

## Effects of PGI<sub>2</sub> and TxA<sub>2</sub> Analogs and Inhibitors in Orthodontic Tooth Movement

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**Abstract:** This study evaluates the effects of prostacyclin (PGI<sub>2</sub>) and thromboxane A<sub>2</sub> (TxA<sub>2</sub>) in orthodontic tooth movement and osteoclastic activity in rats. The study sample consisted of 150 male Sprague-Dawley rats. The rats were randomly divided into five equal groups, and each group was again equally divided into three subgroups (SGs). Twenty grams of reciprocal force was applied to maxillary incisors of the rats with a spring bent from 0.35 mm stainless steel wire, except for the rats in the last SG. Iloprost (PGI<sub>2</sub> analog), indomethacin (PGI<sub>2</sub> inhibitor), U 46619 (TxA<sub>2</sub> analog), and imidazole (TxA<sub>2</sub> inhibitor) were dissolved in 0.9% NaCl (saline solution), and each material was prepared in three different concentrations (10<sup>-4</sup>, 10<sup>-5</sup>, and 10<sup>-6</sup> M/L). Iloprost was administered (20 µL/12 hours) in the first three SGs with the sequence of 10<sup>-4</sup>, 10<sup>-5</sup>, and 10<sup>-6</sup> M/L. Indomethacin, U 46619, and imidazole were administered in the next nine SGs with the same sequence and dose. In SG 13, 0.9% NaCl solution was administered (20 µL/12 hours) to the rats together with orthodontic force. Only orthodontic force was not used in SG 14, and neither any solution nor orthodontic force was used in the last SG. The rats were sacrificed on the fifth day of the experiment, premaxillae were dissected, and cross samples were taken. The results showed that PGI<sub>2</sub> and TxA<sub>2</sub> analogs increased the number of multinuclear osteoclasts, osteoclastic bone resorption, and rate of orthodontic tooth movement. (*Angle Orthod* 2004;74:526–532.)

**Key Words:** Osteoclast; Prostacyclin; Thromboxane A<sub>2</sub>; Orthodontic tooth movement

### INTRODUCTION

Orthodontic tooth movement requires remodeling of the alveolar bone,<sup>1</sup> and prostaglandins (PGs), cyclic adenosine monophosphate, and cyclic guanosine monophosphate have been suggested to be of major importance in bone remodeling.<sup>2–6</sup> Prostaglandin E<sub>1</sub> (PGE<sub>1</sub>) and prostaglandin E<sub>2</sub> (PGE<sub>2</sub>) have been of interest in the majority of previous studies in orthodontics, and it was reported that these arachidonic acid metabolites increased the rate of orthodontic tooth movement in humans<sup>7</sup> and animals.<sup>8–13</sup>

PGI<sub>2</sub> and TxA<sub>2</sub> are two other arachidonic acid metabolites that were shown to be synthesized in human body, and they have been extensively investigated in various fields of

medical science.<sup>14–22</sup> However, it has also been observed in a number of studies<sup>19–22</sup> that there was an inverse proportion between these two mediators. Grodzinska and Marcinkiewicz<sup>19</sup> reported that PGI<sub>2</sub> administration decreased TxA<sub>2</sub> production in platelet-rich plasma. Hanazaki et al<sup>20</sup> observed in their study that warm ischemic liver damage, in mongrel dogs, was protected by PGE<sub>1</sub> administration by way of suppressing the increased TxA<sub>2</sub> production and increasing PGI<sub>2</sub> production. Makino and Kamata<sup>21</sup> also found an inversely proportional relationship between PGI<sub>2</sub> and TxA<sub>2</sub> in an experimental study with diabetic rats.

It was shown that, PGE<sub>2</sub>, PGI<sub>2</sub>, and TxA<sub>2</sub> were at higher levels in inflammatory tissues, and these metabolites played an important role in periodontal disease. ElAttar and Lin<sup>23</sup> suggested that PG levels were considerably higher in inflammatory gingival tissues. Dewhirst et al<sup>24</sup> indicated that PGI<sub>2</sub> synthesis was increased in the inflammatory gingival tissues, and TxA<sub>2</sub> was mostly found deep in periodontal pockets. Rifkin and Tai<sup>25</sup> found significantly higher levels of TxA<sub>2</sub> in inflammatory periodontal tissues of dogs but could not definitely determine the relationship between TxA<sub>2</sub> and bone loss. Saito et al<sup>26</sup> used 9,11-epithio-11,12-methano thromboxane A<sub>2</sub> (a stable TxA<sub>2</sub> analog) in mouse marrow culture and observed that multinuclear osteoclast-like cells were increased.

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Accepted: August 2003. Submitted: May 2003.

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Although  $\text{PGE}_1$  and  $\text{PGE}_2$  are well known to increase the rate of tooth movement, a literature review shows that  $\text{PGI}_2$  and  $\text{TxA}_2$  were not evaluated extensively in orthodontics. This study compares the effects of  $\text{PGI}_2$  and  $\text{TxA}_2$  analogs and inhibitors on orthodontic tooth movement.

