown that 0.6 mT 50 Hz PEMFs increased BMP-Smad1/5/8 signaling in primary rat calvaria cells and BMPRII silencing inhibited the effects of PEMFs on osteoblasts [23]. Moreover, PEMFs up-regulated the expression of BMPRII at the base of the cilium, and cilium abrogation by knockdown of IFT88 inhibited PEMFs effects on BMPRII. Interestingly, 75 Hz 2.5 mT PEMFs have been shown to up-regulate the expression of A_{2a} and A_3AR adenosine receptors in hFOB 1.19 osteoblastic cells [24]. Not much is known about the localization of adenosine receptors at the base of primary cilia, but it is possible to see a common denominator here and hypothesize that a relevant part of PEMFs action is mediated by up-regulation of membrane trafficking and therefore membrane receptors and that, given the important role of primary cilia in organizing transmembrane structures, primary cilia are required for PEMF activity in bone cells and in other cell types. Further research should focus on elucidating these mechanisms as they may represent the key to unlock the potential of electromagnetic fields in several clinical applications, not least in bone regeneration.

Conflict of Interest

The authors have no conflict of interest to disclose.

Acknowledgement

The authors would like to thank Dr. Arianna Smerieri for her advice.

Bibliography

1. Sollazzo V., *et al*. "Effects of pulsed electromagnetic fields on human osteoblastlike cells (MG-63): a pilot study". *Clinical Orthopaedics and Related Research* 468.8 (2010): 2260-2277.

2. Wang J., *et al*. "Pulsed electromagnetic field may accelerate in vitro endochondral ossification". *Bioelectromagnetics* 36.1 (2015): 35-44.

3. Fu YC., *et al*. "A novel single pulsed electromagnetic field stimulates osteogenesis of bone marrow mesenchymal stem cells and bone repair". *PLoS One* 9.3 (2014): e91581.

4. Lim K., *et al*. "Effects of electromagnetic fields on osteogenesis of human alveolar bone-derived mesenchymal stem cells". *BioMed Research International* (2013): 296019.

5. Sun LY., *et al*. "Pulsed electromagnetic fields accelerate proliferation and osteogenic gene expression in human bone marrow mesenchymal stem cells during osteogenic differentiation". *Bioelectromagnetics* 31.3 (2010): 209-219.

6. Jing D., *et al*. "Circadian rhythm affects the preventive role of pulsed electromagnetic fields on ovariectomy-induced osteoporosis in rats". *Bone* 46.2 (2010): 487-495.

7. Bassett CA., *et al*. "A non-operative salvage of surgically-resistant pseudarthroses and non-unions by pulsing electromagnetic fields. A preliminary report". *Clinical Orthopaedics and Related Research* 124 (1977): 128-143.

8. Bassett CA., *et al*. "Treatment of ununited tibial diaphyseal fractures with pulsing electromagnetic fields". *Journal of Bone and Joint Surgery-American* 63.4 (1981): 511-523.

9. Griffin XL., *et al*. "Electromagnetic field stimulation for treating delayed union or non-union of long bone fractures in adults". Cochrane Database of Systematic Reviews 4 (2011): CD008471.

10. Markov MS. "Magnetic field therapy: a review". *Electromagnetic Biology and Medicine* 26.1 (2007): 1-23.

11. Liu C., *et al*. "Effect of 1 mT sinusoidal electromagnetic fields on proliferation and osteogenic differentiation of rat bone marrow mesenchymal stromal cells". *Bioelectromagnetics* 34.6 (2013): 453-464.

12. Lei T., *et al*. "Effects of four kinds of electromagnetic fields (EMF) with different frequency spectrum bands on ovariectomized osteoporosis in mice". *Scientific Reports* 37.1 (2017): 553.

13. Zhou J., *et al*. "Different electromagnetic field waveforms have different effects on proliferation, differentiation and mineralization of osteoblasts in vitro". *Bioelectromagnetics* 35.1 (2014): 30-38.

14. Satir P. "CILIA: before and after". *Cilia* 6 (2017): 1.

15. Pedersen LB., *et al*. "The ciliary cytoskeleton". *Comprehensive Physiology* 2.1 (2012): 779-803.

16. Yan JL., *et al*. "Pulsed electromagnetic fields promote osteoblast mineralization and maturation needing the existence of primary cilia". *Molecular and Cellular Endocrinology* 404 (2015): 132-140.

17. Hoey DA., *et al*. "A role for the primary cilium in paracrine signaling between mechanically stimulated osteocytes and mesenchymal stem cells". *Biochemical and Biophysical Research Communications* 412.1 (2011): 182-187.

18. Delling M., *et al*. "Primary cilia are not calcium-responsive mechanosensors". Nature 531.7596 (2016): 656-660.

19. Schneider L, *et al*. "PDGFRaα signaling is regulated through the primary cilium in fibroblasts". *Current Biology* 15.20 (2005): 1861-1866.

20. Satir P., *et al*. "The primary cilium at a glance". *Journal of Cell Science* 123.4 (2010): 499-503.

21. Sorkin A and von Zastrow M. "Endocytosis and signalling: intertwining molecular networks". *Nature Reviews Molecular Cell Biology* 10.9 (2009): 609-622.

22. Clement CA., *et al*. "TGF-β signaling is associated with endocytosis at the pocket region of the primary cilium". *Cell Reports* 3.6 (2013): 1806-1814.

23. Xie YF., *et al*. "Pulsed electromagnetic fields stimulate osteogenic differentiation and maturation of osteoblasts by upregulating the expression of BMPRII localized at the base of primary cilium". *Bone* 93 (2016): 22-32.

24. Vincenzi F., *et al*. "Pulsed electromagnetic fields increased the anti-inflammatory effect of A_{2A} and A_3 adenosine receptors in human T/C-28a2 chondrocytes and hFOB 1.19 osteoblasts". *PLoS One* 8.5 (2013): e65561.

Volume 14 Issue 4 September 2017