
Comparative evaluation of pulpal blood flow during incisor intrusion

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Aim: The aim of the present study was to evaluate and compare changes in pulpal blood flow (PBF) as a result of maxillary incisor intrusion achieved by one of two methods (utility arches or mini-implants).

Materials and methods: Thirty subjects were divided into three groups, the first of which underwent maxillary incisor intrusion using utility arches (UA) and a second group, intrusion via mini-implants (MI). The third group acted as a control. An intrusive force of 100 g was applied to the upper incisors in the treatment groups, whereas no force was applied to the anterior teeth in the control group. A laser Doppler flowmeter (LDF) was used to measure PBF at baseline (T0) and during incisor intrusion at 24 hours (T1), three days (T2), seven days (T3) and three weeks (T4). Statistical changes in PBF were assessed by the Wilcoxon Signed Rank and Mann-Whitney U tests, with significance set at $p < 0.05$.

Results: The mean PBF in the UA and MI groups decreased significantly from T0 to T1 ($p < 0.001$), slightly increased at T2 and continued to increase gradually at T3. PBF attained levels similar to those measured prior to intrusion at T4. No significant changes in PBF were observed in the control group over the course of the study. The only statistically significant difference between the UA and MI groups were at T1 and T2, at which time the MI group had lower PBF values ($p < 0.001$).

Conclusions: Despite slight regressive changes in pulpal tissue observed over the short term, PBF values tended to return to initial levels within three weeks, indicating that changes observed in PBF with the UA and MI intrusion methods are reversible. Although the changes in PBF could not be directly related to the method of intrusion employed, in general, a more severe drop in PBF was observed in the MI group during the first three days of intrusion.

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Introduction

One of the most challenging problems encountered in clinical dentistry is the correction of a deep bite. This is often accomplished through the intrusion of anterior teeth using a conventional intrusion arch,¹ a three-piece intrusion arch,² or a utility arch.³ The intrusive force applied by utility arches to the incisor brackets is usually anchored to the maxillary molars,³ which are subjected to an extrusive force and a tip-back moment.^{3,4} Although utility arches can successfully correct a deep bite by obtaining incisor intrusion, their treatment efficiency is limited by undesirable side effects, such as incisor proclination and unwanted distal tipping of posterior anchor molars.⁴ Headgear, elastics, adjacent teeth and various appliances have been suggested as anchorage support; however, most

methods require a high level of patient compliance to be successful.⁵ Recently, mini-implants have been used as anchorage devices to successfully intrude maxillary incisors with few side effects.^{5,6} Mini-implants offer several advantages, including their small size, low cost and easy insertion and removal.⁷ Moreover, the intrusion of maxillary incisors can be achieved without adverse effects on vertical molar position⁶ and treatment efficiency is minimally dependent upon patient cooperation.

Most research comparing alternative methods of overbite correction has focused on the amount and duration of intrusion, skeletal variables, incisor axial inclination and molar movement.^{5,8,9} However, a literature review uncovered no studies that compared the effect of different intrusion methods on pulpal

blood flow (PBF). Previous studies measuring changes in PBF have relied on histological observation,¹⁰ pulp-tissue respiration rates,¹¹ direct microscopic observation¹² and fluorescent microsphere injection.^{13,14} These methods involve technical limitations that allow teeth to be observed once and only after extraction. In contrast, laser Doppler flowmetry (LDF) is a non-invasive method that can repeatedly measure PBF of the same tooth without causing pulpal damage.^{15,16} As a non-invasive, objective, painless and semi-quantitative method, LDF has been used successfully to investigate vascular changes associated with orthodontic tooth movement.¹⁷⁻²⁰ Currently, one published study has evaluated PBF values in human teeth after the application of intrusive forces with conventional intrusion methods,¹⁷ and only one study has evaluated PBF of maxillary incisors with a mini-implant system.¹⁸ Given the scarcity of LDF/PBF data related to intrusion methods, the present study aimed to comparatively evaluate the PBF generated during anterior maxillary intrusion using either utility arches or mini-implants.

Materials and methods

Subjects and intrusive force application

The present study was approved by the Institutional Review Board and Ethics Committee of the Ministry of Health's Kecioren Training and Research Hospital (Study No: B.10.4.ISM.4.06.68.49/157) and complied with the principles of the Helsinki Declaration. Informed consent was obtained from all participants. In total, 30 healthy patients (age range: 18–25 years; mean age: 21.7 years) with an anterior deep bite of 4 mm or more were selected from the patients presenting to the hospital for treatment. Patients with missing teeth in the maxillary anterior area, a history of incisor trauma, root canal treatment, caries, restorations or previous orthodontic treatment, or an excessive gingival display were excluded from the study. In addition, in order to eliminate the need for aligning and levelling, patients with crowding in the maxillary anterior segment were also excluded.