## MATERIALS AND METHODS

The study comprised 150 adult male Sprague-Dawley rats of approximately the same age with an average initial weight of  $196.95 \pm 33.917$  g. The rats were randomly divided into five equal groups, with 30 rats in each group. Each group was again divided equally into three subgroups (SGs), and 15 SGs were obtained. The animals were fed a standard pellet diet with tap water ad libitum. The rats were anesthetized with enflurane inhalation anesthetic (10 mg/kg) before various procedures.

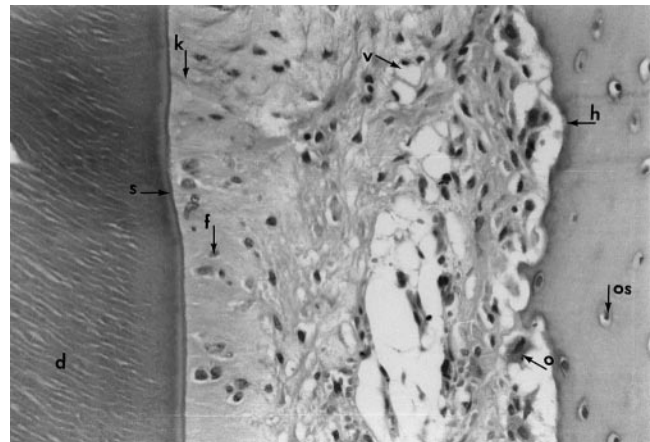
Orthodontic force was applied in all the SGs except the last SG. Holes were prepared on maxillary incisors of the rats, and 20 g of reciprocal force was applied to the teeth with a spring bent from 0.35 mm stainless steel wire. The springs were placed on a grid and activated as a single arm with a plier. The force was measured with a gauge, and the springs were not reactivated during the experiment. Direct linear measurements of tooth separation were recorded at days 1 and 5 between the mesial corners of the upper incisors by two authors using a sliding caliper. Occlusal radiographs of all the rats were obtained in the beginning and at the end of the experiment.

Iloprost ( $\text{PGI}_2$  analog), indomethacin ( $\text{PGI}_2$  inhibitor), U 46619 ( $\text{TxA}_2$  analog), and imidazole ( $\text{TxA}_2$  inhibitor) were dissolved in 0.9% NaCl (saline solution). Each material was prepared in three different concentrations,  $10^{-4}$ ,  $10^{-5}$ , and  $10^{-6}$  M/L. Iloprost was administered (20  $\mu\text{L}/12$  hours) in SGs 1, 2, and 3 as  $10^{-4}$  M/L in the first,  $10^{-5}$  M/L in the second, and  $10^{-6}$  M/L in the third SG. Indomethacin (SGs 4, 5, and 6), U 46619 (SGs 7, 8, and 9), and imidazole (SGs 10, 11, and 12) were administered to these nine SGs, respectively, with the same sequence and dose. The final three SGs were evaluated as control SGs. In SG 13, 0.9% NaCl solution was administered to the rats, together with orthodontic force, with the same amount and prescription (20  $\mu\text{L}/12$  hours). The experimental solutions were injected into the subperiosteum area adjacent to the left and right upper incisors. Only orthodontic force was administered in SG 14, and neither any solution nor orthodontic force was used in the last SG. The rats were monitored throughout the experiment. On the fifth day of the experiment, the rats were sacrificed, and the premaxillae were dissected and placed in 10% formalin. After fixation, the springs were removed, and the premaxillae were decalcified with 9% formic acid.

The decalcified premaxillae were then fixed again in the same manner and hemisectioned into block sections at the coronal, middle, and apical thirds of the right and left upper



**FIGURE 1.** Coronal section obtained from premaxilla (d, dentin; m, enamel; p, pulp; f, fibroblast; pm, periodontal membrane; and ak, alveolar bone).

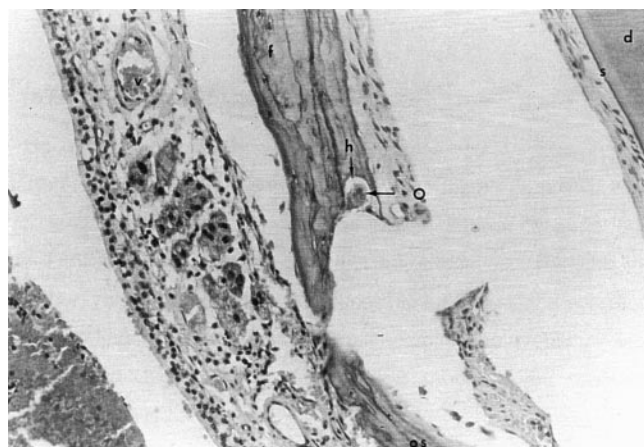


**FIGURE 2.** Stained section of an iloprost subgroup sample. Osteoclast (o), Howship lacuna (h), capillaries (v), cement (s), collagen fibers (k), fibroblast (f), osteosit (os), and dentin (d).

incisor roots. The sections were obtained perpendicular to the roots of the teeth (Figure 1). The sections were washed, trimmed, and run through routine paraffin embedment. The paraffin blocks were serially sectioned at four- to six- $\mu\text{m}$  intervals in the frontal plane. The sections were mounted on glass microscope slides and stained with hematoxylin and eosin. Multinuclear osteoclasts on the stained sections were counted by two pathologists, twice, at different times using a light microscope (Figures 2 and 3). The experiment was carried out according to the guidelines for the use of experimental animals of Gulhane Military Medical Academy.

