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Tooth movement and vascularity of the dental pulp: a pilot study

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The effect of orthodontic tooth movement on the dental pulp was assessed histologically in twelve subjects. The participants in this study required the extraction of at least two maxillary first premolars for orthodontic treatment. They were asked to wear a maxillary removable appliance that acted to move a randomly determined premolar in a buccal direction. The appliance was designed to avoid contacting the contra-lateral tooth that was used as the matched control. The appliance was initially worn for a week to ensure patient comfort and cooperation. The appliance was then activated and the patient dismissed. After two weeks, the appliance was reactivated. Both the control and experimental teeth were extracted three weeks later, on the thirty-fifth day of activated appliance wear. The teeth were fixed, decalcified and sectioned. The sections were stained with haematoxylin and eosin for histological examination. This investigation demonstrated that orthodontic tooth movement did have an effect upon the dental pulp, causing vasodilation in the pulp of an orthodontically stressed tooth.
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Introduction

Very little work has been done in the study of the effects of orthodontic forces on the dental pulp in human samples with matched controls. Research has usually involved the use of animal models. Of the work done on human subjects, there have been few studies with matched controls.[1,2]

Orthodontic tooth movement causes resorption of bone on the side of pressure and apposition of bone on the side of tension.[3] The predominant cell type in the dental pulp is the fibroblast. However, the pulp also contains odontoblasts, Schwann cells, endothelial cells, blood cells and undifferentiated mesenchymal cells. Histologically, the pulp can be divided into a central zone and a peripheral zone. In the central zone large arteries, veins and nerves enter the apical canal and proceed to the coronal pulp chamber. Fibroblasts are the predominant cell type in this region. The peripheral zone is characterised by an odontogenic zone including odontoblasts.[4,5,6]

Stuteville[7] was the first to suggest that orthodontic force may cause injury to the dental pulp, resulting in hyperemia or vascular stasis. Research[8] prior to this had suggested that orthodontic forces had no effect on the pulp. Oppenheim[9] described the phenomenon of diapedesis of erythrocytes and leukocytes, which he attributed to an increased pressure within the veins. Forces acting on the apical region of the tooth causes an impairment of venous return. The resulting vascular stasis causes an accumulation of fluid between the pulpal tissue elements and the formation of vacuoles in the odontoblasts.

Astendig and Kronman[10] noted that in dogs there was an increase in collagenous fibres throughout the pulp, and a corresponding decrease in the cellular content of the pulp subsequent to orthodontic tooth movement. There was a decrease in fibroblasts, but an increase in the number of inflammatory cells, especially in the coronal portion of the pulp. The number of blood vessels also diminished. Stenvik,[11] in a study on the effect of intrusion on teeth, also

noted an increased number of macrophages and leukocytes, both within and outside the vessels in some teeth.

Stenvik and Mjor[12] noted that inflammatory responses were not generally seen in their study on tooth intrusion and that vacuolisation of the odontoblastic layer seemed to be a good indicator of changes in the dental pulp resulting from tooth intrusion. Although vacuolisation of the odontoblastic layer was a more common finding in orthodontically intruded teeth, the authors could not be certain that this was not an artifact of specimen preparation rather than the effect of intrusive forces.

Stanley *et al.*[13] suggested that there may be capillaries that remain non-functional until the pulp is traumatised, thus allowing for an increased blood flow to the pulp. Using fluorescent microspheres, Kvinnsland *et al.*[14] showed a substantial increase in blood flow in the dental pulp of mesially tipped rat molars. Nixon *et al.*[15] noted a significant vascular effect on the pulp due to orthodontic forces. They noted an increase in the numbers of functional pulpal vessels in rat molars after application of an orthodontic force.

A study of the pulpal tissue respiration rate by Hammersky *et al.*[16] showed that orthodontic force of a relatively short duration (72 hours) reduced the oxygen consumption rate in the human premolar. Another point of interest noted was a correlation between increasing age and a greater degree of depression in the pulpal respiration rate. Stenvik[11] noted in a study of human subjects that the degree of pulpal reaction to orthodontic forces was dependent upon magnitude of force and the degree of root development.

Materials and methods

Fifteen orthodontic patients requiring bilateral extraction of maxillary first premolars were selected within the Orthodontics Section of the University of Queensland Dental School. Patients undergoing orthodontic treatment involving extraction of the upper first premolars were invited to participate in the study. The target sample size was fifteen subjects. Written permission was sought on the basis of informed consent. The guidelines for human experimentation prescribed by the NHMRC of Australia were followed and the project was approved by the institutional ethics committee. The subjects selected did not have any prior history of dentinal hypersensitivity, restorative or endodontic involvement in either the experimental or control teeth.

