The effect of acetaminophen, ibuprofen, and misoprostol on prostaglandin E_2 synthesis and the degree and rate of orthodontic tooth movement

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Cells of the nervous, immune, and endocrine systems become involved in the activation and response of the periodontal ligament (PDL) and alveolar bone cells during tooth movement.[1] The exact mechanism by which teeth move has not yet been determined. However, it has been postulated that movement may be mediated through the local production and action of prostaglandins (PGs).[2-4] The purpose of the study was to compare the effects of three common types of painrelievers on PGE_2 synthesis and orthodontic tooth movement.

Arachidonic acid, present in the membrane phospholipid of cells, can be released by phospholipidases activated by cellular damage or by any nondestructive pertubation of the membrane. Arachidonic acid is metabolized by two main enzyme pathways: cyclo-oxygenase and lipoxygenase. The products of arachidonic acid metabolism, namely PGs of the E and F series, prostacyclin (PGI_2) and the leukotrienes, are integral components of the inflammatory reaction. Inflammatory cells produce cytokines, which mediate various stages of inflammation. Some of these cytokines, namely interleukin 1∂ and 1β, have been implicated in the mediation of bone remodeling processes in vitro.[5,6] It is not clear whether the action of IL-1 on target cells involves stimulation of arachidonic acid metabolism.

Abstract

The present study compared the effect of acetaminophen, ibuprofen and misoprostol on PGE_2 synthesis and orthodontic tooth movement. Guinea pigs were randomly assigned into one of three test groups or a control group. Each group received study treatments every 12 hours as an orthodontic force was applied to the maxillary incisors. Direct linear measurements of tooth separation were recorded at days 2, 4, 6, 10, and 11, and inflammatory exudate from the periodontal ligament (PDL) space was extracted and quantitatively analyzed radioimmunologically for the presence of PGE_2 at days 4 and 9. Comparing the concentration of PGE_2 in sample extracts, a significant difference ($P=0.001$) was found among drug groups. A highly significant difference was found between the mean tooth separation among the various drug groups ($P<0.001$). At day 11 the misoprostol group exhibited 4.49 ± 0.49 mm of separation; ibuprofen 2.56 ± 0.11 mm, and the control and acetaminophen groups exhibited similar degrees of tooth separation: 3.31 ± 0.07 mm and 3.31 ± 0.08 mm, respectively. A highly significant difference occurred between the mean rates of tooth separation among the various drug groups after day 8 ($P<0.001$). Results of this study suggest that acetaminophen is the analgesic of choice for the relief of minor discomfort associated with orthodontic treatment.

Key Words

Prostaglandin E_2 • Tooth movement • Inflammation • Nonsteroidal anti-inflammatory drugs

Submitted: April 1995 **Revised and accepted:** August 1995 Angle Orthod 1996; 66(5):339-350.

Prostaglandins of the E and F series have been implicated in bone remodeling activities, particularly resorption.[7,8] Once formed, PGs of the E series are stable, but they are rapidly inactivated due to the enzyme action of 15-hydroxy prostaglandin dehydrogenase. Prostaglandins of the E series play an important role in the pathogenesis of chronic periodontitis by regulating production of osteoclast-activating factor in activated lymphocytes,[10] inducing an increase in size[11] or number[12] of osteoclasts and accelerating release of lysosomal enzymes and collagenase from activated macrophages.[13] Researchers have postulated that prostaglandin E_2 is a mediator of bone resorption in periodontal disease,[14-17] trauma,[18] and malignancies.[19-22] Local PGE_2 levels in the gingival tissues have been shown to correlate with periodontal tissue destruction.[23,24] PGE_2 has been shown to stimulate bone resorption,[3,8,19,25-28] decrease collagen synthesis,[8,29,30] and cause increases in cyclic AMP.[31-35] It is possible, however, that the ability of PGE_2 to increase cellular cyclic AMP may be related less to its resorptive effects than to its inhibitory effect on new bone formation.[30] Prostaglandin $F_{2\partial}$ ($PGF_{2\partial}$) is found in mineralized tissues,[36] where it may be associated with bone apposition and resorption.[37]

Prostaglandins and tooth movement

Yamasaki et al.[3] reported the role of PGs as biochemical mediators of bone resorption induced by tooth movement in rats. Local injection of PGE_1 or PGE_2 resulted in a dose-dependant increase in the appearance of osteoclasts at the site of tooth movement, while administration of indomethacin, a specific inhibitor of prostaglandin synthesis,[38] was found to have an inhibitory effect on the appearance of osteoclasts. Chumbley[39] supported the histologic data by showing that indomethacin reduced the rate of orthodontic tooth movement in cats by half. Chao et al.[40] showed that in rats treated with daily submucosal injections of 50μg of PGE_2 per kilogram for 5 consecutive days, alveolar bone resorption was activated during tooth movement. Sandy and Harris[41] found a decrease in osteoclasts and tooth movement in rabbits where PG synthesis was inhibited by flurbiprofen. Mohammed et al.[42] found significant inhibition of tooth movement in rats when indomethacin was administered.