## Statistical analysis

The statistics were performed by SPSS 10.0 (SPSSFW, SPSS Inc, Chicago, Ill) statistical package. Descriptive statistics were shown as mean  $\pm$  standard deviation (Table 1). The intragroup differences were investigated by Kruskal-



**FIGURE 3.** Stained section of an imidazole subgroup sample. Osteoclast (o), Howship lacuna (h), capillaries (v), cement (s), fibroblast (f), osteositis (os), and dentin (d).

Wallis test. The intersubgroup differences of the findings were determined with Mann-Whitney *U*-test (with Bonferroni correction). *P* values less than or equal to .005 were evaluated as statistically significant.<sup>27</sup>

## RESULTS

### Intragroup differences

Statistically significant differences were found at osteoclasts (O) in the coronal (c) ( $P = .003$ ) and middle (m) ( $P = .004$ ) thirds of the roots and for the intracincisal measurements (M, in mm) ( $P = 3.404 \times 10^{-4}$ ) in the imidazole group. *P* values were significant in all the parameters in the control group, and they were found as  $P = 4.409 \times 10^{-4}$ ,  $P = 1.940 \times 10^{-4}$ ,  $P = 4.077 \times 10^{-4}$ , and  $P = 2.699 \times 10^{-4}$ , respectively (Table 2).

### Comparison of the analogs, inhibitors, and control SGs within the groups

The differences between SGs 1 and 3 were significant at O (c) ( $P = .001$ ), O (m) ( $P = .004$ ), and O (apical [a]) ( $P = .004$ ), and the differences between SGs 4 and 6 were significant at O (c) ( $P = .004$ ) and O (a) ( $P = .004$ ). The difference at M (mm) ( $P = .001$ ) was the only significant finding in the comparison of SGs 10 and 11, whereas the differences at O (c) ( $P = .002$ ), O (m) ( $P = .002$ ), and M (mm) ( $P = .001$ ) were found significant when SGs 10 and 12 were compared with each other. In the evaluation of the

**TABLE 1.** Arithmetic Means and  $\pm$ Standard Deviations of Osteoclasts (O) in the Coronal (c), Middle (m), and Apical (a) Thirds of the Roots and Intracincisal Measurements (M) at the End of the Experiment

| Subgroups                     | O (c) (psi) |       | O (m) (psi) |       | O (a) (psi) |       | M (mm) |       |
|-------------------------------|-------------|-------|-------------|-------|-------------|-------|--------|-------|
|                               | Mean        | SD    | Mean        | SD    | Mean        | SD    | Mean   | SD    |
| 1—Iloprost ( $10^{-4}$ )      | 13.300      | 0.675 | 7.900       | 1.100 | 10.200      | 0.789 | 5.900  | 0.738 |
| 2—Iloprost ( $10^{-5}$ )      | 12.500      | 1.080 | 7.500       | 1.080 | 9.400       | 1.075 | 5.900  | 0.737 |
| 3—Iloprost ( $10^{-6}$ )      | 11.900      | 0.738 | 6.900       | 0.738 | 8.900       | 0.738 | 5.000  | 0.667 |
| 4—Indomethacin ( $10^{-4}$ )  | 6.900       | 0.738 | 3.000       | 0.667 | 4.000       | 0.667 | 3.200  | 0.789 |
| 5—Indomethacin ( $10^{-5}$ )  | 7.600       | 0.699 | 3.300       | 0.823 | 4.600       | 0.699 | 3.600  | 0.966 |
| 6—Indomethacin ( $10^{-6}$ )  | 8.200       | 0.789 | 3.500       | 0.527 | 5.200       | 0.788 | 4.100  | 0.738 |
| 7—U 46619 ( $10^{-4}$ )       | 11.500      | 1.269 | 6.300       | 0.949 | 8.500       | 1.269 | 5.700  | 0.675 |
| 8—U 46619 ( $10^{-5}$ )       | 11.300      | 0.948 | 5.800       | 1.032 | 8.300       | 0.948 | 5.800  | 0.919 |
| 9—U 46619 ( $10^{-6}$ )       | 9.900       | 0.994 | 5.000       | 1.154 | 6.900       | 0.994 | 4.700  | 0.675 |
| 10—Imidazole ( $10^{-4}$ )    | 7.200       | 0.919 | 2.800       | 0.632 | 4.400       | 0.699 | 3.300  | 0.675 |
| 11—Imidazole ( $10^{-5}$ )    | 8.000       | 0.816 | 3.300       | 0.675 | 4.600       | 0.699 | 4.700  | 0.675 |
| 12—Imidazole ( $10^{-6}$ )    | 8.800       | 0.789 | 4.000       | 0.667 | 4.900       | 1.000 | 5.000  | 0.816 |
| 13—Saline + force (control 1) | 8.100       | 1.197 | 3.200       | 1.033 | 5.500       | 0.849 | 4.500  | 0.527 |
| 14—Force (control 2)          | 8.200       | 0.919 | 3.200       | 0.920 | 5.500       | 0.972 | 4.500  | 0.527 |
| 15—Control 3                  | 1.700       | 0.675 | 1.300       | 0.483 | 1.700       | 0.675 | 0.170  | 0.067 |

