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ORTHODONTIC TOOTH MOVEMENT IN LOW-DOSE FOR DIFFERENT COURSES METHYLPREDNISOLONE-TREATED RATS

Harraa S. Mohammed -Salih
Orthodontic Department/ College of Dentistry/ Baghdad University

ABSTRACT
The aim of this study is to investigate the effect of different courses of glucocorticosteroid treatment on orthodontic tooth movement (OTM). A 'Split- mouth design performing orthodontic tooth movement in 30 male Wistar albino rats divided into three groups: control (n = 10), short-course (n = 10) and long-course (n = 10). Short and long-course groups received corticosteroid treatment (5 mg/kg/day of methylprednisolone) for 3 and 7 weeks, respectively, while no pharmacological treatment was performed in the control group. The upper right 1st molar was moved mesially for 21 days in all three groups with a closing-coil spring delivering 20 g of force to cause orthodontic tooth movement by means of fixed orthodontic appliance. Measurements were carried using a digital caliper at 0, 1, 2 and 3 wks after appliance placement. Calculations of the differences between the I-M distances at the appliance and non-appliance sides were considered to represent the actual experimental tooth movement caused by the orthodontic appliance. The results showed that the amount of orthodontic tooth movement is significantly greater in the short-course group compared with the long-course and control groups at the same magnitude after 1, 2 and 3 weeks of OTM, ($P < 0.001$). Higher rate of OTM was found in steroid treated groups than control group. Among steroid treated groups short-course group showed higher rate of OTM compared to long-course group, therefore; the clinical consequences could induce a passive treatment phase during the periods where steroids are administered.

KEY WORDS: Steroid; orthodontic tooth movement; rats.

INTRODUCTION

Orthodontic tooth movement (OTM) has been defined as a result of a biologic response to interference in the physiologic equilibrium of the dentofacial complex by an externally applied force[1]. It has been shown that OTM can be influenced by general and local pharmacological modulation; patients requiring orthodontic treatment can be anticipated to present variations from normal bone turnover due to metabolic disease or medication, e.g. steroid treatment of allergies[2]. Orthodontic patients may be affected by systemic diseases that need medical treatment with drugs that could possibly affect bone metabolism[3]. Corticosteroids are a class of steroid hormones, produced in the adrenal cortex. They are involved in many physiologic systems, such as stress response, inflammatory and immune responses, carbohydrate metabolism, protein catabolism, and blood electrolyte levels. Corticosteroids are commonly used to treat many different diseases because of their anti-inflammatory effect. Allergy, asthma, dermatitis and eczema are all diseases with high incidence and rapidly increasing prevalence commonly treated with corticosteroids[4]. In physiological doses, corticosteroids are administered to reinforce deficient endogenous hormones. In certain pathological status, much larger doses of glucocorticoids are administered with the following aims: decreasing inflammation, suppressing the immune response, affecting the hemopoietic system and metabolism[5].

Corticosteroid treatment has been shown to interfere with OTM rate and tissue reaction in animal studies. Only a few authors have examined the effects of glucocorticoids on OTM, and no study was found dealing with mineralocorticoids. The glucocorticoids that have been studied are cortisone, prednisolone, and methylprednisolone. Controversial data are available concerning the effect of orthodontic treatment under corticosteroid treatment on OTM .The effect of cortisone on OTM was investigated in rabbits. Cortisone acetate was injected at a dosage of 15 mg/kg/day for 4 days before and during the application of an orthodontic force of approximately 100 cN for 14 days. Compared with the controls, this regimen led to a significant increase in the rate of OTM. Also, the relapse rate was faster in the experimental group than in the control animals[6]. Prednisolone was administered at 1 mg/kg/day in rats for an induction period of 12 days, followed by an experimental period of 12 days. During the latter phase of the study, the first molar was moved mesially with a force of 30 cN. This therapy had no significant effect on the rate of OTM[7]. An experimental design was used in another study in which methylprednisolone was given at a dosage of 8 mg /kg/ day. In the chronic group, an induction period of 7 weeks was used; then OTM was performed for 3 weeks with a force of 25 cN. This led to an increase in the rate of OTM. However, in the acute and control group without an induction period, methylprednisolone had no effect on the rate of OTM[8]. The differences in the results of these studies probably reflect the combined effects of the dosages, the induction periods, the amount of orthodontic force applied and the relative anti-inflammatory activity of the glucocorticoids tested. In the present study methylprednisolone, one of the most widely