In monkeys, Yamasaki et al.[43] showed that administration of PGE_1 or PGE_2 in gingiva near orthodontically treated canines nearly doubled the rate of canine retraction as compared with controls. In a clinical study[44] that followed, PGE_1 was administered locally near ortho-dontically treated teeth and movement was enhanced. However, patients experienced significant pain during the injection because PGs biochemically mediate the amount of cyclic AMP, which modulates norepinephrine at the neural synapse.[45] In rats, Lee[46] administered gingival injections of PGE_1 (5μg/kg) twice daily, or a constant systemic administration (7.5 ng/kg/min) by an osmotic pump. In both cases the number of osteoclastic lacunae in periodontal pressure sites increased, compared with non-PGE_1 treated animals. Cementum and dentin remodeling in specimens with local application of PGE_2 combined with orthodontic tooth movement has been found not to differ significantly from groups in which only orthodontic force has been applied.[47] This finding raises the possibility of administering PGE_1 or PGE_2 systemically during orthodontic treatment in an effort to enhance the rate of tooth movement.

Orthodontic forces

Smith and Storey,[48] Storey and Smith,[49] and Storey[50] found significant differences in the rate of tooth movement and the nature of associated tissue changes when various forces were applied to teeth. With heavy force (150 grams), tissue disruption predominates, while light force (25 grams) induces orderly tissue disruption and remodeling of bone and connective tissues in the guinea pig.[50]

Utley[51] found no significant difference in the rate of canine movement in the cat with forces ranging from 40 to 560 grams. Fortin[52] and Furstman et al.,[53] in the monkey and dog, respectively, found significant differences in the rate of tooth movement between heavy and light forces. Botting and Storey[54] demonstrated that with different forces there are significant differences in the rate and nature of tissue changes associated with tooth movement in the guinea pig. Generally, it is the magnitude of the force that will determine the duration of hyalinization. This will be shorter within the light force level, although there is a tendency for longer initial hyalinization periods and formation of secondary hyalinized zones when excessively strong forces are applied.[55] Increasing the magnitude of the force does not increase the rate of tooth movement. The purpose of applying a light force is to increase cellular activity without causing undue tissue compression and to prepare the tissues for further changes. In each species there is an optimal range of force necessary to induce a clinically acceptable rate of tooth movement. Various forces to induce orthodontic tooth movement have been reported in the literature.[39,41,42,56]

Drugs and prostaglandin synthesis

Nonsteroidal anti-inflammatory drugs exhibit analgesic, antipyretic, and anti-inflammatory actions as a result of the inhibition of PG biosynthesis from arachidonic acid. Aspirin-like drugs have been shown to inhibit PG release from human platelets by Smith and Willis,[57] from the perfused dog spleen by Ferriera et al.,[58] and PG synthesis in cell-free homogenates of guinea pig lung by Vane.[38] Aspirin and other nonsteroidal anti-inflammatory drugs (NSAIDs) are potent inhibitors of cyclo-oxygenase,[38] an enzyme involved in the catabolism of arachidonic acid. In the guinea pig, oral administration of aspirin was found to effectively inhibit PG synthesis at the level of the bronchioles, but did not appear to significantly affect tooth movement.[56] However, the force used in this study may not have provoked significant PG synthesis in the tissues surrounding the teeth.

Ibuprofen, a NSAID, is available as a prescription drug or in several nonprescription products. The recommended dosage of the nonprescription product is 200 mg every 4 to 6 hours while symptoms persist. Although 400 mg may be used, no more than 1.2 gms should be taken in a 24-hour period.

Inhibition of alveolar bone loss has been shown to occur in dogs treated daily with 4 mg/kg of ibuprofen over a 13-month period.[59] Given systemically, ibuprofen has been shown to consistently inhibit localized osteoclastic bone resorption[60] and synthesis of PGs in the middle ear effusions of chinchillas in which otitis media had been induced.[61]

Acetaminophen has both analgesic and antipyretic properties, but produces no antirheumatic or anti-inflammatory activity. It has been shown to either inhibit or stimulate PG synthesis, depending on the tissue, preparation of the tissue, and constituents of the incubation milieu. Acetaminophen may fail to exhibit anti-inflammatory activity because it does not concentrate in areas of inflammation.[62] Vane[38] found acetaminophen to have a weak inhibitory effect on PG synthesis in the guinea pig lung, strong antipyretic and analgesic actions, and lack of anti-inflammatory activity. It seems that antipyretics, which lack an anti-inflammatory action, inhibit PG synthetase only in the central nervous system and not in peripheral tissues; this is supported by the fact that acetaminophen was inactive as an anti-inflammatory agent in the rat paw edema test.[63] A popular nonprescription form of acetaminophen is available in 325 mg tablets (Tylenol; McNeil Pharmaceuticals, Fort Washington, Penn). The recommended adult dose is 325 to 650 mg every 4 hours, not to exceed 4 grams daily. The analgesic oral dose ED_{50} in the adjuvant arthritic rat was found to be over 200 mg/kg.[64]