**TABLE 2.** Intragroup Differences of the Analog, Inhibitor, and Control Groups

| Groups       | O (c) (psi) |                          | O (m) (psi) |                          | O (a) (psi) |                          | M (mm)   |                          |
|--------------|-------------|--------------------------|-------------|--------------------------|-------------|--------------------------|----------|--------------------------|
|              | $\chi^2$    | <i>P</i>                 | $\chi^2$    | <i>P</i>                 | $\chi^2$    | <i>P</i>                 | $\chi^2$ | <i>P</i>                 |
| Iloprost     | 10.030      | .007                     | 9.016       | .011                     | 8.501       | .014                     | 9.132    | .010                     |
| Indomethacin | 10.144      | .006                     | 2.842       | .241                     | 9.553       | .008                     | 5.148    | .084                     |
| U 46619      | 9.154       | .010                     | 6.184       | .045                     | 9.154       | .010                     | 9.473    | .009                     |
| Imidazole    | 11.377      | .003*                    | 11.226      | .004*                    | 1.418       | .492                     | 15.971   | $3.404 \times 10^{-4}$ * |
| Control      | 20.058      | $4.409 \times 10^{-4}$ * | 17.095      | $1.940 \times 10^{-4}$ * | 20.215      | $4.077 \times 10^{-4}$ * | 21.040   | $2.699 \times 10^{-4}$ * |

\* Significance level  $\alpha = .005$ .



**TABLE 3.** Comparison of the Analogs (1, 2, 3, 7, 8, 9), Inhibitors (4, 5, 6, 10, 11, 12), and Control (13, 14, 15) Subgroups Within the Groups

| Subgroups   | O (c) (psi) |                            | O (m) (psi) |                            | O (a) (psi) |                            | M (mm) |                            |
|---|-------------|----------------------------|-------------|----------------------------|-------------|----------------------------|--------|----------------------------|
|   | Z           | P                          | Z           | P                          | Z           | P                          | Z      | P                          |
| 1 (10 <sup>-4</sup> )–2 (10 <sup>-5</sup> ) (PGI <sub>2</sub> )   | 1.751       | .105                       | 0.823       | .436                       | 1.734       | .105                       | 0.267  | .853                       |
| 1 (10 <sup>-4</sup> )–3 (10 <sup>-6</sup> ) (PGI <sub>2</sub> )   | 3.190       | .001*                      | 2.914       | .004*                      | 2.914       | .004*                      | 2.437  | .023                       |
| 2 (10 <sup>-5</sup> )–3 (10 <sup>-6</sup> ) (PGI <sub>2</sub> )   | 1.310       | .218                       | 1.310       | .218                       | 1.044       | .353                       | 2.711  | .011                       |
| 4 (10 <sup>-4</sup> )–5 (10 <sup>-5</sup> ) (PGI <sub>2</sub> )   | 1.931       | .075                       | 0.773       | .529                       | 1.767       | .123                       | 0.881  | .436                       |
| 4 (10 <sup>-4</sup> )–6 (10 <sup>-5</sup> ) (PGI <sub>2</sub> )   | 2.914       | .004*                      | 1.699       | .143                       | 2.865       | .004*                      | 2.257  | .035                       |
| 5 (10 <sup>-4</sup> )–6 (10 <sup>-5</sup> ) (PGI <sub>2</sub> )   | 1.693       | .123                       | 0.849       | .481                       | 1.693       | .123                       | 1.238  | .247                       |
| 7 (10 <sup>-4</sup> )–8 (10 <sup>-5</sup> ) (TxA <sub>2</sub> )   | 0.198       | .853                       | 1.270       | .247                       | 0.198       | .853                       | 0.452  | .684                       |
| 7 (10 <sup>-4</sup> )–9 (10 <sup>-6</sup> ) (TxA <sub>2</sub> )   | 2.604       | .011                       | 2.310       | .023                       | 2.604       | .011                       | 2.706  | .009                       |
| 8 (10 <sup>-5</sup> )–9 (10 <sup>-6</sup> ) (TxA <sub>2</sub> )   | 2.607       | .011                       | 1.527       | .143                       | 2.607       | .011                       | 2.567  | .011                       |
| 10 (10 <sup>-4</sup> )–11 (10 <sup>-5</sup> ) (TxA <sub>2</sub> ) | 1.764       | .105                       | 1.636       | .143                       | 0.418       | .739                       | 3.334  | .001*                      |
| 10 (10 <sup>-4</sup> )–12 (10 <sup>-6</sup> ) (TxA <sub>2</sub> ) | 3.191       | .002*                      | 3.123       | .002*                      | 1.149       | .315                       | 3.445  | .001*                      |
| 11 (10 <sup>-5</sup> )–12 (10 <sup>-6</sup> ) (TxA <sub>2</sub> ) | 1.922       | .075                       | 2.071       | .063                       | 0.771       | .481                       | 0.856  | .436                       |
| 13 (s + f)–14 (f) (control)                                       | 0.198       | .853                       | 0.080       | .971                       | 0.121       | .912                       | 0.000  | 1.000                      |
| 13 (s + f)–15 (c3) (control)                                      | 3.860       | 1.082 × 10 <sup>-5</sup> * | 3.594       | 1.299 × 10 <sup>-4</sup> * | 3.856       | 1.082 × 10 <sup>-5</sup> * | 3.883  | 1.082 × 10 <sup>-5</sup>   |
| 14 (f)–15 (c3) (control)  | 3.836       | 1.082 × 10 <sup>-5</sup> * | 3.575       | 1.299 × 10 <sup>-4</sup> * | 3.853       | 1.082 × 10 <sup>-5</sup> * | 3.883  | 1.082 × 10 <sup>-5</sup> * |

