Effects of Static and Pulsed Electromagnetic Fields on Orthodontic Tooth Movement: An Animal Study

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ABSTRACT

Background: Electromagnetic field therapy has emerged as a non-invasive and effective modality for enhancing bone quality and quantity during the healing process. This study aims to determine the effects of pulsed electromagnetic fields (PEMF) and static magnetic fields (SMF) of the same intensity and duration on orthodontic tooth movement.

Methods: Forty-five male albino rabbits were randomly assigned to three groups (15 rabbits per group): control group, PEMF group and SMF group. The PEMF group was exposed to a pulsed electromagnetic field generated by a commercially available portable device (square waveform, 120 Gauss intensity, 9.6 Hz frequency, with 1 hour of daily exposure for 3 weeks). The SMF group was subjected to a static magnetic field generated by neodymium iron boron permanent magnets (120 Gauss intensity, with 1 hour of daily exposure for 3 weeks). Five rabbits from each group were sacrificed at the end of each week for analysis.

Result: The rate of tooth movement, amount of new bone formation and the count and activity of osteoblasts and osteoclasts were significantly higher in the PEMF group as compared to the control group. The SMF group also exhibited significantly higher results in most of the measured variables over the 3 weeks relative to the control group, although the outcomes were less pronounced than those observed in the PEMF group.

Key words: Orthodontic tooth movement, Pulsed electromagnetic field, Static magnetic field.

INTRODUCTION

Orthodontic treatment typically lasts 24 to 36 months, which concerns most patients with fixed appliances. Methods to speed up tooth movement include Photobiomodulation, mechanical vibration, electromagnetic fields and surgical approaches. After discovering bone's piezoelectric properties, Bassett *et al.* (1976) first reported that a changing electromagnetic field can induce a current in bone without using implanted electrodes (Stark and Sinclair, 1987). There are experimental and clinical studies reporting that magnetic field can cause positive progress in various aspects of the body, including increased cell membrane permeability (Öcal *et al*., 2020). The term magnetic fields (MFs) defines the effect that particles with electric charge create around them (Cevik *et al*., 2017). Magnetic fields (MF) can be categorized into two types based on whether their intensity and direction change over time: static magnetic fields and pulsed (time-alternating) electromagnetic fields (Fan *et al*., 2021). Electromagnetic fields consist of interrelated magnetic and electric fields, these fields can penetrate tissues without a decrease in intensity passing through cell membranes and influence cellular responses.

In this research, we studied the effects of a commercially available, affordable, small-sized portable pulsed electromagnetic field generator on the rate of tooth movement and compare results with those obtained from a static magnetic field of the same intensity and exposure duration. This study aims to evaluate the effects of pulsed electromagnetic fields (PEMF) and static magnetic fields (SMF) on tooth movement speed. It will also examine histological changes in alveolar bone, including new bone formation and the presence of osteoblasts and osteoclasts, along with immunohistochemical changes in bone formation factor (bone alkaline phosphatase) and bone resorption factor (tartrate-resistant acid phosphatase 5b).

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MATERIALS AND METHODS

This research was performed from July 2023 to May 2024. The experimental phase of this research received approval from the Ethics Committee of the College of Dentistry at Mosul University (approval code: UoM.Dent. 23/35, No. 35 on 4/6/2023). All required measures were taken to minimize animal pain or discomfort during the whole study period. The animal housing and follow-up were done in the College of Veterinary Medicine (University of Mosul, Mosul, Iraq), while experimental parameters and histology were

conducted in the College of Dentistry (University of Mosul, Mosul, Iraq).

The study sample consisted of 45 local rabbits, which were randomly divided into 3 groups (15 rabbits in each group):

Group 1: Control group (C), this group received an orthodontic appliance to produce 50-gram force in distal direction on lower incisors.

Group 2: PEMF group (pulsed electromagnetic field); use the same orthodontic force in addition to the application of pulsed electromagnetic field.

Group 3: The SMF group (static magnetic field), use the same orthodontic force in addition to exposure to static magnetic field.

Orthodontic force application lasts for total three weeks and at the end of each week, 15 rabbits (5 from each study groups) were sacrificed.

Orthodontic appliance

The orthodontic appliance includes two stainless steel bands with tubes (2.5 mm wide, 0.022 × 0.028 inch size) attached to lower incisors. The setup consists of two bands, an open coil spring, an archwire and a ligature wire, similar to appliance used by Al Zubaidi *et al.* (2024). A 7.5 mm nickel-titanium open coil spring (light force, 0.010 × 0.030 inches) was used, generating 50 grams of force when compressed into a 3.5 mm space (initially present between two adjacent tubes). The 3.5 mm distance is set as a key to maintaining a consistent 50-gram force across all samples (Fig 1).