Misoprostol (Cytotec; G.D. Searle and Co, Skokie, Ill), a synthetic PGE_1 analog, is available only as a prescription product in 100 μg or 200 μg tablets. Nonsteroidal anti-inflammatory drugs inhibit PG synthesis within the gastric mucosa, which may contribute to mucosal damage. Misoprostol can increase secretion of duodenal bicarbonate and gastric mucus production,[65] and is the theoretical basis for prescribing misoprostol when a NSAID is used long-term. Misoprostol in a mouse model provides analgesia in a dose-related manner equivalent to that of morphine—the ED_{50} being on the order of 2 mg/kg (~10^{-5} M). Misoprostol has the capacity to work synergistically with recognized NSAIDs such that the usual level of analgesia can be achieved with considerably lower doses.

Hypothetically, because acetaminophen is inactive as an anti-inflammatory agent in peripheral tissues,[63] it should have no adverse effect on PG biosynthesis and subsequent bone resorption associated with orthodontic tooth movement, unlike the NSAID ibuprofen. Misoprostol should enhance tooth movement. Previous in vivo studies have demonstrated that administration of PGs enhances bone resorption associated with orthodontically treated teeth.[3,40,43,44,46] The duration of this study was selected somewhat arbitrarily: Patients treated in the orthodontic department reported discomfort associated with their teeth and supporting periodontium that they felt warranted the use of an OTC analgesic for 2 to 5 days. However, patients have reported discomfort for up to 10 days following their appointments. Patients also reported predosing with an OTC analgesic prior to appointments.

Materials and methods

Experimental model and design

Male guinea pigs (Hartley strain) ranging in weight from 290 to 380 grams were obtained from ACE Laboratories (Boyer Town, Penn). Maturation was determined by initial body weight to be 6 to 8 weeks of age. Animals were housed separately at the central animal facility of Temple University School of Medicine. The environment was maintained at a constant temperature (72° F), they received overhead lighting 12 hours per day, and were fed pellets and water ad libitum.

Guinea pigs were chosen primarily because

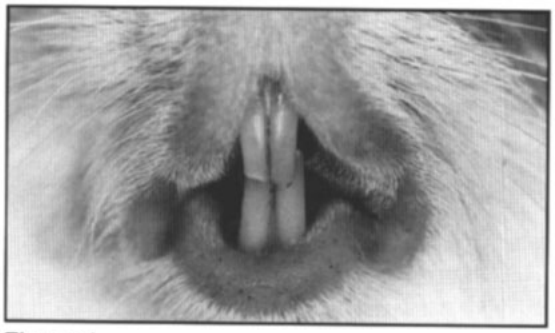

Figure 1

Figure 2

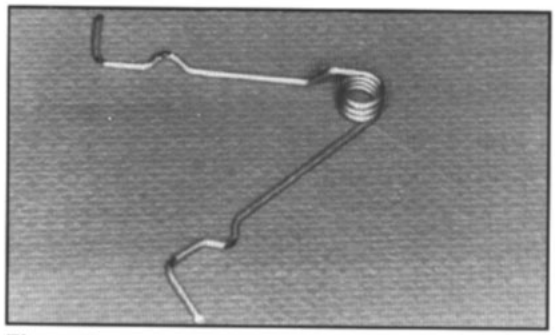

Figure 3

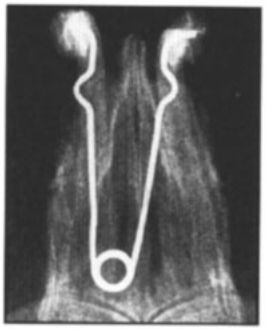

Figure 4

Figure 1
Natural dentition in the adult guinea pig.

Figure 2
Radiograph of the premaxillary suture in the adult guinea pig.

Figure 3
TMA spring (.016')-side view

Figure 4
Radiograph of premaxillary suture following incisor separation

they have periodontal structures and incisors that can be successfully manipulated by orthodontic mechanotherapy (Figure 1). Although these teeth erupt continuously, the arrangement of the hard and soft tissues is similar to that in man. The interpremaxillary suture in a guinea pig is fused at 6 weeks of age;[50] thus, separation of the maxillary incisors occurs primarily by orthodontic tooth movement without the added complication of orthopedic separation between the two halves of the premaxilla (Figure 2). Males were used to eliminate any hormonal variability due to the female reproductive cycle.[66]

Drug regimen

Forty (n=40) guinea pigs were randomly assigned to one of three test groups or a control group, each consisting of 10 animals. The control group received 0.4% carboxymethylcellulose (CMC) stock solution, a suspending agent used in the pharmacologic preparations for test groups 3 and 4.