The participants were randomly distributed between two study groups and one control group (N = 10 participants/40 teeth per group). In the first study group, a utility arch (UA) was used to apply an intrusive force of 100 g to the maxillary central and lateral incisors (approximately 25 g per tooth). In

the second study group, the same force was applied using mini-implants (MI) as points of application. The control group received no orthodontic treatment. Stainless steel brackets (0.018" × 0.025") were bonded to the four maxillary anterior teeth and first molars in the UA group and a conventional 0.016" × 0.016" utility archwire (Elgiloy, Rocky Mountain Morita Co., Tokyo, Japan) was inserted which bypassed the canines and premolars. Tip-back bends, at an angle of approximately 45° to the horizontal plane, were incorporated mesial to the first molars. The utility arch was adjusted so that each incisor received an intrusive force of approximately 25 g (100 g for the four incisors). Metal brackets of the same type were bonded to the maxillary incisors of the MI group and a 0.016" × 0.022" stainless-steel segmental archwire was inserted. The archwire incorporated hooks located at points corresponding to the central and lateral incisor contact points. Self-drilling mini-implants (1.3 mm dia.; 5 mm length) (Absoanchor, Dentos, Daegu, South Korea) were inserted into the alveolar bone at the muco-gingival junction between the roots of the maxillary central and lateral incisors (one each on the left and right side). Standard periapical radiographs were taken to verify the position of the mini-implants in relation to the neighbouring roots. An intrusive force of 100 g (25 g per tooth) was delivered using a nickel-titanium closed-coil spring (3M Unitek, CA, USA) placed between the hooks of the passive arch and the mini-implants, which were loaded immediately after insertion. In each study group, the magnitude of the intrusive force was checked and calibrated using a gram-force gauge (Correx; Ortho Care, Saltaire, UK) during initial activation (baseline) and prior to each LDF evaluation after intrusion.²¹ At the end of each recording point, the intrusive force was reactivated to 100 g. Intrusion continued until an appropriate upper lip-to-incisor relation and optimal overbite were achieved (treatment time: five to six months). Treatment lasted nine months and included a review period.

Laser Doppler flowmeter

PBF was measured using an LDF (Periflux PF 4001, Perimed, Järfälla, Sweden), which recorded the amount of backscattered light. The LDF output signal voltage was linearly related to red-blood-cell flow (number of cells × average velocity), which was recorded in perfusion units (PU) to provide a relative

measurement of blood flow. The LDF used a 1 mW He-Ne laser with a wavelength of 632.8 nm. A straight probe (PF 416, Perimed, Järfälla, Sweden), with a diameter of 2 mm, was used to conduct a laser light-beam of 125 μm (fibre-to-fibre distance: 500 μm) to the measurement site within the dental pulp and to retrieve the backscattered light from the flowmeter. Prior to each measurement, the probe was calibrated for zero voltage and a motility standard of 250 PU using a plastic block (Perimed, Järfälla, Sweden).

Recording procedures

LDF measurements were recorded just prior to intrusion (T0) and after 24 hours (T1), three days (T2), seven days (T3) and three weeks of intrusion (T4). The accuracy and reproducibility of measurements were achieved by providing each patient with a custom-fabricated splint of self-curing acrylic resin that was used to secure the probe in the appropriate position between the gingival margin and the orthodontic bracket. The archwires were removed for a few minutes in each recording session in order to place the splint. The patients were allowed to rest in a supine position in the dental chair for approximately 10 minutes, after which the splints were placed on the teeth and the lips were retracted using cotton rolls. LDF measurements were continuously recorded for each tooth (experimental incisors as well as control teeth) until two minutes of stable PBF data values were registered on the flowmeter screen. All measurements were performed by the same operator (S.E.) under standard clinical conditions and at a constant room temperature. Attempts were made to minimise bias due to subject and probe movement, and pulse rate

and blood pressure were recorded throughout the measurement sessions. None of the participants reported any pain or discomfort during the procedure.

During each session, the mean PUs for the teeth were calculated for the phase of stable values, with peaks attributable to movement artefacts excluded. Data recorded by LDF was transferred to a computer connected to the RS-232 port of the flowmeter using system-specific software (PeriSoft for Windows, Perimed) and stored for later analysis.