Each subject was provided with an upper removable appliance designed to tip one premolar buccally and to maintain the position of the contralateral control tooth. The tooth to be moved, either a 14 or 24, was chosen randomly. A standard appliance was used, consisting of Adam's clasps on 16 and 26, a labial bow on the four upper incisors, an anterior bite plate and a palatal piston screw (Dentaurum*) to the tooth to be moved. The bite plate unloaded the teeth from occlusal forces while the piston screw provided a standard force mechanism. Each patient wore the appliance for a period of seven days to ensure that it was comfortable and could be worn continuously. The screw was then activated and the tooth to be moved was subjected to a buccal force for a period of fourteen days. The appliance was then reactivated on the fourteenth day. The experimental and control teeth were then extracted with forceps under local anaesthetic at same appointment on the thirty-fifth day after initial activation. By this stage, the experimental teeth had on average been moved a total of 2 mm at the level of the crown.

Upon extraction, the two premolar teeth were placed immediately in Bouin's solution for two minutes to perfuse the apical tissues. In order to enable sufficient perfusion of fixative into the dental pulp, a cut was made on the buccal surface of the root, at approximately one third of the root length coronal of the root apex. The tooth was then fixed in Bouin's solution at 4° C for forty-eight hours. The teeth were coded numerically for blind analysis. This information was not decoded until the study was complete. Personal details or other identifying criteria were not recorded on the specimen containers.

Following two changes of buffer, the teeth were decalcified in a solution of 20 per cent v/v formic acid (90 per cent concentration) and 10 per cent v/v formalin in distilled water at room temperature with continual agitation for up to two weeks, with the end point of demineralisation being determined radiographically. The teeth were then washed in distilled water for four hours, dehydrated with graded ethanol, cleared in cedarwood oil and embedded in paraffin by standard histological procedure. Serial 5 µm sections were cut and stained with haematoxylin and eosin for light microscopy histological evaluation.

The sections were evaluated histologically to ascertain the effects of orthodontic movement on the vasculature of the dental pulp. The odontoblast layer of the sections was examined for possible vacuolization, and the pulpal tissue examined for possible signs of disruption and inflammation under a light microscope at 10x, 25x, 40x and 100x magnification by two independent observers.

Initial histological examination of the samples revealed that most of the changes occurring in the blood vessels of control and experimental teeth could be found in the region of the cemento-enamel junction (CEJ). Accordingly, a decision was made to

* Dentaurum, D-7530P Pforheim, Fed. Rep. of Germany.

assess only the blood vessels around that area. The blood vessels in the teeth that had been experimentally moved appeared to be dilated and congested when compared with the control teeth. In some instances, the vascular differences manifested as an increase in the size of the blood vessels due to vasodilation and congestion with blood cells. In other cases, the vascular difference was a combination of vasodilation as well as an increase in the number of small blood vessels within the pulps of the orthodontically moved teeth.

In an attempt to quantify these differences, it was decided to digitise the blood vessels to enable some form of quantitative comparison between the two groups. Although this form of analysis would not differentiate between vasodilation and an increase in the number of blood vessels within the pulp, it would allow a quantitative comparison of the blood vessel area between the experimental teeth and their matched controls.

Each slide was first mounted and examined under a light microscope† at 25 x magnification and the CEJ found. The pulpal tissue between the buccal and lingual CEJ and slightly coronal and apical to the CEJ was then examined in greater detail. A video image of the area was captured via a video camera‡ and transferred to a computer§ via a video capture program.‖ The digital image from the computer was then transferred to an image analysis program.¶ During this transfer, the colour digital image was converted to a black and white image, which was then used to determine the area of blood vessels within the region to be examined (Figure 1).

The blood vessel area in the region examined was assessed by digitising the outlines of blood vessels. To facilitate uniformity of results, a blood vessel for the purpose of this experiment was defined as an endothelial lined space, with or without the presence of red blood cells. It is important to note that:

1. Red blood cells that were not enclosed by an endothelial lining were not included when assessing blood vessel area.
2. Any spaces present that were not lined by endothelial cells were excluded from the assessment, as these spaces could represent an artefact of slide preparation rather than a blood vessel. To aid in identifying and digitising the blood vessels during this phase, the region of interest was examined under 100 x magnification. At this magnification, the smaller blood vessels could be identified, and the shapes of the blood vessels could be more accurately determined, aiding in the digitising of the blood vessel outline on the computer image.

Once all the blood vessels in the region of interest on each slide had been digitised, the results were added to provide the total blood vessel area in the region examined. In order to facilitate comparison between the control and experimental teeth, the total pulpal area examined on each slide was also measured. This was defined as the area examined, minus any dentine and artefacts present in the section examined.

By using a ratio of blood vessel area against the total area examined for each tooth, it was possible to compare differences in blood vessel area between control and experimental teeth.

