



The effects of local pulsed magnetic field with intraosseous implant on clotting time and calcium level

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Abstract

Background: Many researches indicated that magnetic field had effect on clotting time and serum calcium level when it was used for stimulation of bone healing and growth. This in vivo study was carried out to evaluate the effect of local pulsed magnetic on clotting time and serum calcium level when used for stimulation of intraosseous implant.

Materials and Methods: Forty eight oryctologus cuniculus male rabbits were used in this study, they divided into two controlled groups and two experimental groups and each group consist of 12 animals, all experimental groups exposed to (50 gauss) pulsed magnetic field strength. Clotting time and calcium level measured at intervals of time (2, 4, 6 and 8) weeks of healing periods.

Results: the result indicated that theirs no significant differences between the controlled group and other groups for (50 Gauss) pulsed magnetic field which indicate no harmful effect of magnetic field on clotting time and calcium level.

Conclusion: The use of (50 Gauss) pulsed magnetic field show no noticeable affect on serum calcium level and clotting time.

Keyword: Pulsed magnetic field, clotting time, serum calcium level.

Introduction

The effect of magnetic fields on clotting time was investigated in rats by Zalyvbovaskya, 1973 and their result indicated that the blood coagulation in rate become longer in time after exposure to the electromagnetic wave¹. Hannow, 1991 showed that in his study the exposure of rabbits to(6 gauss) static and magnetic field lead to increasing the clotting time², and the same results had been showed by Payman, 1995 while same result was obtain when 100mT was used for whole body exposure in the rabbit³. Study in vitro by Tashaev

et al., 1997 demonstrated the effect of electromagnetic field on the stability of blood formed elements in an artificial closed circuit and their results indicated that the resistance of erythrocytes increased in the circuit by electromagnetic field⁴. Researches by Robert, 2001 on the effect of 10Hz electromagnetic field on the morphology of the red and white blood cells in groups of volunteers. These disturbing frequencies of 10 Hz to 100 Hz were mediated by QLink (electromagnetic device). After 72 hours, the red blood cells appeared to

be more round and symmetrical which is the normal healthy profile. There was a significant notable improvement in the morphology of the live and coagulated blood of all participants when wearing a QLink pendant. It is hypothesized that the improvement in the morphology of the blood is a direct result of the disturbing low EMF⁵. Studies by Korpinar *et al.*, 2002, on the effect of 300MHz on blood parameters (measured erythrocyte, thrombocyte, leukocyte and coagulation time and their results shows no significant results between all parameters in the studies they showed slight increasing in coagulation time⁶. Frankle *et al.*, 1998 demonstrated that blood, like all tissues, contains electrically charged ions. A physics principle known as Faraday's Law states that a magnetic field will exert a force on a moving ionic current and the extension of Faraday's law called the Hall effect which states that when a magnetic field is placed perpendicular to the direction of flow of an electric current, it will tend to deflect and separate the charged ions. While the deflection of ions will be in opposite directions depending on the magnetic pole encountered and the charge of the ion, this force is not based on the attraction or repulsion of like and unlike charges⁷.

Materials and Methods

Study design:-

The experimental animals were divided into four groups, two experimental and two controls, each group consist of twelve animals, 5mm length of IMTEC Sendax implants were inserted at 2 cm away from proximal end of lateral surface of left tibia for (A and B groups) of the rabbits while in groups (C and D) 24 cart Gold implant of 5mm in length inserted in same site and as follows:-

Group A: - Consist of twelve animals, with titanium implant and this group not exposed to magnetic field and this group considered as a control titanium group.

Group B: - Consist of twelve animals, with titanium implant and this group exposed to pulsed magnetic field at the implant site and this group considered as experimental group

Group C: - Consist of twelve animals, with gold implant and this group was not exposed to the magnetic field and this group considered as a control gold group.

Group D: - Consist of twelve animals, with gold implant and this group exposed to pulsed magnetic field at the implant site.