Statistical analysis

Statistical analysis was performed using the MedCalc 12.7.7 software program (MedCalc Software bvba, Ostend, Belgium). Changes in PBF within and between groups were assessed by the Wilcoxon Signed Rank and Mann-Whitney *U* tests, respectively, with a statistical significance set at $p < 0.05$.

Results

The patients were clinically evaluated and the magnitude of the intrusive force was verified at each time point. Patients reported no discomfort, and there was no clinical sign of discolouration or pulpal pain in any tooth. The initial intrusive force (100 g) in the UA group was observed to decrease over time (98 g at T1, 97 g at T2, 95 g at T3 and 83 g at T4), whereas the initial intrusive force (total of 100 g) in the MI group remained constant (99.7 g at T1, 99.5 g at T2, 99.5 g at T3 and 99 g at T4).

Table I shows the maxillary incisor PBF values at T0, T1, T2, T3 and T4 for each group. Baseline PBF values did not differ significantly between the study

Table I. Maxillary incisor PBF values at T0, T1, T2, T3 and T4 for each group.

	T0	T1	T2	T3	T4
<i>Utility arch group (N = 40)</i>					
Mean \pm SD	11.67 \pm 0.88	7.56 \pm 0.52	7.57 \pm 0.52	9.27 \pm 1.10	11.67 \pm 0.88
Med (min-max)	11.73 (10.16-13.41)	7.47 (5.91-8.69)	7.49 (5.91-8.69)	9.41 (6.50-11.07)	11.75 (10.16-13.41)
<i>Mini-implant group (N = 40)</i>					
Mean \pm SD	11.66 \pm 0.87	7.11 \pm 0.42	7.20 \pm 0.39	9.34 \pm 0.83	11.66 \pm 0.87
Med (min-max)	11.71 (10.06-13.41)	7.09 (5.91-7.99)	7.16 (6.35-8.05)	9.32 (8.05-12.85)	11.71 (10.06-13.41)
<i>Control group (N = 40)</i>					
Mean \pm SD	11.64 \pm 0.93	11.67 \pm 0.95	11.69 \pm 0.81	11.66 \pm 0.90	11.66 \pm 0.87
Med (min-max)	11.73 (10.16-13.41)	11.69 (10.06-13.51)	11.73 (10.07-13.41)	11.71 (10.16-13.51)	11.63 (10.16-12.99)

and control groups (11.67 ± 0.88 , 11.66 ± 0.87 and 11.64 ± 0.93 PU ($p > 0.05$), respectively, for UA, MI and control groups). No significant changes in PBF were observed in the control group over the course of the study. However, the mean PBF decreased significantly ($p < 0.001$) from T0 to T1 in the UA group (7.56 ± 0.52 PU) and the MI group (7.11 ± 0.42 PU); increased slightly from T1 to T2 (UA: 7.57 ± 0.52 PU, $p = 0.066$; MI: 7.20 ± 0.39 PU, $p < 0.001$); and increased significantly from T2 to T3 (UA: 9.27 ± 1.10 and MI: 9.34 ± 0.83 PU, $p < 0.001$) and from T3 to T4 to attain levels similar to those measured prior to intrusion (UA: 11.67 ± 0.88 , $p = 0.150$; MI: 11.66 ± 0.87 PU, $p = 0.102$). The mean PBF values of the two study groups varied significantly at T1 and T2 ($p < 0.001$), but not at T0 ($p = 0.919$), T3 ($p = 0.593$), or T4 ($p = 0.904$). The changes in maxillary incisor PBF that occurred over time in the study groups, compared with the relatively constant PBF in the control group, are illustrated in Figure 1.

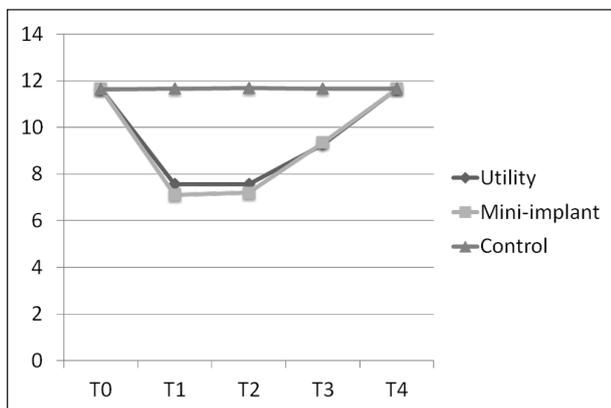


Figure 1. The changes in the maxillary incisor PBF over time in the study groups, compared to the relatively constant PBF in the control group.