Group 1: Control-0.4% carboxymethylcellulose administration 1.66 ml/kg q 12 h (volume effect).

Group 2: Misoprostol administration 100 μg/kg q 12 h.

Group 3: Acetaminophen administration 200 mg/kg q 12 h.

Group 4: Ibuprofen administration 30 mg/kg q 12 h.

Drugs were administered 1 hour before placement of the orthodontic appliances. NSAIDs given 1 hour prior to the initiation of orthodontic tooth separation were shown most effective in suppressing the appearance of osteoclasts.[3] Animals were weighed (OHaus Autogram 1000; Florham Park, NJ) each morning and dosages adjusted accordingly. Selected dosages were to approximate the level that creates analgesia, without adverse effects on behavior or general health. The animals did not appear to experience pain during the experiment: Their eating habits were not altered and their weight continually increased after day 3. All groups received study treatments by gastric lavage every 12 hours using a 10 cc disposable syringe (Becton Dickinson and Co, Rutherford, NJ) and stainless steel biomedical needle (Perfectum size 13; Popper and Son Inc, New Hyde Park, NY) over a period of 11 days.

Three trials were conducted. The first trial consisted of a sample population of 8 (2 animals per test group). The second and third trials consisted of sample populations of 16 each (4 animals per test group).

Drug preparations

Suspensions were prepared at the beginning of each trial and were thoroughly mixed before administration. Misoprostol stock solution (60 μg/ml) was prepared by dissolving the compound (misoprostol:HPMC dispersion [1:100]; G.D. Searle Co, Skokie, Ill) in distilled water. Stock solution was stored at 2 - 8° C. Acetaminophen and ibuprofen suspensions were prepared with 0.4% carboxymethylcellulose. Acetaminophen stock solution (120 mg/ml) was prepared with acetaminophen powder, USP (Miles Laboratories Inc, Elkhart, Ind) and stored at 2 - 8° C. Ibuprofen suspension (Children's Motrin; McNeil Pharmaceutical, Fort Washington, Penn) 100 mg/5 ml stock solution was purchased from McNeil Pharmaceutical and stored at room temperature.

Orthodontic appliance

Appliances consisting of a single 0.016" titanium molybdenum alloy wire (TMA, Ormco Corp; Glendora, Calif) formed into a helical torsion spring with a coil, 4 turns, 2 mm in diameter, and arms 12 mm in length (Figure 3) were directly bonded to the labial surface of the maxillary incisors. A vertical step was placed in one arm 1 mm anterior to the coil, thereby allowing both arms to lie parallel. Horizontal V-bends were placed in the arms 12 mm anterior to the coil to prevent labial displacement of the appliance. Horizontal bends 90° outward and 3 mm anterior to the V-bends served as a means for attaching the appliance to the labial surfaces of the incisors.

Technique for positioning appliances

Animals were anesthetized with 2 mg/kg of Acepromazine (Ayerst Laboratories, Inc, New York, NY) and 60 mg/kg of Ketalar (Parke-Davis, Morris Plains, NJ) i.p. A 0.5 mm undercut was placed between the maxillary incisors at the gingival papilla with the aid of an electric handpiece (Dremel Moto-Tool, Model 285; Racine, Wisc) and a #1 round carbide bur (Premier; Norristown, Penn). The undercut served to stabilize the appliance during the bonding procedure and to resist occlusogingival displacement. The mandibular incisal edges were reduced using the Dremel Moto-Tool and a #700-010 steel crosscut fissure bur (Brasseler USA, Savannah, Ga) to prevent occlusal interference with the appliance.

Labial surfaces of the maxillary incisors were debrided and dried with moisture-free air. Following a 60-second etch with 40% phosphoric acid gel (Onyx L/G Black Etch; Class I Orthodontics, Lubbock, Tex), the enamel surface was rinsed with water for 20 seconds and dried until the surface acquired a chalky appearance. Primer (Transbond; Unitek/3M, Monrovia, Calif) was applied to the etched enamel surface and light cured for 10 seconds (Ortholux Light Unit #712-019; Unitek/3M, Monrovia, Calif). The helical end of the spring lay passively against the palate. Each arm was inserted through the interproximal contact until engaged in the interproximal undercut, then retained in position with light cured adhesive (Transbond; Unitek/3M, Monrovia, Calif). Several layers of adhesive were placed to compensate for the adhesive wear brought about by the animals' continuous gnawing behavior.

Measurement of force

Initial force exerted by each spring was determined, prior to insertion, with a dial gauge (Accu-Force III digital force gauge; Ametek, Largo, Fla) accurate to 0.1 grams. With both arms touching, the springs were capable of exerting a reciprocal lateral force of 25 ± 0.1 grams. When passive, the arms were 45° to each other and 12 mm apart at their ends. One investigator fabricated all the springs. Titanium molybdenum alloy possessing half the force and twice the working range of stainless steel was capable of exerting a light and continuous expansile force. The spring was placed interproximally, so that tension occurred in the PDL on the mesial and compression on the distal side of the treated incisors.