Biochemical serum analysis:

Serum calcium was investigated for both experimental and control groups on sacrifice day. The rabbits placed in clean placed and the heart region was palpated by fingers and 5cc needle introduced into the heart just in time after slaughtering until three ml of blood was aspirated, the blood was immediately poured into a clean and sterile test tube. For preparing serum, the blood which kept in sterile test tube (without anticoagulant) was placed in water bath at 37⁰C for 15 minutes till coagulate was occurred. Then isolation of serum was made by centrifuging (3000 rpm) for 10 minutes and then serum calcium level was determined calorimetrically by spectrophotometer in Laboratory of Hawler teaching Hospital.

Materials and Equipments, figures (1):

A- Equipments:

- 1-Spectrophotometer (CECIL, CE2021, model 2000).
- 2-Therostated water bath
- 3-Centrifuge machine (HITACHI, 05P-21)

B-Materials:

- 1-Test tube
- 2-Micropipet
- 3-Reactive reagent (BIOLABO REF 80004)

Procedure:

Take 0.5 ml of R1 (Calcium Tampon) Buffer solution and it was mixed with 0.5 ml of R2 (Calcium chromogene) so 1ml of working reagent prepared and it was mixed with 25 μ l of the serum of the rabbit, wait for 15minutes at the room temperature 25-30C after that the calcium level was measured by Spectrophotometer at wave length 578 nm. The calcium level was measured according to the following formula:

$$\frac{A_{\text{sample}}}{A_{\text{stander}}} \times C_{\text{stander}} = \text{Calcium concentration mg/dl}$$

A: absorbent of the wave, C: concentration

Clotting time measurement:Apparatus:-

- 1- Capillary tube (without anticoagulant), figure (2).
- 2-Stop watch

Procedure:-

- 1-The stop watch started as soon as the blood appeared in the syringe.
- 2-The capillary tube was filled with the blood quickly.
- 3-Small piece of the tube was broke at (15 second) intervals until the blood shows thread formation.

Coil design: -

The stimulator is attached in series to a pair of coils with a total nominal inductance of (2.4 milliHenry). The diameter of the coils was determined by the size of the tibiae bone of the animals. The coils are wound from 0.060mm diameter insulated copper and attached to the stimulating device by 2m long of 0.7mm² cross-section of cable. The coil was placed directly above the surgical site and fixed to this area by using surgical plaster which can be replaced without any

complication ,a current set at range (0-2 A) producing a field of (0-10mT) at its center it was checked by Teslameter, figure (3). The coils protective covering was made of an prosthodontic cold cure Acrylic resin.

Results**Clotting time:-**

The clotting times were measured in all groups of the rabbits and the effect of magnetic field on the clotting time was observed and the result was shown in table (1) ,from the table it was clear that all clotting time was within normal range .

Table (2) showed that the differences in clotting time between all groups and control group of the rabbit was insignificant (P >0.05).

Biochemical serum analysis

The level of the calcium concentration in the serum was measured at the time of sacrifice in each group of the rabbits as shown in the table (3) and the result indicated that all groups of the rabbit were within normal range .

Statistical analysis by t-test \pm SE was indicated no significant differences between serum calcium levels in all groups of the rabbits table (4).

Discussion

Some researchers showed increase in clotting time and decreasing in serum calcium level when the rabbits whole body exposed to a magnetic field for different period of time^{2,3,4,6,8}.while in our study all groups showed normal clotting time and calcium level because of the low magnetic field of the implant and the pulsed and static magnetic field used in this study had a local effect on tibiae bone so they showed no systemic effect on clotting time and serum