PEMF device and exposure

In the PEMF group, we used the MiraMate Mini Magic - Portable PEMF device (China) to generate pulsed electromagnetic fields. It produces a square waveform with a frequency range of 9.6 Hz. For standardization, the PEMF coils were mounted on a plastic holder in a Helmholtz configuration, keeping them parallel and 17 mm apart (equal to the coil radius) to create a uniform magnetic field (Darendeliler *et al.,* 2007; Dennis, 2020).

A plastic rod was attached to the left side of the holder, passing through the center of the left coil and ending

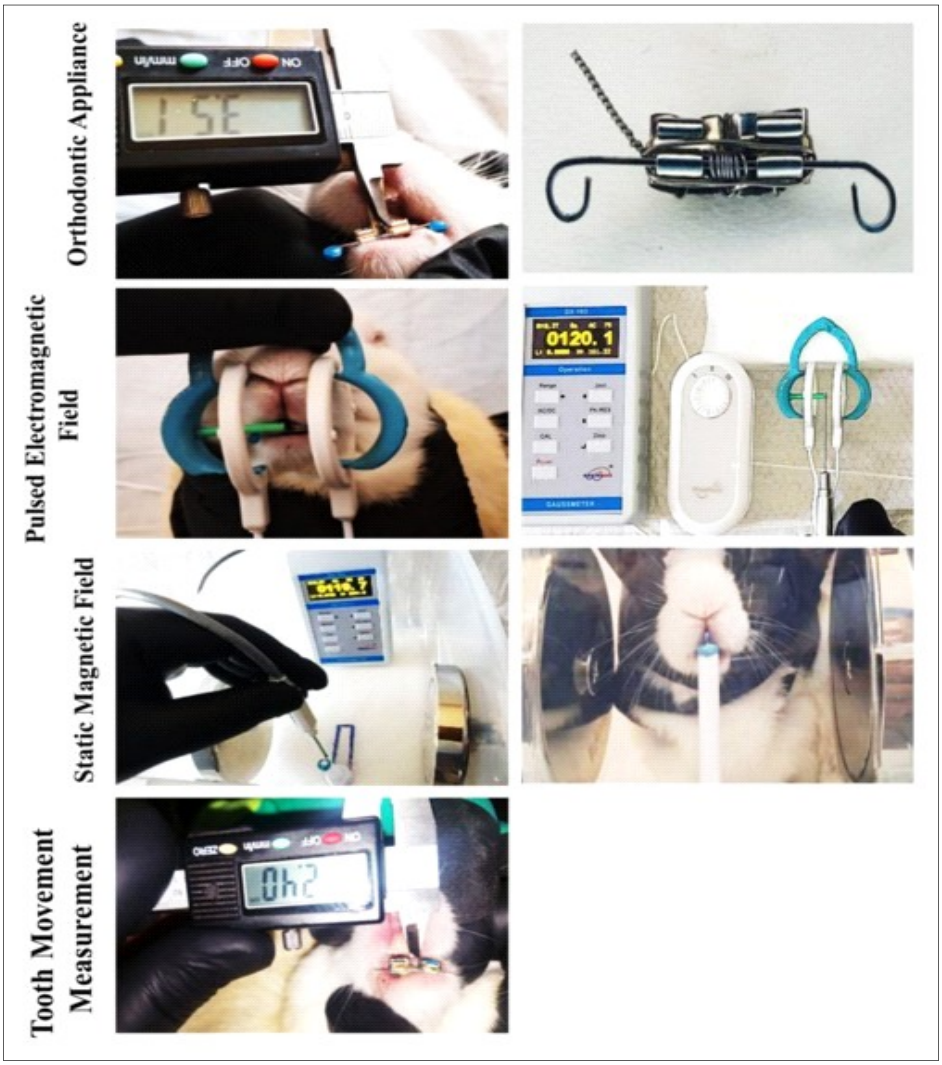


Fig 1: Medical appliances used to measure the studied parameters.

midway between the two coils (8.5 mm). The rod's tip was always positioned at the rabbit's lip midline, serving as a consistent reference point during sessions. At its end, a Gauss meter probe could measure peak magnetic flux intensity when needed (Fig 1).

The intensity of the pulsed electromagnetic field was measured using a handheld AC/DC Gauss meter (DX-103, DEXINGMAG Company, China). The peak reading during each session was found equal to 120 Gauss (12 mT). The PEMF exposure lasted 1 hour per day, split into two 30-minute sessions-one in the morning and one in the evening-over 21 days.