Recording tooth separation

Prior to placing the appliances, no measurable space existed between the maxillary incisors. Measures of separation were made by two observers blinded to treatment allocation. Intraoral measurements were made using three direct linear measures, which were then averaged. Measurements were recorded at the interproximal undercuts using a digital measuring caliper (Mitutoyo Corp, Japan) accurate to .01 mm. Measurements were recorded at days 2, 4, 6, 8, 10, and 11.

Radioimmunoassay technique

A radioimmunoassay technique was used to quantitatively analyze samples of inflammatory exudate from the PDL space for the presence of PGE_2. The prostaglandin E_2 [^{125}I] Radioimmunoassay Kit (Catalog #NEK-020A, Dupont New England Nuclear; Boston, Mass) is based on the use of an iodinated analog of prostaglandin E_2 (PGE_2 [^{125}I]) as tracer and lyophilized rabbit anti-PGE_2 as the antiserum (specific antibody). The basic principle of this radioimmunoassay technique is competitive binding, where a radioactive antigen competes with a nonradioactive antigen for a fixed number of antibody binding sites. When unlabeled antigen from standards or samples and a fixed amount of tracer (labeled antigen) are allowed to react with a constant and limiting amount of antibody, decreasing amounts of tracer are bound to the antibody as the amount of unlabeled antigen is increased. Separation of the antibody-antigen complexes from free antigen is achieved by precipitation of the antibody-bound tracer with polyethylene glycol in the presence of carrier immunoglobin. After centrifugation, the supernatant containing the unbound antigen is decanted, and the pellet containing the antibody-antigen complex is quantified using a gamma counter. Results obtained for the standards are used to construct a standard dose-response curve from which the unknowns are read by interpolation. The sensitivity of the assay kit has been determined by the manufacturer to be approximately 0.13 picograms of PGE_2 added. From our pretrial study conducted with 8 animals, we found that baseline readings of local PGE_2 concentrations in the PDL space could not be determined due to the lack of ample inflammatory exudate that could be aspirated and successfully analyzed with this radioimmunologic procedure.

Animals were anesthetized with 2 mg/kg of Acepromazine and 60 mg/kg of Ketalar. An aspirating syringe (Hamilton 10 µl syringe, #701-1LT; Reno, Nev) was used to collect

Table 1
Comparison of mean separation (millimeters) according to trial

Day	Trial 1 (n=8) Mean ± S.D.	Trial 2 (n=16) Mean ± S.D	Trial 3 (n=16) Mean ± S.D	F Statistic	P
2	1.45 ± 0.23	1.38 ± 0.12	1.46 ± 0.16	3.052	0.064
4	1.86 ± 0.34	1.75 ± 0.19	1.92 ± 0.26	2.562	0.096
6	2.32 ± 0.39	2.11 ± 0.28	2.31 ± 0.29	2.505	0.100
8	2.71 ± 0.54	2.70 ± 0.43	2.74 ± 0.47	0.503	0.611
10	3.13 ± 0.68	3.06 ± 0.56	3.21 ± 0.73	1.227	0.309
11	3.42 ± 0.73	3.35 ± 0.68	3.49 ± 0.84	0.338	0.716

Table 3
Mean concentration of PGE_2 from sample extracts at days 4 and 9

Drug group	*Mean±SD
1 (CMC)	6.68±2.30
2 (Misoprostol)	4.99±2.54
3 (Acetaminophen)	1.62±2.12
4 (Ibuprofen)	0.32±0.55

*Mean concentrations reported in picograms (pg)

Table 2
Comparison of mean weight (grams) according to drug group

Day	Group 1 Control (CMC) Mean ± S.D.	Group 2 (Misoprostol) Mean ± S.D.	Group 3 (Acetaminophen) Mean ± S.D.	Group 4 (Ibuprofen) Mean ± S.D.	F Statistic	P
1	333.40 ± 17.41	337.70 ± 23.80	337.20 ± 21.57	341.90 ± 19.73	0.554	0.650
2	324.80 ± 18.60	323.40 ± 21.63	326.00 ± 24.41	321.70 ± 26.03	0.113	0.952
3	338.90 ± 15.09	331.70 ± 21.65	342.30 ± 20.95	323.90 ± 36.49	1.764	0.177
4	344.00 ± 19.32	338.70 ± 27.44	352.80 ± 21.65	327.30 ± 43.23	2.564	0.075
5	350.60 ± 20.25	343.80 ± 29.49	357.30 ± 21.55	329.80 ± 44.92	2.444	0.085
6	370.00 ± 15.29	361.20 ± 25.53	378.50 ± 18.69	351.70 ± 37.55	2.180	0.113
7	379.00 ± 14.19	368.90 ± 23.25	387.40 ± 16.36	360.90 ± 34.42	2.282	0.101
8	387.30 ± 19.78	372.40 ± 26.79	391.80 ± 20.76	370.50 ± 38.36	1.851	0.161
9	390.40 ± 25.02	370.90 ± 36.47	392.20 ± 28.13	371.90 ± 38.30	2.008	0.136
10	388.80 ± 22.00	377.30 ±32.46	393.80 ± 33.18	368.30 ± 35.76	2.407	0.088
11	399.40 ± 23.11	384.50 ± 31.96	407.60 ± 34.01	378.20 ± 34.58	3.155	0.040