used corticosteroids which has almost no mineralcorticoid effects, on bone metabolism in rats was used with therapeutic dosages of 5 mg/kg/day with short and long-course of administration as the same regime and experimental design of kalia et al[8] but with decreasing the dose of the drug to be comparable with low doses of corticosteroid and also the amount of orthodontic force applied was decreased to be 20g to compensate the differences in the results showed in the previous investigations.

MATERIALS AND METHODS

Animals and experimental protocol

Thirty adult male Wistar albino rats, weighing (220-320g) aged 12- week were used for this experiment. The rats were kept in the animal department of (National Center for Drug Control and Research/Baghdad-IRAQ) in separate cages in a12:12 hour light/dark environment at a constant humidity and temperature of 23°C according to the National Research Council's guide for the care and use of laboratory animals and accessed to drinking water *ad libitum* and standard laboratory rat pellets.

According to the pharmacological treatment [8], the rats were randomly divided into three groups:

Group I: a control group (n = 10) without any pharmacological treatment but received orthodontic treatment for 3 weeks (week 1–3).

Group II: a short-course group (n = 10) received Methylprednisolone and orthodontic treatment simultaneously for 3 weeks (week 1–3).

Group III: a long-course group (n = 10) received Methylprednisolone For 7 weeks (week 1–7) and orthodontic treatment for the last 3 weeks (week 5–7).

The experimental groups received 5 mg/kg/day of methylprednisolone (Solu-medrol; Pharmacia NV/SA, Puurs-Belgium) intramuscularly every 24 hrs for the prescribed number of days[9].

Orthodontic Appliance Design

The appliance was inserted under general anaesthesia, induced by an intramuscular injection of a mixture of ketamine (90 mg/kg body weight) and xylazine (10mg/kg body weight). Orthodontic tooth movement was generated by the insertion of a stainless steel ligature-wire with a diameter of 0.009" and 4mm in length interdentally between the 1st and 2nd maxillary molars, which looped around the cervical part of the 1st molar. It was ligated tightly to ensure maximum stabilization of the wire to which a closing-coil spring (9 mm in length, Dentaurum, Germany) was attached, and the end of the wire was bent carefully toward the buccal surface of the tooth by double ended-ligature tucker to avoid any mechanical trauma to the surrounding oral tissues and the slippage of the coil. To compensate the conical shape of the rats' incisors and subsequently prevent the slippage of the wire as well as the appliance, a TEC-TORQUE, angled hand piece (W&H-Austria) with an inverted-cone bur was used to make grooves cervically on the disto-labial surfaces of both maxillary incisors to which another preformed stainless steel ligature wire, with a diameter of 0.009" and 5mm length, was looped. The ligature wire ligated tightly to which the other end of the closing-coil spring was attached, so that the closing-coil spring of fixed orthodontic appliance was delivering a total orthodontic force of 20 g for mesial traction of maxillary 1st molar was measured by pressure-gauge (CORBLX, Dentarum – Germany). As the closing-coil spring was being attached to the ligature wire, the end of the ligature wire had been carefully adapted toward the distal grooves by using Adam's plier. In order to avoid any mechanical trauma from the appliance to the surrounding oral tissues and to ensure maximum stability of the appliance, a light-cured filling composite material was added to the maxillary incisors. The appliance was checked weekly to ensure any loose or damage to the appliance. Consequently a mesially directed orthodontic force to the maxillary 1st molar with the incisors were used as anchorage teeth resulted in mesial traction of the 1st molar and space creation between the 1st and 2nd molar teeth. According to Ren et al [10] to limit the influence of inter-animal variation in response to metabolic stimuli, a split-mouth design was used as the right side served as the appliance side and the left side served as the non-appliance side, (Figure 1).