\* Significance level  $\alpha = .005$ .**TABLE 4.** Comparison of the Analog and Inhibitor Subgroups with Saline + Force (SF) Subgroup

| Subgroups                             | O (c) (psi) |                            | O (m) (psi) |                            | O (a) (psi) |                            | M (mm) |                            |
|---------------------------------------|-------------|----------------------------|-------------|----------------------------|-------------|----------------------------|--------|----------------------------|
|                                       | Z           | P                          | Z           | P                          | Z           | P                          | Z      | P                          |
| 1-Iloprost (10 <sup>-4</sup> )–SF     | 3.836       | 1.082 × 10 <sup>-5</sup> * | 3.827       | 1.082 × 10 <sup>-5</sup> * | 3.839       | 1.082 × 10 <sup>-5</sup> * | 3.375  | 4.871 × 10 <sup>-4</sup> * |
| 2-Iloprost (10 <sup>-5</sup> )–SF     | 3.807       | 1.082 × 10 <sup>-5</sup> * | 3.891       | 1.083 × 10 <sup>-5</sup> * | 3.827       | 1.082 × 10 <sup>-5</sup> * | 3.395  | 4.871 × 10 <sup>-4</sup> * |
| 3-Iloprost (10 <sup>-6</sup> )–SF     | 3.829       | 1.082 × 10 <sup>-5</sup> * | 3.833       | 1.082 × 10 <sup>-5</sup> * | 3.845       | 1.082 × 10 <sup>-5</sup> * | 1.699  | .143                       |
| 4-Indomethacin (10 <sup>-4</sup> )–SF | 2.310       | .023                       | 0.403       | .739                       | 3.225       | .001*                      | 3.212  | .002*                      |
| 5-Indomethacin (10 <sup>-5</sup> )–SF | 1.150       | .280                       | 0.241       | .853                       | 2.277       | .029                       | 2.207  | .035                       |
| 6-Indomethacin (10 <sup>-6</sup> )–SF | 0.159       | .912                       | 0.810       | .481                       | 0.729       | .529                       | 1.258  | .280                       |
| 7-U 46619 (10 <sup>-4</sup> )–SF      | 3.738       | 2.165 × 10 <sup>-5</sup> * | 3.747       | 2.165 × 10 <sup>-5</sup> * | 3.754       | 2.165 × 10 <sup>-5</sup> * | 3.224  | .002*                      |
| 8-U 46619 (10 <sup>-5</sup> )–SF      | 3.742       | 2.165 × 10 <sup>-5</sup> * | 3.662       | 4.333 × 10 <sup>-5</sup> * | 3.758       | 2.165 × 10 <sup>-5</sup> * | 2.962  | .003*                      |
| 9-U 46619 (10 <sup>-6</sup> )–SF      | 2.908       | .004*                      | 2.905       | .004*                      | 2.758       | .007                       | 0.640  | .522                       |
| 10-Imidazole (10 <sup>-4</sup> )–SF   | 1.735       | .105                       | 0.888       | .436                       | 2.648       | .011                       | 3.224  | .002*                      |
| 11-Imidazole (10 <sup>-5</sup> )–SF   | 0.276       | .796                       | 0.321       | .796                       | 2.277       | .029                       | 0.640  | .579                       |
| 12-Imidazole (10 <sup>-6</sup> )–SF   | 1.346       | .218                       | 1.844       | .089                       | 1.401       | .190                       | 1.440  | .190                       |

\* Significance level  $\alpha = .005$ .

control SGs, significant differences were found at all the parameters between SG 13–SG 15 and SG 14–SG 15. In both the comparisons the differences were found as  $P = 1.082 \times 10^{-5}$ ,  $P = 1.299 \times 10^{-4}$ ,  $P = 1.082 \times 10^{-5}$ , and  $P = 1.082 \times 10^{-5}$ , respectively (Table 3).