inflammatory exudate samples in duplicate from the PDL compression site on the mesial of the left maxillary incisor at days 4 and 9. The needle of the syringe was inserted to the depth of the periodontal ligament space, and by gently pulling back on the syringe plunger, negative pressure was created, thus allowing aspiration of 0.2 ml of inflammatory exudate. Samples were immediately assayed quantitatively for the presence of PGE_2.

Upon completion of the data collection at day 11, each specimen's maxilla was radiographed to verify the continuity of the interpremaxillary suture and separation of the incisors (Figure 4). The degree of tooth separation was recorded photographically at 1:1 magnification with a Nikon N2000 camera (Lester A. Dine Inc, Palm Beach Gardens, Fla) on Kodak Plus-X pan 125 film (Eastman Kodak Co, Rochester, NY). Appliances were retrieved with an adhesive-removing instrument (AEZ Model #803-00210; Ormco Corp, Glendora, Calif). The animals were not followed after retrieval of the appliances. Force levels generated by the appliances were verified with the dial gauge. The springs exhibited superior memory and lack of distortion of the helix or arms.

Statistical analysis

Analysis of variance (ANOVA) was performed to determine whether or not statistically significant differences in weight existed between trials. The mean change in tooth separation as measured by direct linear measurement was calculated. Two-tailed paired *t*-tests were used to determine whether or not significant differences occurred between measurement points, as well as to determine whether or not significant differences occurred between the mean weight change within drug groups and trials. Analysis of covariance controlling for differences in weight at baseline was performed to determine if there were significant differences between the

degree and rate of maxillary incisor separation, and comparative rate of change among drug groups. Pearson correlation coefficients were performed to determine if a correlation existed between the concentration of PGE_2 in sample extracts and the degree of tooth separation in the various drug groups. Both 1- and 2-tailed criteria for significance were applied.

Results

Changes in weight

A statistically significant difference ($P < 0.001$) was found in the baseline weights (day 1) of animals between trials. Differences in baseline weights may be attributed to the animals in trial 2 being 6 to 7 weeks old rather than 8. Weight differences did not affect tooth separation, and analyses of covariance revealed no significant difference between the mean separation among trials from days 2 to 11 ($P > .05$) (Table 1). No significant difference in mean body weight was found between the various drug groups at the onset of the experiment ($P = 0.650$) (Table 2). Postanesthesia weight loss occurred in all groups at day 2. By the third day, the mean weight for the control and acetaminophen groups surpassed initial weights at the onset of the experiment and between the fourth and sixth day for the misoprostol and ibuprofen groups. Within drug groups, a significant difference ($P < 0.05$) in mean weight change occurred between days 1 and 2 due to the transient loss in weight following anesthesia and appliance placement. A significant difference in mean weight change occurred after days 4 or 6 due to steady increases in body weight ($P \leq 0.005$).

Effect of drugs on prostaglandin E_2 concentration

Quantitative evaluation of PGE_2 from PDL inflammatory extracts of the guinea pig revealed a highly significant difference when comparing the mean concentrations of PGE_2 among the four drug groups ($P = 0.001$). Compared with group 1, group 2 showed a slight decrease, group 3 a fourfold decrease, and group 4 nearly a twenty-onefold decrease in the mean concentration of PGE_2 produced in the PDL space (Table 3).

Effect of drugs on orthodontic tooth movement

Analysis of covariance revealed a highly significant difference between the mean tooth separation among the groups tested ($P < 0.001$) (Figure 5). At day 11, group 2 exhibited the greatest degree of tooth separation (4.49±0.49 mm) (Figure 6), group 4 the smallest (2.56±0.11 mm), and groups 1 and 3 exhibited a similar degree of tooth