Static magnetic field (SMF) exposure

SMF was generated using two Neodymium-iron-boron (NdFeB) permanent magnets (Wukong, China), each with a 120 mm diameter and 18 mm thickness, attached to the right and left sides of a modified plastic cage that held the rabbits during exposure. The cage, measuring 17 cm in width, 30 cm in depth and 25 cm in height, was large enough to keep the rabbits in place, preventing significant changes in body position. The magnets were positioned so that the N pole of the right magnet faced the S pole of the left, orienting the magnetic field from right to left. A plastic screw (12 cm long) was inserted at the base of the container, aligned with the rabbit's lip midline as a reference point. The screw was fixed at a position that produced a peak reading of 120 Gauss, measured by a Gauss meter probe at its end (Fig 1). The rabbits were exposed to SMF for 1 hour per day, split into two 30-minute sessions-one in the morning and one in the evening-throughout the experimental period.

Measurement of rate of tooth movement

The rate of tooth movement was measured by the direct method using a digital caliper between the mesioincisal angle of the lower incisors. The measurement of tooth movement was performed at 10 times point: 1, 3, 5, 7, 9, 12, 14, 16, 18 and 21 days (Fig 1).

Histological analysis

Sections of lower incisors, including periodontal ligament and alveolar bone, were examined under a light microscope. The sections were categorized into two regions: The upper region (U), encompassing the alveolar bone and PDL around the upper part of the root cervically, both medially and distally and the lower region (L), covering the lower part of the root apically, both medially and distally.

Statistical analysis

Statistical analysis was conducted using IBM SPSS statistics 25. The data were tested for their normal distribution by using the Shapiro-Walk test and all data were normally distributed. Comparison among the three groups regarding tooth movement, new bone formation area, osteoblast and osteoclast count were done by using one-way ANOVA and Duncan post hoc multiple comparisons test.

Results and Discussion

Rate of orthodontic tooth movement

It is evident that the PEMF group exhibited the fastest rate of tooth movement from day 3 to day 16, with a significant difference compared to the other groups. The SMF group also began to show a faster rate of tooth movement from day 9 to day 16, with a significant difference compared to the control group. However, on days 1, 18 and 21, there was no significant difference in the rate of tooth movement among all the groups (Table 1).

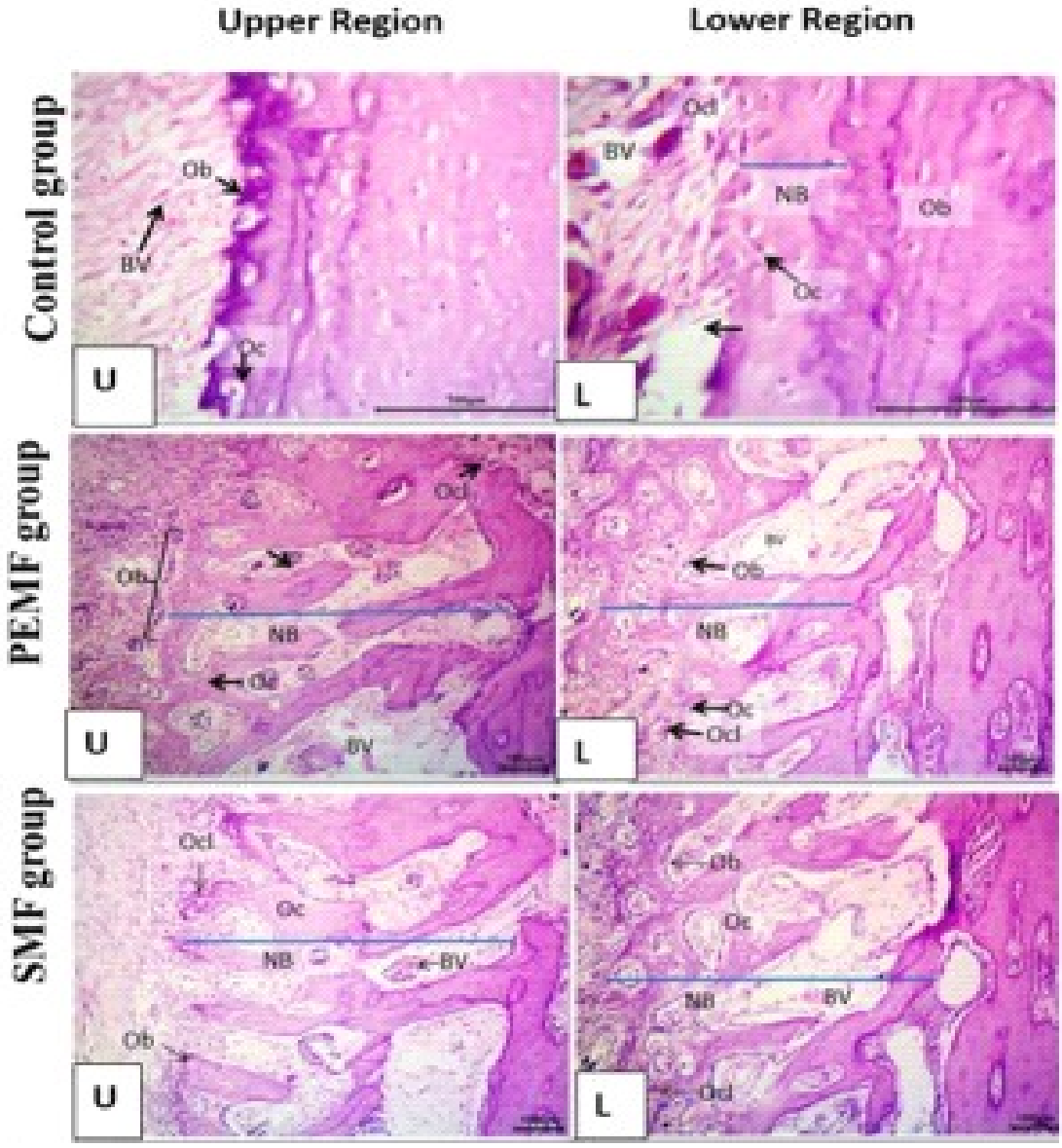
New bone formation (Osteoid)