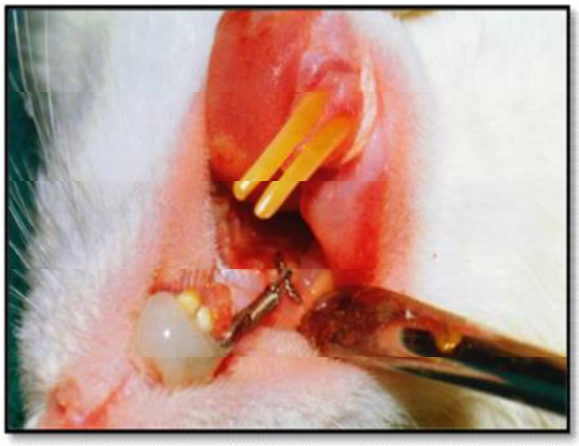

FIGURE 1: Orthodontic fixed appliance in situ.

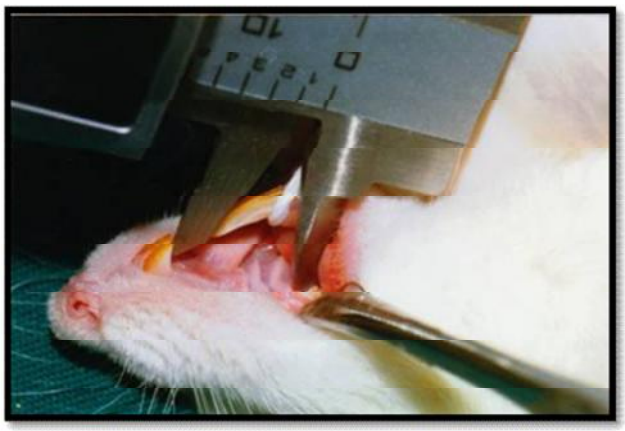

FIGURE 2: Measurements of (I-M distance) at both appliance (right) and non-appliance (left) sides.

Measurements of Orthodontic Tooth Movement

The measurements of the amount of OTM were done clinically. Measurements with a digital caliper (Japan) were performed while the animals were under general anesthesia at 0, 1, 2 and 3 weeks after appliance placement. Calculations of the differences between the

(Incisor-Molar) I-M distances at the appliance and non-appliance sides were considered to represent the actual experimental tooth movement caused by the orthodontic appliance, (Figure 2).

The magnitude of tooth movement was measured as the following numerical expression: ($d_M = d_R - d_L$) Where d_M is the magnitude of tooth movement, d_R is the distance of appliance side (right) and d_L is that of non-appliance side (left), they were expressed as positive values. The final linear measurement value for each case represents the mean of two measurements taken by two observers[11].

STATISTICAL ANALYSIS

The results were expressed by measuring the magnitude of orthodontic tooth movement in mm at both appliance and non-appliance sides at 1, 2, 3 weeks after appliance placement for each group. Statistical analyses were performed; means and standard errors of the mean were calculated for each group. Paired t- test for difference in mean changes after 1, 2, 3 weeks compared to baseline (at 1st day of appliance insertion) for each group at both appliance and non-appliance sides. One-way analysis of variance (ANOVA) for difference in mean changes after 3 week compared to baseline between the 3 study groups was used. LSD was used for the difference in mean between 2 studied groups. A value of $P<0.05$ was considered significant.

RESULTS

The body weight of rats in control group decreased transiently after 1st week till 3rd week but subsequently recovered with steadily weight gained, whereas in short-course group decreased dramatically during the experimental period, and in the long-course group it increased from the beginning of steroid treatment till the end of the 4th week when orthodontic appliance was in place, (Figure 3).