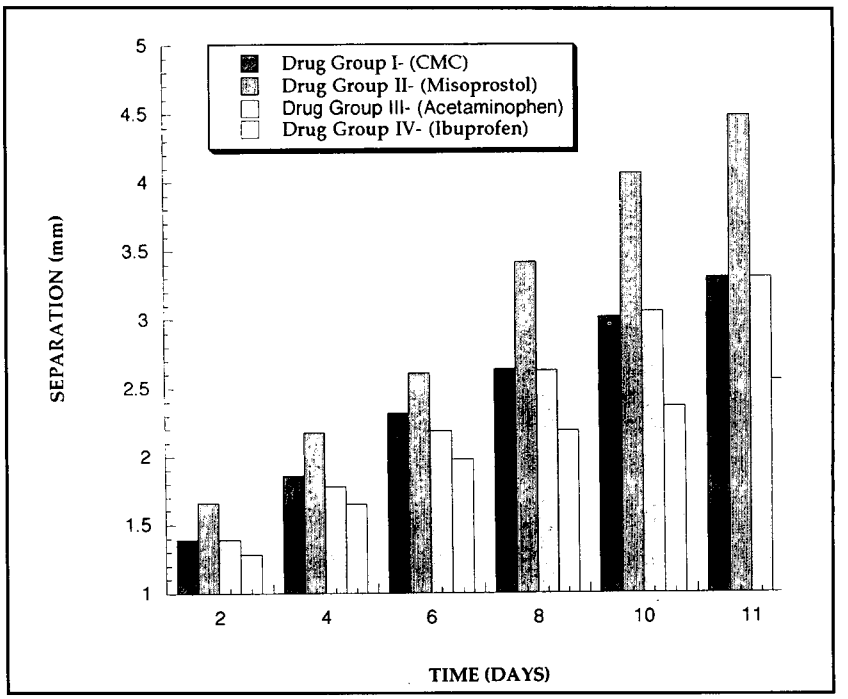

Figure 5

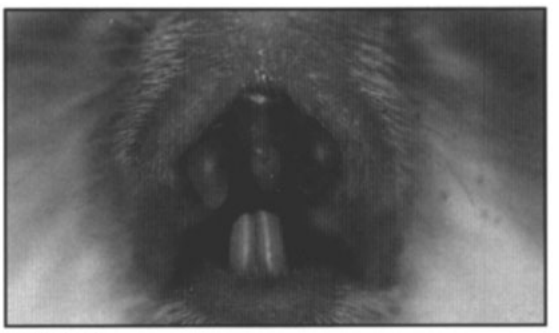

Figure 6

Figure 5
Comparison of mean separation (mm) according to drug group

Figure 6
Incisor separation in group 2 (misoprostol)-day 11

Figure 7
Comparison of separation (mm) according to drug group

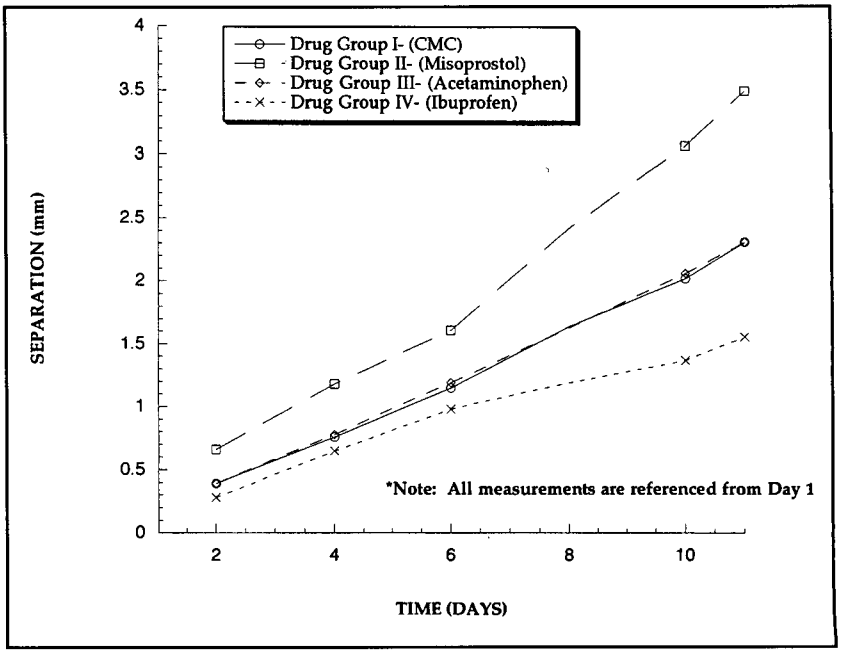

Figure 7

separation (3.31±0.07 mm and 3.31±0.08 mm, respectively). Analysis of covariance revealed a highly significant difference between the mean rates of tooth separation among the various drug groups after day 8, which may be attributed to enhanced osteoclastic activity. Analysis of covariance revealed a highly significant difference between the comparative rates of change in separation among drug groups after day 2 ($P \leq 0.001$) (Figure 7).

Discussion

The present study compared the effect of three test agents on the degree and rate of orthodontic tooth movement in guinea pigs. Acetaminophen and ibuprofen were selected for comparison because they are the most widely recommended over-the-counter analgesics for the relief of minor dental discomfort. Misoprostol was selected to establish whether an orally administered PGE_1 analog would enhance orthodontic tooth movement. Previous in vivo studies[3,39,41,42] have shown NSAIDs to have an inhibitory effect on orthodontic tooth movement. However, no biochemical assays have been performed to determine their effect upon local concentrations of PGs.