From Table (2), the PEMF group exhibited a significantly greater amount of bone formation compared to the other

Table 1: Amount of orthodontic tooth movement among all groups.

Groups	Day	n	Mean±SD	P value	Day	n	Mean±SD	P value
Control	1	15	0.77±0.13A	0.846	12	10	2.18±0.18A	0.03
PEMF		15	0.81±0.14A			10	2.58±0.29B	
SMF		15	0.79±0.14A			10	2.36±0.25AB	
Control	3	15	1.36±0.15A	0.0001	14	10	2.46±0.18A	0.0001
PEMF		15	1.55±0.29B			10	2.9±0.23C	
SMF		15	1.21±0.23A			10	2.67±0.25B	
Control	5	15	1.57±0.12A	0.034	16	5	2.88±0.08A	0.053
PEMF		15	1.74±0.27B			5	3.16±0.23B	
SMF		15	1.55±0.2A			5	3.04±0.13AB	
Control	7	15	1.67±0.12A	0.001	18	5	3.18±0.08A	0.133
PEMF		15	1.88±0.29B			5	3.38±0.18A	
SMF		15	1.68±0.23A			5	3.26±0.13A	
Control	9	10	1.93±0.16A	0.02	21	5	3.44±0.11A	0.245
PEMF		10	2.38±0.32C			5	3.62±0.15A	
SMF		10	2.06±0.26AB			5	3.48±0.13A	

Data expressed as Mean±SD, Different letters vertically within same time point indicate significant differences among groups at $p<0.05$ using Duncan multiple test.



New bone formation (NB) (blue line), alveolar bone (AB), osteoblasts (Ob), osteoclast (Oc) and blood vessels (BV).
U= upper region and L= Lower region. H and E stain, (100X), Scale bar= 100 µm.

Fig 2: Histological sections from the control group, PEMF group and SMF group at week 3 post-orthodontic appliance at the upper region (left panel) and lower region (right panel).

Table 2: Comparison of surface area of the new bone formation in all groups at 1, 2 and 3 weeks.

Group	Week 1		Week 2		Week 3	P-value
	Mean±SD	P-value	Mean±SD	P-value	Mean±SD	
C-U	148887±20429A		190838±7220A		273833±22752A	
C-L	136699±18208A		177515±8265A		252103±11100A	
PEMF-U	225170±11390B		368817±39251D		520022±35468C	
PEMF-L	228820±15298B	0.0001	332997±29058C	0.0001	535676±36680C	0.0001
SMF-U	157023±28309A		229067±27566B		376789±30323B	
SMF-L	142061±21638A		239220±21979B		365619±20824B	

Data expressed as Mean±SD measured in cubic micrometer (μm^2), Different letters vertically within the same period indicate significant differences among groups at $p<0.05$ using Duncan multiple test, C- control, U- upper region, L- lower region.

two groups at all-time points (Fig 2). On the other hand, the SMF group exhibited a significantly greater amount of bone formation compared to the control group at 2 and 3 weeks only (Fig 2).

Osteoblast count

Table (3) indicates significant differences in osteoblast numbers between the PEMF group and the other two groups at 1, 2 and 3 weeks, with the PEMF group showing the highest counts. Significant differences were also observed between the SMF and control groups at 1 and 3 weeks, but not at 2 weeks.

Osteoclast count

Table (3) displays the osteoclast numbers. The PEMF group demonstrated a significant increase in osteoclast numbers compared to the other groups throughout all periods. The SMF group showed a significant difference over the control group at 1 and 2 weeks, but not at 3 weeks.