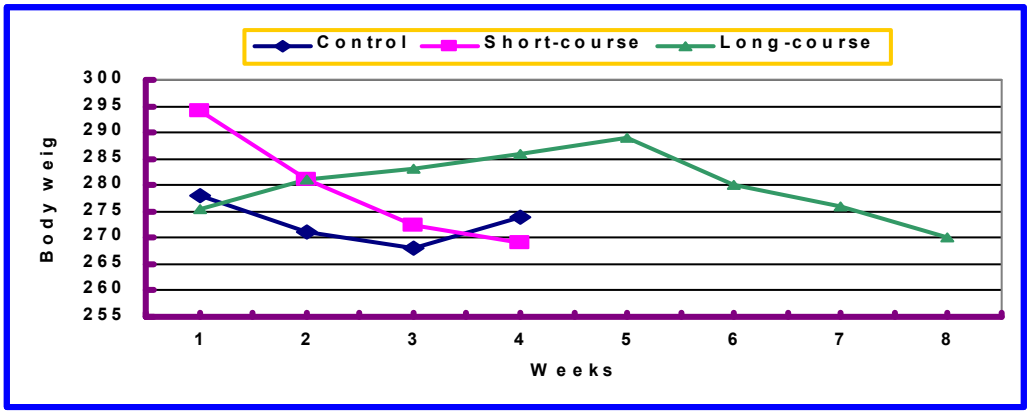

FIGURE 3: Linear chart showing the mean changes in animal body weight of the experimental groups during the experimental period.

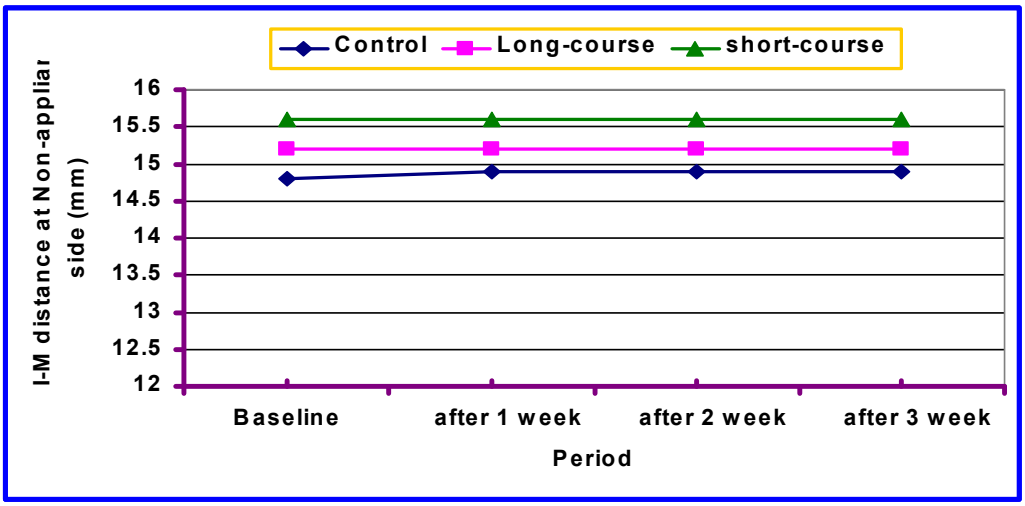

FIGURE 4: Linear chart showing the mean changes in I-M Distance (mm) at Non-appliance side by time and Groups.

TABLE 1: The Difference in Mean Changes in I-M Distance (mm) at Non-appliance side by time and Groups

	Baseline	after 1 week	Changes after 1 week	after 2 weeks	Changes after 2 weeks	after 3 weeks	Changes after 3 weeks
Control							
Range	(13.21 - 16.4)	(13.22 - 16.42)	(-0.01 - 0.03)	(13.22 to 16.41)	(-0.01 to 0.03)	(13.23 to 16.42)	(-0.02 to 0.03)
Mean	14.85	14.86	0.009	14.86	0.013	14.86	0.016
SD	1.032	1.031	0.013	1.028	0.016	1.027	0.016
SE	0.326	0.326	0.004	0.325	0.005	0.325	0.005
N	10	10	10	10	10	10	10
P (Paired t-test)			0.05[NS]		0.028		0.011
Short-course							
Range	(14.44 to 16.62)	(14.43 to 16.64)	(-0.01 to 0.02)	(14.43 to 16.64)	(-0.01 to 0.03)	(14.44 to 16.64)	(0 to 0.03)
Mean	15.58	15.58	0.008	15.59	0.010	15.59	0.015
SD	0.698	0.706	0.010	0.703	0.012	0.703	0.008
SE	0.221	0.223	0.003	0.222	0.004	0.222	0.003
N	10	10	10	10	10	10	10
P (Paired t-test)			0.037		0.023		<0.001
Long-course							
Range	(13.06 to 16.96)	(13.06 to 16.95)	(-0.01 to 0.01)	(13.07 to 16.96)	(-0.01 to 0.01)	(13.07 to 16.97)	(0 to 0.02)
Mean	15.22	15.22	0.002	15.23	0.005	15.23	0.012
SD	1.322	1.319	0.009	1.318	0.007	1.319	0.008
SE	0.418	0.417	0.003	0.417	0.002	0.417	0.002
N	10	10	10	10	10	10	10
P (Paired t-test)			0.51[NS]		0.05[NS]		0.001

Note: All changes were compared to baseline values

TABLE 2: The Difference in Mean Changes in I-M Distance (mm) at Appliance side by time and Groups.