In our study, group 1 received no active pharmacological agent, so it may be assumed that the mean concentration of PGE_2 released locally within the PDL was a result of the orthodontic force. (Table 3) The decrease seen in group 2 may be attributed to lower body weight with a resultant decrease in the production of inflammatory exudate. The decrease seen in group 3 may be due to local peripheral anti-inflammatory activity of acetaminophen in the guinea pig model. The dramatic decrease seen in group 4 may be attributed to the anti-inflammatory activity of this NSAID. Groups 1 and 3 exhibited a weak positive correlation between the degree of tooth separation and PGE_2 concentrations, 0.387 and 0.053, respectively. Group 2 exhibited a strong positive correlation, 0.787, while group 4 exhibited a weak, negative correlation, -0.505. This weak negative correlation indicates that as levels of this NSAID increased, the PGE_2 concentration in the PDL decreased.

Tooth movement can be enhanced with local injections of PGE_1 or PGE_2.[3,40,43,44] However, local injection of PGs elicits significant pain.[47] Since most studies that associated PGs with tooth movement were performed prior to the development of the orally administered PGE analog misoprostol, there remains some doubt as to its efficacy and potency in laboratory animals. Since ibuprofen and acetaminophen have not been studied as extensively as other analgesics, there remains doubt as to their efficacy in producing analgesia in laboratory guinea pigs. Due to the multiple doses mandated by our study, three factors in the dosing regimen needed to be considered: drug dosage, route of administration and frequency of drug administration.

Since misoprostol has a tendency to cause diarrhea, a lower systemic dose than that which causes analgesia in the mouse was considered. At a dosage of 100 μg/kg, a significant increase in the degree and rate of tooth movement was noted with no symptoms of diarrhea. Administration by gastric lavage allowed systemic administration of a PG and made painful local injections unnecessary.

Ibuprofen has adverse effects on the G.I. system when administered in high therapeutic doses. A dosage of 30 mg/kg was found to result in a significant decrease in the degree and rate of tooth movement without any adverse effects on the animal's behavior or general health. Animals appeared not to experience pain since their eating habits were not affected and their body weight continued to increase during the course of the experiment.

A dosage of 200 mg/kg of acetaminophen was selected because the analgesic oral dose ED_{50} in the rat has been determined to be approximately 200 mg/kg.[64] This dosage was found to have no adverse effect on tooth movement or general health.

Tooth movement

Tooth movement rates can vary, even when a relatively constant force is applied to a tooth. During the first day, rapid tooth movement oc-

curs, resulting from displacement of soft tissue so that the tooth within its bony socket comes into contact with the bone on the pressure side. The rate of tooth movement then decreases as the bone on this side is resorbed. In the rabbit and rat (but not the mature guinea pig) a contributing factor to the tooth movement is the lateral movement of the premaxillary bones. A force of 25 grams was selected because this 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 periodontal tissues in the guinea pig.[8] The appliances generated 25 ± 0.1 grams of force when the arms were fully compressed and were passive when the arms were 12 mm apart; at least 15 ± 0.1 grams of force was exerted between the incisors at all times. No orthopedic separation was noted in the interpremaxillary suture when pre- and posttreatment radiographs were compared.

Summary and conclusions

This study demonstrates that ibuprofen significantly inhibits PGE_2 production in the PDL of the guinea pig. Associated with this decrease in local PGE_2 production, we observed a marked decrease in the degree and rate of orthodontic tooth movement. Acetaminophen had an inhibitory effect on peripheral PG production at the level of the PDL; however, the degree or rate of tooth movement was not significantly different than in the control. Misoprostol had an insignificant inhibitory effect on local PGE_2 production; however, the degree and rate of tooth movement was enhanced in comparison with the other test groups. This acceleration of tooth movement may be attributed to the enhanced bone resorbing activity of PGE_1, which supports the histological data of Yamasaki[3,67] and Lee.[46]

By recommending an OTC analgesic which exhibits minimal adverse effects on PG biosynthesis, clinicians may reduce treatment time. With a reduction in treatment time, patients may be receptive to comprehensive orthodontic treatment. Associated with a shorter treatment time is the reduced risk of adverse sequelae associated with long-term orthodontic therapy.

A long-term investigation comparing the relationship between OTC analgesics, local PG synthesis, and orthodontic tooth movement is recommended. Histological studies are needed to examine the effects of various synthetic PGs on the cementum of root surfaces and supporting alveolar bone. The issue of administering an oral PG to facilitate orthodontic treatment also requires further clinical study.

Acknowledgments

This project has been supported by the Temple University Orthodontic Alumni Association and a grant awarded by the American Association of Orthodontists Foundation. Sincere gratitude is expressed to Edward H. Hamilton Jr., DDS, former chairman, Department of Orthodontics; and Lee Liu-Chen, PhD, professor of pharmacology, for their interest and counsel in the conduction of this research investigation. Special thanks are extended to Jean Morrison of Unitek/3M; Ruthann Butterfield of Ormco Corporation; and Searle Research and Development for their generosity.

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