### Comparison of the analog and inhibitor SGs with saline + force SG

Statistically significant differences were observed in the comparisons of SG 1 and saline + force (SF) and SG 2–SF. In both the comparisons,  $P$  values at O (c), O (m), and O (a) were found to be  $1.082 \times 10^{-5}$ , whereas it was  $4.871 \times 10^{-4}$  at M (mm). Significant differences were also observed at O (c) ( $P = 1.082 \times 10^{-5}$ ), O (m) ( $P = 1.082 \times 10^{-5}$ ), and O (a) ( $P = 1.082 \times 10^{-5}$ ) in the SG 3–SF comparison. The differences were significant at O (a) ( $P = .001$ ) and M (mm) ( $P = .002$ ) when SG 4 and SF were compared. In SG 7–SF comparison, the significant differences were at O (c) ( $P = 2.165 \times 10^{-5}$ ), O (m) ( $P = 2.165$

$\times 10^{-5}$ ), O (a) ( $P = 2.165 \times 10^{-5}$ ), and M (mm) ( $P = .002$ ). Similar findings were also observed at O (c) ( $P = 2.165 \times 10^{-5}$ ), O (m) ( $P = 4.333 \times 10^{-5}$ ), O (a) ( $P = 2.165 \times 10^{-5}$ ), and M (mm) ( $P = .003$ ) in SG 8–SF comparison. When SG 9 was compared with SF, the differences at O (c) ( $P = .004$ ) and O (m) ( $P = .004$ ) were statistically significant. Finally, when SGs 10, 11, and 12 were compared with SF, a significant difference was found only in SG 10–SF comparison at M (mm) ( $P = .002$ ) (Table 4).

### Comparison of the analog and inhibitor SGs of PGI<sub>2</sub>

When analog and inhibitor SGs of PGI<sub>2</sub> were compared, the differences at O (c) ( $P = 1.082 \times 10^{-5}$ ), O (m) ( $P = 1.082 \times 10^{-5}$ ), O (a) ( $P = 1.082 \times 10^{-5}$ ), and M (mm) ( $P = 1.082 \times 10^{-5}$ ) were found significant in the comparison of SGs 1 and 4. In SG 2–SG 5 comparison, the significant differences were again observed at all the parameters as  $P = 1.082 \times 10^{-5}$ ,  $P = 1.082 \times 10^{-5}$ ,  $P = 1.082 \times 10^{-5}$ ,

**TABLE 5.** Comparison of the Analog (1, 2, 3, 7, 8, 9) and Inhibitor (4, 5, 6, 10, 11, 12) Subgroups at the Same Concentrations

| Subgroups                                    | O (c) (psi) |                           | O (m) (psi) |                           | O (a) (psi) |                           | M (mm) |                           |
|--|-------------|---------------------------|-------------|---------------------------|-------------|---------------------------|--------|---------------------------|
|  | Z           | P                         | Z           | P                         | Z           | P                         | Z      | P                         |
| 1 (10 <sup>-4</sup> )–4 (10 <sup>-4</sup> )  | 3.860       | 1.082 × 10 <sup>-5*</sup> | 3.865       | 1.082 × 10 <sup>-5*</sup> | 3.865       | 1.082 × 10 <sup>-5*</sup> | 3.847  | 1.082 × 10 <sup>-5*</sup> |
| 2 (10 <sup>-5</sup> )–5 (10 <sup>-5</sup> )  | 3.838       | 1.082 × 10 <sup>-5*</sup> | 3.854       | 1.082 × 10 <sup>-5*</sup> | 3.842       | 1.082 × 10 <sup>-5*</sup> | 3.663  | 7.577 × 10 <sup>-5*</sup> |
| 3 (10 <sup>-6</sup> )–6 (10 <sup>-6</sup> )  | 3.847       | 1.082 × 10 <sup>-5*</sup> | 3.876       | 1.082 × 10 <sup>-5*</sup> | 3.847       | 1.082 × 10 <sup>-5*</sup> | 2.437  | .023                      |
| 7 (10 <sup>-4</sup> )–10 (10 <sup>-4</sup> ) | 3.833       | 1.082 × 10 <sup>-5*</sup> | 3.859       | 1.082 × 10 <sup>-5*</sup> | 3.841       | 1.082 × 10 <sup>-5*</sup> | 3.868  | 1.082 × 10 <sup>-5*</sup> |
| 8 (10 <sup>-5</sup> )–11 (10 <sup>-5</sup> ) | 3.827       | 1.082 × 10 <sup>-5*</sup> | 3.859       | 1.082 × 10 <sup>-5*</sup> | 3.845       | 1.082 × 10 <sup>-5*</sup> | 2.567  | .011                      |
| 9 (10 <sup>-6</sup> )–12 (10 <sup>-6</sup> ) | 2.321       | .029                      | 1.994       | .075                      | 3.258       | .001*                     | 0.856  | .436                      |
| 1 (10 <sup>-4</sup> )–7 (10 <sup>-4</sup> )  | 2.912       | .003*                     | 3.328       | 3.247 × 10 <sup>-4*</sup> | 2.786       | .005*                     | 0.622  | .579                      |
| 2 (10 <sup>-5</sup> )–8 (10 <sup>-5</sup> )  | 2.263       | .029                      | 2.835       | .004*                     | 2.120       | .043                      | 0.311  | .796                      |
| 3 (10 <sup>-6</sup> )–9 (10 <sup>-6</sup> )  | 3.454       | 3.247 × 10 <sup>-4*</sup> | 3.133       | .002*                     | 3.454       | 3.247 × 10 <sup>-4*</sup> | 1.011  | .393                      |
| 4 (10 <sup>-4</sup> )–10 (10 <sup>-4</sup> ) | 0.842       | .436                      | 0.691       | .579                      | 1.327       | .247                      | 0.247  | .853                      |
| 5 (10 <sup>-5</sup> )–11 (10 <sup>-5</sup> ) | 1.137       | .315                      | 0.210       | .853                      | 0.000       | 1.000                     | 2.459  | .019                      |
| 6 (10 <sup>-4</sup> )–12 (10 <sup>-6</sup> ) | 1.457       | .190                      | 1.699       | .143                      | 0.803       | .481                      | 2.202  | .035                      |