Discussion

The aim of the clinical study was to evaluate and compare the PBF values of maxillary incisors over a three-week period during intrusion using either utility arches or mini-implants. Baseline PBF values did not differ significantly between groups and were in the same range reported by previous studies (7.6 to 14 PU).^{22,23} No significant mean change in PBF was observed in the control group over the course of the study, indicating that the change in PBF registered in subjects undergoing intrusion was unrelated to repeated measurement, flowmeter calibration, or test sensitivity. PBF values were significantly lower in both

study groups compared with the control group at 24 hours and three days of intrusion ($p < 0.001$). The sharp decrease in PBF was likely due to strangulation and stasis of blood flow to the pulp, which has been reported by histological studies.^{12,17,24,25} The initial reduction of PBF in the study groups was followed by a pattern of gradual time-related recovery, with a marked increase in PBF observed after seven days of intrusion when compared with the readings at 24 hours and three days. At week three, PBF attained levels similar to those measured prior to the application of an intrusive force in the test groups. The eventual increases in PBF can be explained by reactive hyperaemia compensating for the earlier blood-flow reduction.²⁶

The mean PBF values of the two study groups were found to be similar over the course of the study, with the exception of the 24 hours and three days of intrusion time periods. At these times, the PBF in the mini-implant group was significantly lower than in the utility arch group. This could be due to the fact that extrusion of the anchored molars resulted in less force delivered to the incisors in the utility arch group compared with that delivered to the mini-implant group. Whereas the initial total intrusive force of 100 g had decreased to 97 g after three days and to 83 g after three weeks in the utility arch group, the intrusive force remained constant (total > 99 g) over the course of the study in the mini-implant group, as the molars were not used for anchorage and therefore maintained their positions during incisor intrusion. Although the magnitude of the intrusive force in the utility arch group showed a greater decrease after three weeks of intrusion compared with that seen at one and three days, statistically significant differences between the mean PBF values of the utility arch and mini-implant groups were not observed at T3 or T4. Therefore, it may be speculated that the effect of an intrusive force on PBF is less severe during the later stages of intrusion, especially after the first three days. Overall, the findings of the present study indicate that the initial change in PBF following intrusion is reversible and that the pulpal circulatory system of an incisor tooth can cope with the apical displacement produced by an intrusive force of 25g. These findings are supported by the majority of histological and in vivo human and animal studies reporting a reduction in pulp-tissue vascularity during the initial period of force application.^{10,12,17,24,25,27} Using in vivo

microscopy, Guevara and McClugage¹² observed an initial decrease in blood flow in rats. A histological study by Anstendig and Kronman¹⁰ found a decrease in the number of blood vessels following the application of orthodontic force in dogs, and a study by Konno et al.,²⁷ which evaluated changes in pulpal morphology and blood flow in response to intrusion in an animal model, found that pressure generated by an intrusive force reduced the number of capillary blood vessels below the apical foramen and produced slight degenerative changes in the pulp tissue. Human *in vivo* studies by Brodin et al.,²⁸ Sano et al.¹⁷ and Ikawa et al.,²⁰ evaluating PBF changes using LDF, also reported that intrusive force initially produced a significant reduction in PBF, in support of the current findings. Contrary to these reports, several histological studies^{13,14,29} have reported an increase in pulp-tissue vascular response in animals. A histomorphometric study on rats conducted by Nixon et al.²⁹ reported an increase in the number of functional pulpal vessels after the application of orthodontic force. Kvinnsland et al.¹³ utilised fluorescent microspheres and showed a substantial increase in blood flow in the dental pulp of rat molars following mesial tipping and Vandevska-Radunovic et al.¹⁴ also reported increased blood-flow values in the pulp of actively moved rat molars. In each of these studies, the authors attributed the increased vascular activity to the propagation of an inflammatory reaction. However, vasodilatation and increased vascular permeability with fluid exudation in connective-tissue compartments^{30,31} are not necessarily evidence of an increase in blood flow, as dilation of blood vessels can also occur during stasis in connection with a decrease in blood flow.³² Furthermore, quantitative analysis of blood-flow rates by LDF, which is proven to be the most effective early indicator of pulpal changes, would have yielded better results than the histological observation and fluorescent microspheres used in the earlier studies. Finally, in further contrast to the present results, Barwick and Ramsay¹⁹ reported that the application of a brief intrusive orthodontic force does not alter PBF in humans.¹⁹ This finding of a non-significant reduction in PBF could be explained by the brief time (only four minutes) of force application, the small sample size ($N = 8$), and error variability introduced into the laser Doppler signal.¹⁹