Immunohistochemistry analysis

In immunohistochemistry (IHC) analysis, the bone formation marker (bone alkaline phosphatase, BALP) and the bone resorption marker (tartrate-resistant acid phosphatase, TRAP 5b) were examined on both the upper and lower regions across all groups over 1, 2 and 3 weeks.

Bone alkaline phosphatase

In Table (4), BALP activity score was significantly higher in the PEMF group compared to the other groups (Fig 3). Additionally, the SMF group had significantly higher scores than the control group at all-time points.

Tartrate-resistant acid phosphatase 5b

In Table (5), the highest TRAP 5b score was observed in the PEMF group across all periods (Fig 4). The SMF group displayed significantly higher TRAP 5b scores compared to the control group, but only during the 2 and 3 week periods.

The difference in tooth movement between the PEMF and the control group appeared on day 3 and lasted until day 16. This suggests that PEMF likely began providing a clinical benefit by day 3. After day 16, the lack of significant difference may be attributed to the reduced force in the orthodontic appliance, which might have been insufficient to show a difference between the groups. These findings align with several studies that observed a significantly faster rate of tooth movement in the PEMF groups (Stark and Sinclair, 1987; Zaffe *et al*., 1998; Darendeliler *et al*., 2007 and Dogru *et al*., 2014).

In terms of new bone formation, the PEMF group shows a significant difference as early as 1 week, with the amount of new bone nearly doubling that of the control group by 3 weeks. These findings align with the studies that observed that electromagnetic fields appear to enhance bone quality and quantity (Stark and Sinclair, 1987; Darendeliler *et al*., 1997; Zaffe *et al*., 1998). Many mechanisms proposed for this enhancement in bone formation like activation of primary cilia-associated signaling pathway (Wang *et al*., 2019), activation of Wnt/β-catenin signalling, (Kobayashi-Sun *et al*., 2024), increased expression of piezo 1 and Ca_2+ influx (Chen *et al*., 2023).

Regarding osteoblast count, the PEMF group showed significantly higher results compared to the other groups, with the highest count observed at the 3-week mark. This result agrees with many researchers who studied the effect of pulsed electromagnetic on differentiation, proliferation and maturation of osteoblast (Barnaba *et al.,* 2013; Yan *et al*., 2015; Kobayashi-Sun *et al.,* 2024) but it might disagree with a study who found that exposure to 50 Hz sinusoidal PEMF inhibits the osteoblast proliferation but promotes differentiation and mineralization potentials (Zhou *et al*., 2011).

The bone alkaline phosphatase activity reached its highest score in the PEMF group at the 3-week mark. This corresponds with the osteoblast count, indicating that most osteoblasts in the upper and lower regions were active. These findings align with other research demonstrating the positive effects of PEMF-across various intensities, frequencies and waveforms-on osteoblast cell cultures (Wang *et al*., 2019; Zhou *et al*., 2021; He *et al*., 2022).

Many mechanisms proposed about this PEMF effect on osteoblast and BALP activity, production of the non-toxic level of reactive oxygen species, which induces antioxidative defense mechanisms in osteoblast (Ehnert *et al*., 2017), activation of Wnt/β-catenin signaling (Kobayashi-Sun *et al.,* 2024), increased expression of piezo 1 and Ca_2+ influx

Table 3: Comparison of the counts of the osteoblasts and osteoclasts in all groups after 1, 2 and 3 weeks.

Time	Week 1		Week 2		Week 3	
Groups	Osteoblasts	Osteoclasts	Osteoblasts	Osteoclasts	Osteoblasts	Osteoclasts
C-U	29.8±4.65AB	3.2±1.3AB	65.2±17.19A	7.6±1.14A	105.4±11.45A	16.6±1.67A
C-L	25±5.78A	2.6±1.14A	64.4±12.85A	7.8±1.3A	97.4±5.81A	15.2±2.86A
PEMF-U	44.8±4.86D	8.00±1.58D	98±6.63B	17.6±2.4C	160±7.31C	27.2±3.96B
PEMF-L	44.2±5.26D	7.6±1.81D	120.4±11.45C	15.8±1.92C	163.8±7.56C	24.8±4.08B
SMF-U	34.2±3.11BC	5±0.7C	78.8±6.18A	12.8±1.48B	133.8±4.96B	18.2±1.92A
SMF-L	37.8±3.11C	4.8±0.83BC	77.4±5.31A	12±1.87B	126.6±6.26B	17±2A
P value	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001

Data expressed as Mean±SD and represent total osteoblast/osteoclast count, Different letters vertically within the same period indicate significant differences among groups at $p<0.05$ using Duncan multiple test, C- Control, U- Upper region, L- Lower region.