\* Significance level  $\alpha = .005$ .

and  $P = 7.577 \times 10^{-5}$ , respectively. The differences at O (c) ( $P = 1.082 \times 10^{-5}$ ), O (m) ( $P = 1.082 \times 10^{-5}$ ), and O (a) ( $P = 1.082 \times 10^{-5}$ ) were also significant when SGs 3 and 6 were compared.

#### Comparison of the analog and inhibitor SGs of TxA<sub>2</sub>

Statistically significant differences were found at O (c) ( $P = 1.082 \times 10^{-5}$ ), O (m) ( $P = 1.082 \times 10^{-5}$ ), O (a) ( $P = 1.082 \times 10^{-5}$ ), and M (mm) ( $P = 1.082 \times 10^{-5}$ ) in the comparison of SGs 7 and 10. The differences at O (c) ( $P = 1.082 \times 10^{-5}$ ), O (m) ( $P = 1.082 \times 10^{-5}$ ), and O (a) ( $P = 1.082 \times 10^{-5}$ ) were also significant in the SG 8–SG 11 comparison. The difference was significant only at O (a) ( $P = .001$ ) when SGs 9 and 12 were compared (Table 5).

#### Comparison of the analog SGs of PGI<sub>2</sub> and TxA<sub>2</sub>

The differences between SGs 1 and 7 were statistically significant at O (c) ( $P = .003$ ), O (m) ( $P = 3.247 \times 10^{-4}$ ), and O (a) ( $P = .005$ ). When SG 3 and 9 were compared, significant differences were again found at O (c) ( $P = 3.247 \times 10^{-4}$ ), O (m) ( $P = .002$ ), and O (a) ( $P = 3.247 \times 10^{-4}$ ). The difference was significant only at O (m) ( $P = .004$ ) between SGs 2 and 8 (Table 5).

#### Comparison of the inhibitor SGs of PGI<sub>2</sub> and TxA<sub>2</sub>

Statistically significant differences were not observed for any parameter when inhibitors were compared with each other.

### DISCUSSION

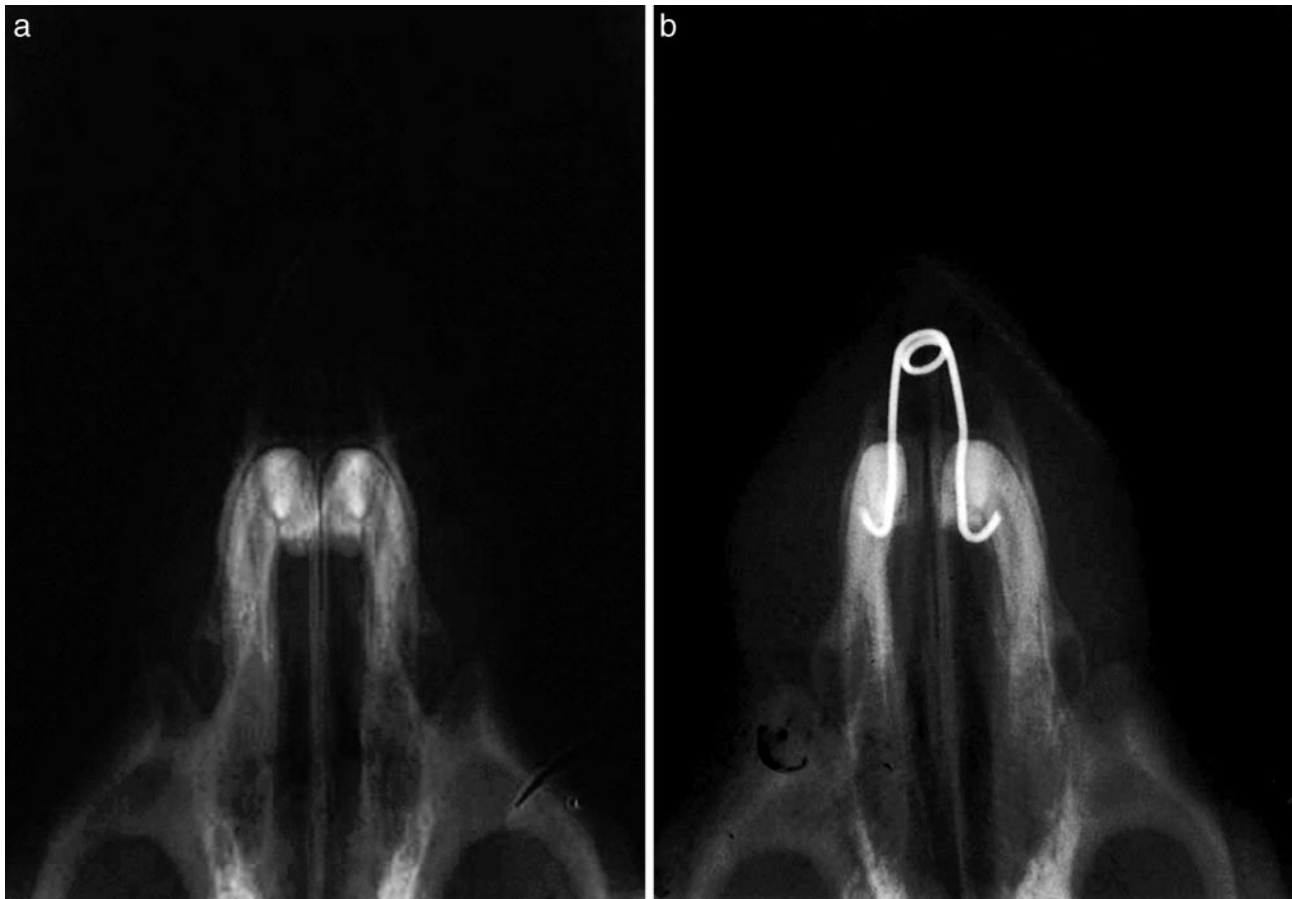
Local microenvironment is central to the regulation of osteoclastic activity, and studies show that different factors might influence the rate of orthodontic tooth movement by way of various biomediators.<sup>3,4,21,27</sup> This study compared the