	Baseline	after 1 week	Changes after 1 week	after 2 weeks	Changes after 2 weeks	after 3 weeks	Changes after 3 weeks
Control							
Range	(13.21 to 16.41)	(12.48 to 15.89)	(-0.94 to - 0.52)	(12.19 to 15.59)	(-1.24 to-0.82)	(11.78 to 14.89)	(-1.77 to -1.43)
Mean	14.9	14.1	-0.8	13.8	-1.1	13.3	-1.6
SD	1	1.1	0.1	1.1	0.1	1	0.1
SE	0.33	0.34	0.04	0.34	0.04	0.33	0.03
N	10	10	10	10	10	10	10
P (Paired t-test)			<0.001		<0.001		<0.001
Short-course							
Range	(14.44 to 16.63)	(12.85 to 14.74)	(-1.9 to -1.17)	(12.23 to 14.05)	(-2.62 to -1.54)	(11.36 to 13.32)	(-3.35 to -2.62)
Mean	15.6	13.9	-1.6	13.4	-2.2	12.5	-3.1
SD	0.7	0.6	0.3	0.6	0.4	0.7	0.3
SE	0.22	0.2	0.08	0.2	0.11	0.23	0.08
N	10	10	10	10	10	10	10
P (Paired t-test)			<0.001		<0.001		<0.001
Long-course							
Range	(13.05 to 16.97)	(12.24 to 15.79)	(-1.18 to -0.75)	(11.67 to 15.36)	(-1.69 to -1.17)	(11.17 to 14.92)	(-2.09 to -1.68)
Mean	15.2	14.3	-1	13.8	-1.5	13.3	-1.9
SD	1.3	1.3	0.1	1.3	0.1	1.4	0.1
SE	0.42	0.42	0.04	0.42	0.05	0.44	0.05
N	10	10	10	10	10	10	10
P (Paired t-test)			<0.001		<0.001		<0.001

Note: All changes were compared to baseline values

Orthodontic tooth movement (mm) in the appliance side (I – M distance) was adjusted to non-appliance side (I – M distance) and changes were compared to baseline (I – M distance measured at the first day of appliance insertion) for each group. At the non-appliance side, there was no significant difference in the mean of I – M distance reduction value between the experimental groups after 1st, 2nd and 3rd weeks of OTM (P>0.05), (Table 1, Figure 4). Whereas at the appliance side, there was a highly significant difference in the mean of I – M distance reduction value between the experimental groups after 1st, 2nd and 3rd weeks of OTM (P<0.001), (Table 2, Figure 5). The highest mean value of orthodontic tooth movements (mm) adjusted as appliance side subscribed from non–appliance side after 1st, 2nd and 3rd weeks was in the short-course group then in the long-course group while the least mean value was in the control group. Quantitatively, the mean changes in OTM after 3 weeks compared to baseline were statistically highly significant between the experimental groups (ANOVA, P<0.001), (Table 3, Figure 6).