calcium level while previous studies showed increase in clotting time and they correlated that to decreasing in calcium level in the blood which is important ion for coagulation of the blood and its participated in bone tissue calcification^{9,10}. So this factor also may be attributed to acceleration of bone healing and coagulation of blood. Table (1) shows the results of the clotting times in all groups of the rabbits which were indicated no clear or notice effects of using local magnetic implant on clotting time of all rabbits, therefore table(2) shows insignificant effect of magnetic field (p.value> 0.01) between all groups of the rabbits and this result disagree with [Payman,1995 ; Kakai, 1995and Shenzhi *et al.*, 2007]^{3,11,12} and this could be due to small area of the body of the rabbit will be effected and also the local effect of small field strength (50 gauss) as it does not cause harmful effect like stronger magnetic field and this result was agree with the results of [Robert, 2001] which indicated in his research that the low electromagnetic field had improvement effects in the morphology of the red blood cells which participants in health condition of all other cells in the body⁵.

Tables (3and 4) reveling the effect of local magnetic field on calcium level in all groups of the rabbits and from these results which indicated no noticeable effects of using local pulsed (50 Gauss) magnetic field on serum calcium level and this is could be due to small area of the body of the rabbit will be effected and also the local effect of small field strength and this results agree with results of [Saygili,1999 and Belossi *et al.*, 2000,Pilla 1997,Ohy 2003] as they found no effect of using local magnetic field on blood composition elements,^{13,14,15,16} and the results disagree with [Payman, 1995, Tashaev, 1997, Korpinar, 2002,and Peerkhdir,

1995] as they showed decreasing in calcium level in blood serum as they used whole body exposure with greater magnetic field strength in their studies and they contribute this effect with increasing in the metabolic activities of the cells of the tissues and they found a positive effect of magnetic field with bone marrow and immune system^{3,4,6,8}.

Conclusion

Both static and pulsed magnetic at field strength (50 gauss) shows no harmful effect on clotting time and serum calcium level when it was used locally for intraosseous implant of the rabbit.

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Table (1): Clotting time (in minutes) for all groups of the rabbits

Weeks	Control group titanium Implant (A group)	Titanium implant With magnetic (B group)	Gold implant (C group)	Gold implant with magnetic (D group)
2	2.30	2.45	2.45	3.15
4	2.45	2.30	3.0	3.00
6	2.30	2.45	2.45	2.15
8	3.15	3.15	2.45	2.30

Table (2): Statistical analysis by t-test±SE for the differences in Clotting times (miutes) between all groups of the rabbits.

Groups	Control Titanium implant (A)	Magnetic Titanium implant(B)	Control Gold implant(C)	Magnetic Gold implant(D)
Mean±SE	2.5± 0.25	2.5±0.19	2.5±0.13	2.6±0.24
Cal.t	t =0.52		t =0.28	
P value	P =0.63(NS)		P =0.79(NS)	

P value < 0.01 significant; NS: Non significant

Table (3): Serum calcium level (mg/100ml) at sacrifice day.

Weeks	Control group titanium Implant (A group)	Titanium implant With magnetic (B group)	Gold implant (C group)	Gold implant with magnetic (D group)
2	10.5	11.2	10.7	10.9
4	11.3	10.6	10.3	11.2
6	10.8	10.4	10.8	11.6
8	10.4	11.3	11.1	10.7

Table (4): Statistical analysis by t-test ±SE for the differences in calcium mean level (mg/100ml) between all groups of the rabbits.

Groups	Control Titanium implant (A)	Magnetic Titanium implant(B)	Control Gold implant(C)	Magnetic Gold implant(D)
Mean±SE	10.7± 0.20	10.8±0.22	10.7±0.16	11.1±0.19

P value < 0.05 significant NS: Non significant



(A)



(B)



(C)



(D)

Figure (1): Materials and equipments used For Biochemical serum analysis, A: Serum separated from clotted blood, B: Reactive reagent (BIOLABO REF 80004), (C): Centrifuge machine, (D): Spectrophotometer .



(B)



(A)

Figure (2) A: Capillary tube without anticoagulant, B: blood taken from the veins at base of the ear of the rabbit.



a



b



c

Figures (3)a.the rabbit with wire for coil stimulation b. Wire and coils , c. Oscilloscope, Power supply and Teslameter