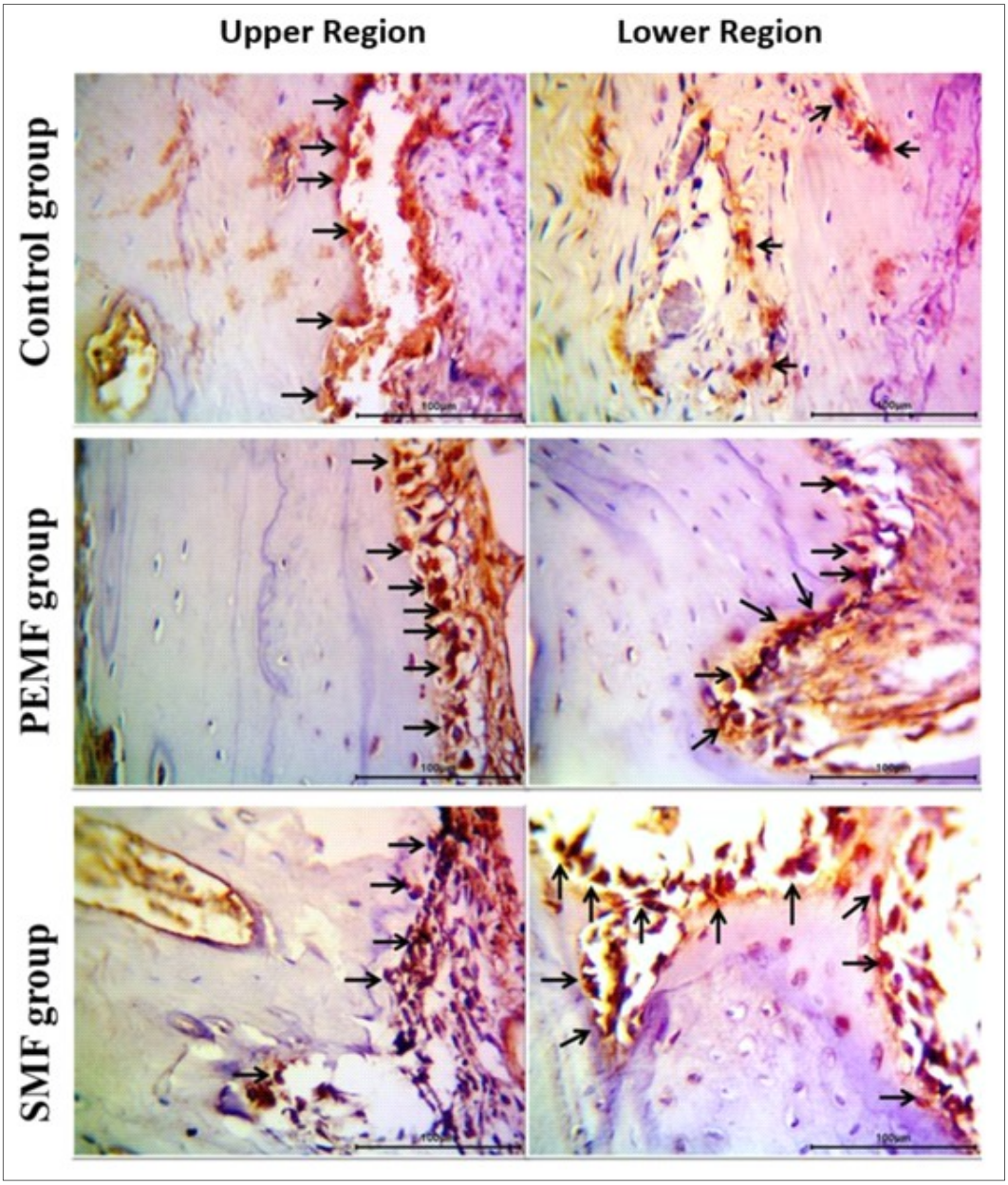


Fig 3: Immunohistochemistry expression of the bone Alkaline phosphatase at week 3 post-orthodontic appliance control.

Table 4: The IHC scores of the bone alkaline phosphatase in all groups after 1, 2 and 3 weeks.