The finding of the present study that changes in blood flow were temporary and were followed by a pattern

of gradual recovery over time, with a marked increase in PBF after seven days and a return to normal within three weeks, is supported by numerous previous reports.^{11,17,18,27,28,33-35} A recent histological study by Derringer et al.³³ examined pulp harvested from human teeth extracted after orthodontic intrusion. The pulps were embedded in collagen and cultured in growth media for up to four weeks. The formation of new micro-vessels between Days 5–10 was confirmed by light microscopy and electron microscopy,³³ with significantly greater numbers of micro-vessels noted between Days 5–10 in orthodontically-moved teeth when compared with controls. Brodin et al.²⁸ and Sano et al.¹⁷ also showed that the orthodontic intrusion of teeth evoked a temporary reduction in PBF. Rohaya et al.³⁵ reported that gingival crevicular fluid levels of aspartate aminotransferase, an intracellular enzyme released to the extracellular environment upon cell death, was highest in the first week of intrusion and gradually lessened during the next three weeks. This also supports the hypothesis that orthodontic treatment can cause temporary, reversible metabolic changes in pulp tissue. Similarly, Grünheid et al.³⁴ found that a number of pathological signs in rat pulp tissue peaked at 24 and 72 hours after force application, but returned to initial levels after 168 hours. It was concluded that controlled mechanical forces during orthodontic treatment, if not excessive, caused only transient pulpal changes, as tissue regeneration was initiated almost immediately after the onset of tooth movement.

In contrast, other studies claim that an intrusive orthodontic force results in permanent damage to pulpal tissue. A histological study by Marshall,³⁶ conducted in 1933, described strangulation and necrosis of pulp in a *Macaca rhesus* monkey after the application of an intrusive force. Spector et al.³⁷ also referred to two cases of human tooth devitalisation resulting from orthodontic therapy. Butcher and Taylor²⁵ reported that vacuole formation, dilation of vessels and thrombosis could be observed in monkey incisors as a result of an orthodontic force; however, the authors questioned their results, since the controls also exhibited vacuole formation and blood vessel congestion. Given the questionable histological methods used in previous studies and the lack of sufficient convincing data from *in vivo* research demonstrating actual microvasculature changes in pulp following the application of orthodontic force, it

is not possible to state unequivocally that orthodontic treatment can induce pulpal death. Therefore, more studies are necessary, and expanding the focus of research into cellular- as well as molecular-level processes taking place within dental pulp tissue after orthodontic tooth movement could yield valuable information.

The orthodontic literature reports only one study that evaluated blood flow in human dental pulp after receiving an intrusive force delivered by a utility arch¹⁷ and one study evaluating blood flow in human dental pulp after receiving an intrusive force via mini-implants.¹⁸ In agreement with the present study, both of these studies found an initial decrease in PBF followed by gradual recovery; however, neither provided a comparison of PBF values between the two intrusion methods.

The present study had two main methodological limitations related to the whole-mouth experimental design and the use of LDF to measure PBF. While a split-mouth design would remove the problem of inter-subject variability and increase the power of a study when compared to a whole-mouth design,^{38,39} the contra-lateral incisors could not be used as controls because all four incisors were simultaneously intruded using either a utility arch or mini-implants. The technical circumstances required that the control and experimental groups be from different individuals, and so the control group used in this study provided useful information about normal range and mean response thresholds for maxillary incisors as well as inter-subject variability.

The LDF and PBF values obtained through this method may be affected by probe positioning,⁴⁰ regional variations in blood flow within the pulp^{14,41} and contamination of the PBF signal from blood flow of non-pulpal origin.^{42,43} In order to stabilise the probe and overcome these problems, the present study used a custom-made acrylic-resin splint to maintain the probe in contact with the tooth and create a reproducible position for follow-up measurements. In conjunction with the splint, cotton rolls were applied between the lip and teeth in order to minimise the contribution of neighbouring pulp and gingiva to the flux signal, since a rubber-dam cannot be applied to teeth with brackets.⁴⁴ A similar technique has been successfully employed in earlier studies.^{44,45} Finally, in order to improve the validity of measurements, special care was taken to maintain ambient temperatures and

patient-related factors such as position, rest and stress levels.⁴⁴

Conclusion

The study findings demonstrated that, despite slight regressive changes in pulpal tissue over the short term, PBF values returned to initial levels within three weeks, indicating that changes observed in PBF via both intrusion methods are reversible. Despite a tendency towards a more severe drop in PBF in the mini-implant group during the first three days of intrusion, no significant differences between utility arch and mini-implant groups were observed at any other time in the study. This indicated that the changes observed were not directly related to the method of intrusion employed.

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