effects of iloprost, indomethacin, U 46619, and imidazole on PGI<sub>2</sub> and TxA<sub>2</sub> synthesis in orthodontic tooth movement.

The experimental studies<sup>1,28–31</sup> related with the rats were reviewed, and five days of experimental period and 20 g of force were selected because 20 g was found to be the optimal force necessary for orthodontic separation without creating separation of the interpremaxillary suture or transfer of nonphysiologic forces to the teeth or supporting tissues in the rat. Because the short cycles in female rats cause hormonal variations, our study was carried out with male rats. Traumatic effects of the springs on the soft tissues were not observed during the experiment. No orthopedic separation was noted in the interpremaxillary suture when the pre- and posttreatment radiographs were compared (Figure 4). Although the statistical analyses were carried out on all the SGs, SG 15 was used only to observe the osteoclast number without any intervention. Because there was no statistically significant difference between SGs 13 and 14, SG 13 served as the major control SG.

The results indicated that multinuclear osteoclasts and orthodontic tooth movement were increased significantly in the analog SGs of PGI<sub>2</sub> and TxA<sub>2</sub>. These increases appeared to be dose dependent and were observed for all parameters at high concentrations (10<sup>-4</sup> and 10<sup>-5</sup>). This finding was similar to those of ElAttar and Lin,<sup>23</sup> Dewhirst et al,<sup>24</sup> and Rifkin and Tai,<sup>25</sup> who mentioned in their previous studies that PG and TxA<sub>2</sub> levels increased in inflammatory periodontal tissues. Our findings also matched with the findings of Saito et al,<sup>26</sup> who observed that TxA<sub>2</sub> analog administration increased the osteoclastlike cells in mouse marrow culture.

Linear measurements showed that the rate of orthodontic tooth movement was more in the iloprost (analog) SG, but the difference was not statistically significant between iloprost and U 46619 analog SGs. However, the number of osteoclasts was significantly greater in the iloprost group at the coronal, middle, and apical sections. In the light of this



**FIGURE 4.** Occlusal radiographs of the rat before (A), after (B) orthodontic tooth movement.

finding it may be concluded that iloprost, as an analog, or  $\text{PGI}_2$  synthesis is more effective in bone turnover.

It was demonstrated in previous studies<sup>3,5,7-12</sup> that PGs play an important role in bone turnover and PG administration enhances the rate of tooth movement. On the other hand, inhibition of PG synthesis significantly decreases the orthodontic tooth movement as Kehoe et al,<sup>11</sup> Mohammed et al,<sup>32</sup> Chumbley and Tuncay,<sup>33</sup> Giunta et al,<sup>34</sup> and Zhou et al<sup>35</sup> showed in their studies with indomethacin. Our findings are similar to these findings, and we also found that indomethacin and imidazole decrease the rate of tooth movement; however, the decrease was statistically significant only at high concentrations ( $10^{-4}$ ). This was related to the short experimental period of our study. Statistically significant differences were not observed between indomethacin and imidazole when inhibitory effects of these two materials were compared.

### CONCLUSIONS

Both iloprost and U 46619 significantly increased the number of multinuclear osteoclasts and the rate of orthodontic tooth movement in rats; however, iloprost administration increased the number of osteoclasts significantly

more than U 46619. Indomethacin and imidazole decreased the rate of tooth movement when they were injected at high concentrations, but a statistically significant difference was not observed between their inhibitory effects. Briefly, the increase in  $\text{PGI}_2$  and  $\text{TxA}_2$  levels, in periodontal tissues, enhanced the orthodontic tooth movement, whereas the decrease in these arachidonic acid metabolites reduced the rate of tooth movement.

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