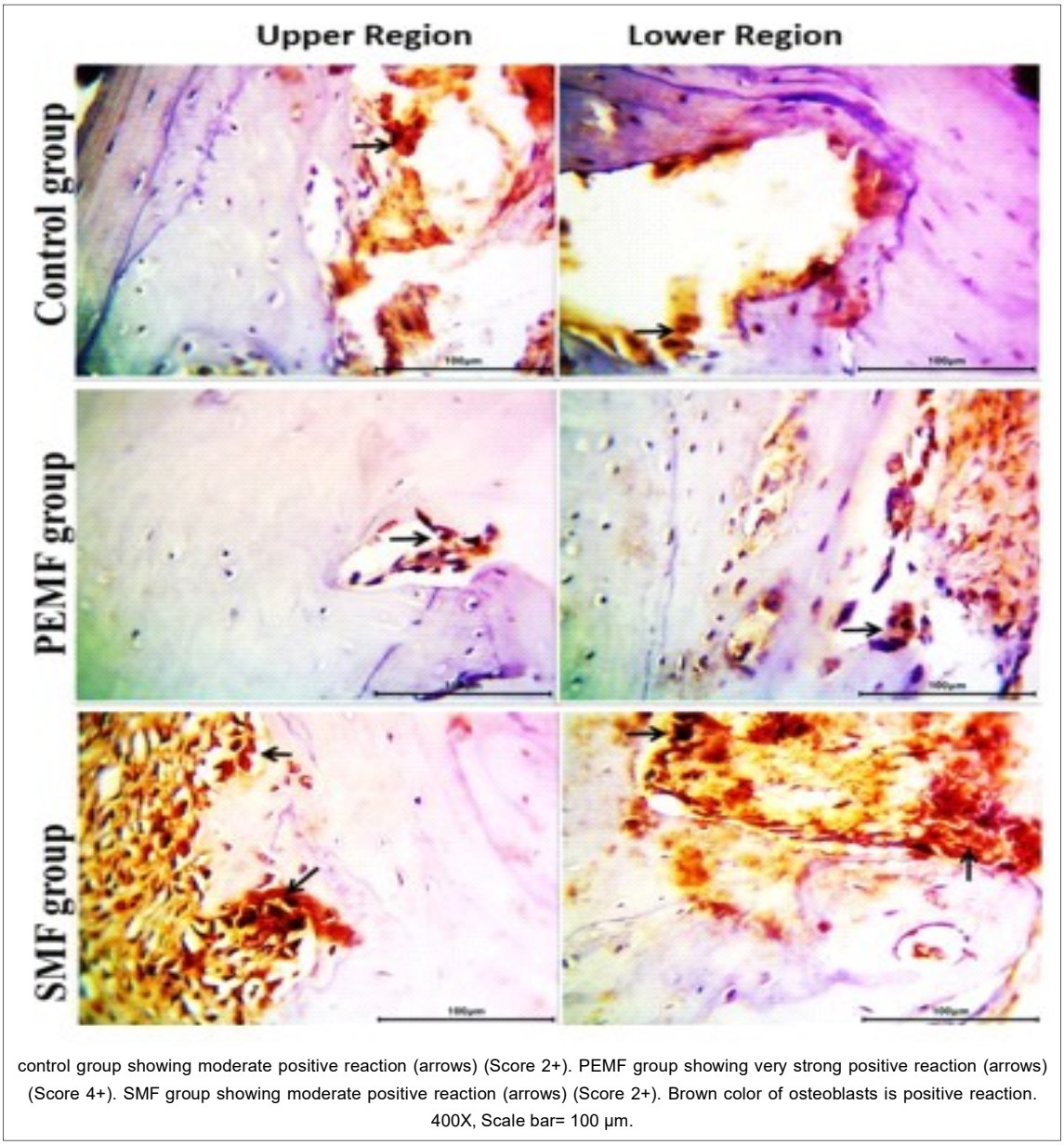
Groups	Region	1 Week	2 Weeks	3 Weeks	*P*-Value
Control group	Upper	0(0.5)Aa	1(1)Ab	2(1)Ac	0.013
	Lower	0(1)Aa	1(0.5)Ab	2(1)Ac	0.008
PEMF group	Upper	3(1)Ba	4(1)Bb	4(0)Bb	0.038
	Lower	3(0.5)Ba	4(0.5)Bb	4(0)Bb	0.016
SMF group	Upper	2(0.5)Ba	3(1)ABab	3(0.5)ABb	0.038
	Lower	2(1)Ba	3(0)ABb	3(1)ABb	0.028
	P-Value	<0.001	<0.001	<0.001	

Data expressed as Median and IQR ((Inter-Quartile-Range) (N= 5). Different small letters among groups in rows mean there is a significant difference at $p \leq 0.05$. Different capital letters among groups in columns mean there is a significant difference at $p \leq 0.05$.

(Chen *et al.,* 2023), primary cilia length extension and increased protein kinase activation (Zhou *et al.,* 2021).

The present study confirmed that the osteoclast counts and TRAP 5b marker for osteoclast activity was significantly higher in the PEMF group across all three time periods studied than in other groups. It concurs with Kobayashi-Sun *et al.* (2024), who found that exposure to 10 mT PEMFs at 60 Hz increased osteoclast numbers. However, this contradicts Wang *et al.* (2021), who reported a decrease in osteoclast numbers with PEMF exposure at 50 and 75 Hz, 1.6 mT and partially agree with Hong *et al.* (2014) found that a 45 Hz PEMF inhibited osteoclast formation and TRAP activity, while a 7.5 Hz PEMF induced osteoclast differentiation and TRAP activity (both at 1 mT).