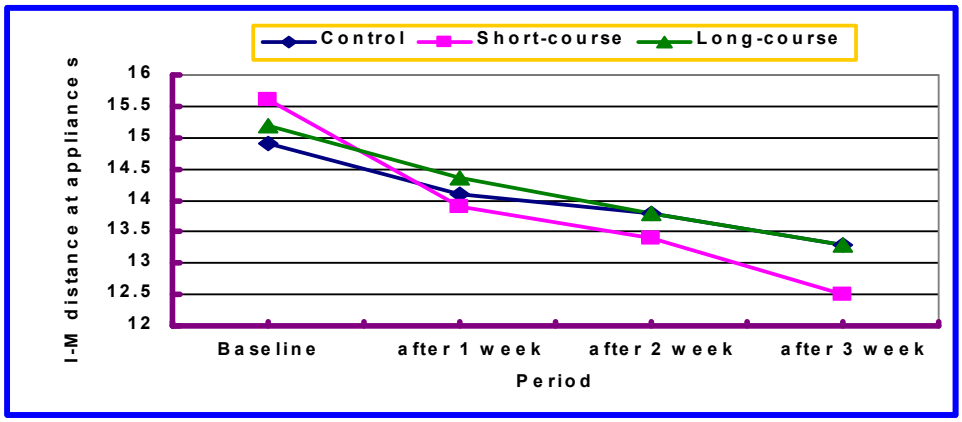

FIGURE 5: Linear chart showing the mean changes in I-M Distance (mm) at Appliance side by time and Groups.

TABLE 3: The Difference in Mean Changes in Orthodontic tooth movement (mm) by time and Groups.

	After 1 week	After 2 weeks	After 3 weeks	Probability
Control				
Range	(-0.93 to -0.54)	(-1.23 to -0.83)	(-1.77 to -1.45)	
Mean	-0.8	-1.1	-1.6	
SD	0.1	0.1	0.1	
SE	0.04	0.04	0.03	
N	10	10	10	
Short-course				
Range	(-1.91 to -1.18)	(-2.64 to -1.55)	(-3.37 to -2.63)	P < 0.001
Mean	-1.6	-2.2	-3.1	
SD	0.3	0.4	0.3	
SE	0.09	0.11	0.08	
N	10	10	10	
Long-course				
Range	(-1.17 to -0.76)	(-1.7 to -1.18)	(-2.11 to -1.69)	
Mean	-1	-1.5	-1.9	
SD	0.1	0.1	0.2	
SE	0.04	0.05	0.05	
N	10	10	10	

*LSD for difference in mean between:
Short-course x Control P <0.001
Long-course x Control P <0.001
Long-course x Short-course P <0.001

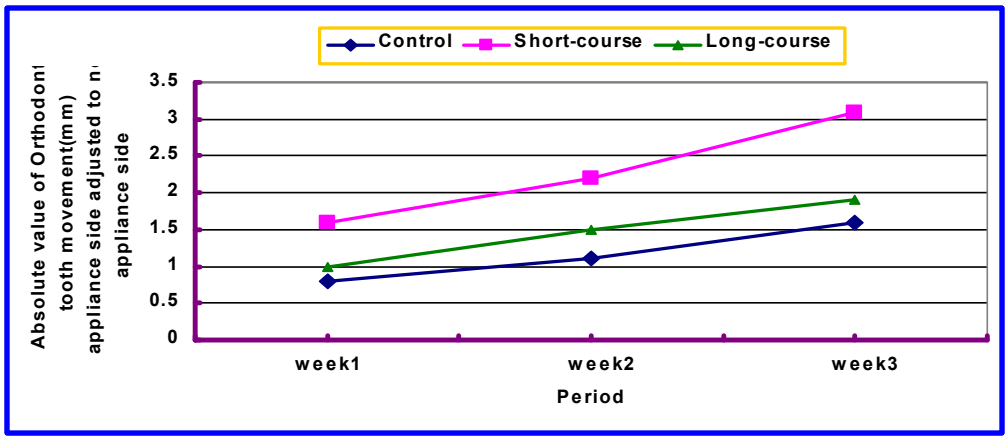

FIGURE 6: Linear chart showing the difference in mean changes in Orthodontic tooth movement (mm) by time and Groups.

DISCUSSION

Corticosteroids are widely used in the treatment of a variety of medical conditions. The results of this study showed an influence of different GC courses and orthodontic treatment on animals' body weight during experimental period. A temporary episode of weight loss in control group was conducted for 1 week following appliance insertion. These findings were in consistent with Ong et al[7]. The Steroid administration resulted in a moderate weight loss in the short-course compared to control group and weight loss was accentuated by the appliance insertion that's verified in the long-course group as weight gain occur from the 1st week of drug administration till the 4th week where orthodontic appliance was inserted. This indicted that the appliance insertion exhibit some discomfort and disability for eating to the animals. Weight gain in the long-course group occur due to the effect of low-dose administration of methylprednisolone for long time which in consistent with Van Balkom et al[12]. All appliance-side 1st molars of the animals showed evidence of tooth movement (reduction in I – M distance measured), with the development of spacing between the first and second molars. No space was observed between the second and third molars, indicating lack of mesial movement of the second molar during the experiment. No tooth movement was evident on the non-appliance side in all experimental groups. In turn, the rate of OTM is of the same magnitude in the three groups, in which the increased rate of OTM has a type of linear relationship that means the rate of increase in the amount of OTM is steady over the experimental period (1-3week). This indicated that the amount of mechanical load exerted by orthodontic appliance on rat molar was the same in all three groups.

In the present study, the results showed that the amount of OTM is significantly greater in the steroid treated groups after 1, 2, and 3week compared with the control group. The rate of OTM increased in both drug groups although the greatest OTM was in the short-course group. The parameters measured had varied between the short and long-course groups indicating that a different clinical reaction can be anticipated in patients undergoing corticosteroid treatment. The explanation is probably different in the two groups. The trend towards increased rate of tooth movement in the short-course group compared to control group could possibly be explained as a reflection of the transition state from the short-course effect to the long-course effect of the drug. This finding is in agreement with Ashcraft et al[6] who performed their study in rabbits, with dosage of (15 mg/kg) and for a period of 14 days showed that the rate of active tooth movement was approximately three to four times greater in steroid group than in controls. Because a general model of increased skeletal resorption has been demonstrated and is attributed to an elevation of osteoclastic activity, it is possible that during conditions stimulating additional resorptive influences (i.e., an orthodontic force) increased resorption might occur. The results of this research tend to support this theory. Whereas a study performed by Ong et al[7] used a lower dosage (1 mg/kg/day) in rats, and they found there was no significant differences in the magnitude of tooth movement between the experimental groups. The same experimental design used in this study was previously performed by Kalia et al[8] in which methylprednisolone was given at a dosage of 8 mg /kg/ day. OTM was performed for 3 weeks with a force of 25 cN. This led to an increase in the rate of OTM in the chronic group whereas methylprednisolone had no effect on the rate of OTM in the acute and control groups. However; direct comparison of these studies with the present data is impossible. Moreover, the treatment regime and duration was not the same except with Kalia et al[8] but he used a dosage of 8 mg/kg/day of methylprednisolone, which is higher than those recommended for more common diseases, such as asthma, but it is comparable with medium and high-oral doses prescribed for inflammatory diseases such as rheumatoid arthritis and renal diseases[14]. While in the present study a dosage of 5 mg/kg/day which is comparable with low-oral doses recommended for more common diseases to keep the detrimental effects of bone loss minimal[9].

However, normal bone remodeling process is fundamental to orthodontics; the increased rate of OTM in the short-course group compared to long-course group could be explained by the effect of GCs on bone remodeling process. There is evidence that during the initial administration of corticosteroids, a period of very rapid bone loss occurs that may not be totally representative of longer-course administration or other chronic conditions[15]. This could be ascribed to the lack of balance between formation activities (inhibited by the drug) and the resorption activities (enhanced by drug administration) occurring in the initial phase of drug administration[16, 17], whereby the long-course group have been reached to a steady state of bone remodeling process during the 4 weeks of drug administration preceding the appliance insertion resulting in accelerating in the rate of OTM compared to the control group. These findings are in accordance with previous investigation that showed increase in the rate of OTM in animals with increased bone turnover rate than in the control group[3].

CONCLUSION & CLINICAL CONSIDERATIONS

1. Increased rate of OTM can be expected with short-course corticosteroid therapy. As asthma and other allergic symptoms are in fact often treated in shorter periods of time; therefore, the orthodontist need to be particularly aware of any adverse clinical consequences of sudden use of GCs by the patient that may lead to undesirable tooth movement and may influence the outcome of mechanotherapy.
2. Another possible option may be, if possible, to postpone the start of orthodontic treatment till the acute phase of the disease is finished.
3. It is imperative that the orthodontist pays close attention to the drug consumption history of each and every patient, before and during the course of orthodontic treatment.

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