The difference in the rate of tooth movement between the SMF and control groups emerged on day 9 and lasted until day 16. This delay suggests that SMF required more time to significantly impact tooth movement compared to PEMF. After day 16, no significant difference was observed, possibly due to tissue adaptation to the static magnetic field or reduced force remaining in the orthodontic appliance. This result agrees with Luo *et al*. (2024) who mentioned that SMF promotes osteoclastogenesis by inducing force loaded periodontal ligament stem cells to secrete interleukin 6 which accelerates orthodontic movement. also consistent with the findings of Shan *et al.* (2021) who used different intensities and durations.



control group showing moderate positive reaction (arrows) (Score 2+). PEMF group showing very strong positive reaction (arrows) (Score 4+). SMF group showing moderate positive reaction (arrows) (Score 2+). Brown color of osteoblasts is positive reaction. 400X, Scale bar= 100 µm.

Fig 4: Immunohistochemistry expression of the *TRAP 5b* at week 3 post-orthodontic appliance.

Table 5: The IHC scores of the tartrate-resistant acid phosphatase 5b in all groups after 1, 2 and 3 weeks.

Groups	Region	1 Week	2 Weeks	3 Weeks	*P*-Value
Control group	Upper	1(1)Aa	1(0.5)Aa	1(1)Aa	0.089
	Lower	1(1)Aa	1(0.5)Aa	2(1)Ab	0.038
PEMF group	Upper	3(1)Ba	3(1)Ba	4(1)Bb	0.038
	Lower	3(0.5)Ba	3(0.5)Ba	4(0.5)Bb	0.013
SMF group	Upper	1(0.5)Aa	2(0.5)ABab	2(1)ABb	0.018
	Lower	1(1)Aa	2(1)ABab	3(1)ABb	0.015
	p value	<0.001	<0.001	<0.001	

Data expressed as Median and IQR ((Inter-Quartile-Range) (N= 5). Different small letters among groups in rows mean there is significant difference at $p \leq 0.05$. Different capital letters among groups in columns mean there is significant difference at $p \leq 0.05$.

In terms of new bone formation, the SMF group demonstrated significantly higher results in weeks 2 and 3 compared to the control group, but consistently lower results than the PEMF group. This is consistent with the findings of Darendeliler *et al.* (1995), who observed increased bone and matrix deposition after SMF exposure and also aligns with other researchers who found that SMF promotes new bone apposition, enhanced osteogenesis and accelerated fracture healing (Kim *et al*., 2017; Li *et al*., 2020; Wang *et al.,* 2023). Many mechanisms proposed for this enhancement in bone formation like activating the phosphorylated AKT pathway (Zhang *et al.,* 2023), reducing Intracellular reactive oxygen species levels (Frachini *et al.,* 2023) and enhancing differentiation *via* FLRT/BMP signalling (Li *et al.,* 2020).

A significant difference in osteoblast numbers between the SMF and control groups was observed at weeks 1 and 3, with no significant difference at week 2. Regarding bone alkaline phosphatase activity, all three time points showed a significant difference between the SMF and control groups. These results align with multiple studies, such as Feng *et al.* (2010), who found that after just 1 day of SMF irradiation, alkaline phosphatase activity significantly increased, showing a 1.5-fold rise, Kim *et al.* (2017) reported that static magnetic fields promoted osteoblastic differentiation by activating various signalling pathways, including Wnt/β-catenin. This stimulation enhanced the activity of early markers such as alkaline phosphatase.

An early significant difference in osteoclast numbers was observed between the SMF and control groups in week 1, continuing into week 2. However, by week 3, no significant difference was found. In terms of TRAP 5b activity scores, no significant difference was observed between the SMF and control groups at week 1, but significant differences emerged during the 2-week and 3-week periods. These results are consistent with Shan *et al.* (2021), who found that more osteoclasts were observed after exposure to 20-204 mT SMF and also align with Luo *et al.* (2024), who discovered that SMF (200±20 mT) promotes osteoclastogenesis by inducing periodontal ligament stem cells to secrete IL-6 and Barnaba *et al.* (2012) who found that cells exposed to SMF (0.9 μT) exhibited a significantly higher TRAP activity after 7 and 10 days.

Conclusion

Within the confines of this study, the pulsed electromagnetic field (PEMF) group demonstrated statistically significant superiority across all assessed parameters, including the rate of tooth movement, new bone formation, as well as osteoblast and osteoclast count and activity. The static magnetic field (SMF) group exhibited notable improvements in most of the measured variables compared to the control group; however, the effects were less pronounced than those observed in the PEMF group.

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Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article.

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