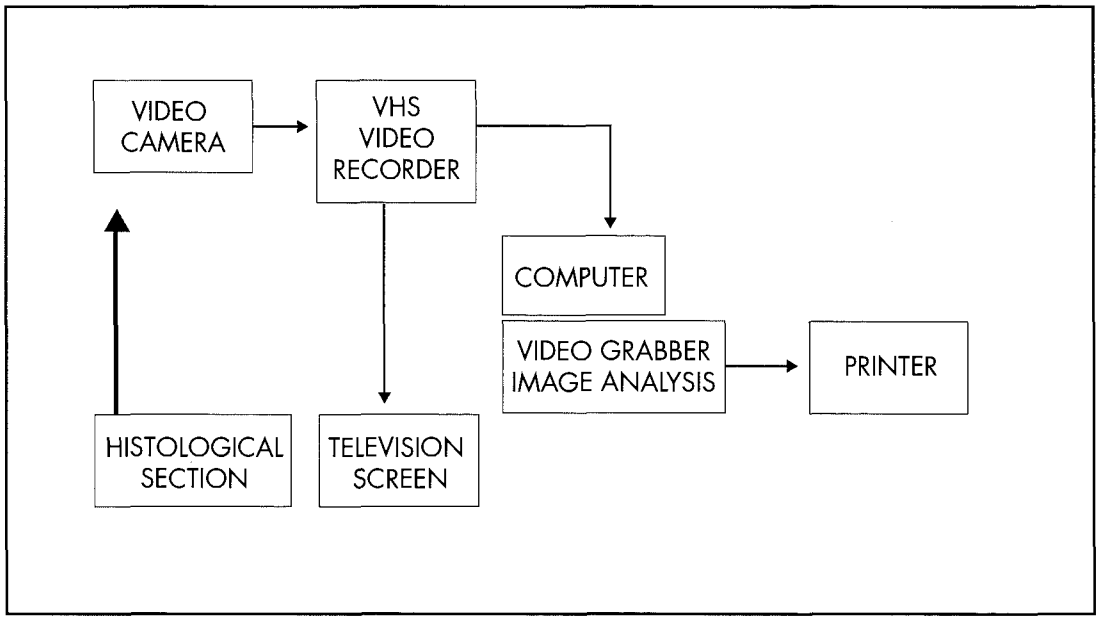

Figure 1. Computer-based image analysis system.

† Olympus BHB.

‡ TMC-6 Video Camera manufactured by Pulnix

§ Apple Macintosh LC630 (c) Apple Computers Inc

‖ Apple Video Player (c) Apple Computers Inc

¶ NIH Image, Version 1.57 by Wayne Rasband, National Institute of Health, U.S.A.

Results

Of the fifteen patients initially recruited for the study, one was excluded because the patient had not been wearing the appliance two weeks prior to extraction. Two patients were also excluded due to the presence of artefacts within the histological sections making analysis impossible. In both cases, problems in sectioning or splitting of the tooth resulted in a tearing of the pulpal tissue.

It was noted that there were vascular differences between the control and experimental teeth. Blood vessels in the teeth that had been experimentally moved appeared to be dilated and congested when compared with the control teeth. The results of the blood vessel area analysis are summarised in Table I and Figure 2.

Table I. Blood vessel area and total pulpal area assessed in both control and experimental sections.

PATIENT	Area of region (in pixels)			
	CONTROL TEETH		EXPERIMENTAL TEETH	
	Pulpal region studied	Blood vessel area in region studied	Pulpal region studied	Blood vessel area in region studied
Patient A	79770	2669	71101	3092
Patient B	75287	2966	72194	4524
Patient C	80875	2380	77574	3103
Patient D	80945	4898	67279	7840
Patient E	81600	5164	77908	5972
Patient F	75696	1808	79336	2527
Patient G	79996	2748	74516	3103
Patient H	81600	4051	75877	4242
Patient I	69825	3103	54445	3271
Patient J	43406	1269	73917	4877
Patient K	81600	1576	76339	2734
Patient L	69195	2869	67591	3778

In all cases examined, there was a difference in blood vessel area between the control and experimental teeth. In all twelve patients assessed in this preliminary study, it was demonstrated that orthodontic tooth movement with a removable orthodontic appliance resulted in an increase in blood vessel area within the pulpal tissue examined. Although the degree of change was variable between different patients, there was a clear difference between the control and experimental teeth in all cases. This difference was found to be significant (at $p<0.01$).

Discussion

A consistent difference was noted in the vascular area between the dental pulps of the control and experimental teeth. In all cases, orthodontic movement resulted in an increase in the blood vessel area of the experimental tooth. The difference between the two groups was statistically significant. This finding supports the suggestion by Stanley *et al.*[13] of the presence of capillaries that are nonfunctional until the pulp is stimulated in some manner. Experiments by Stenvik and Mjor[12] also suggested an increase in vascularity of orthodontically stressed teeth with an increase in sub-odontoblastic capillaries being noted in their experimentally intruded teeth compared with their controls. Nixon *et al.*[15] also noted a significant vascular change with an increase in the numbers of functional pulpal vessels following orthodontic force application.

Studies of pulpal blood flow following orthodontic force application also support the findings of this preliminary study. Kvinnsland *et al.*[14] showed a substantial increase in blood flow in the dental pulp of

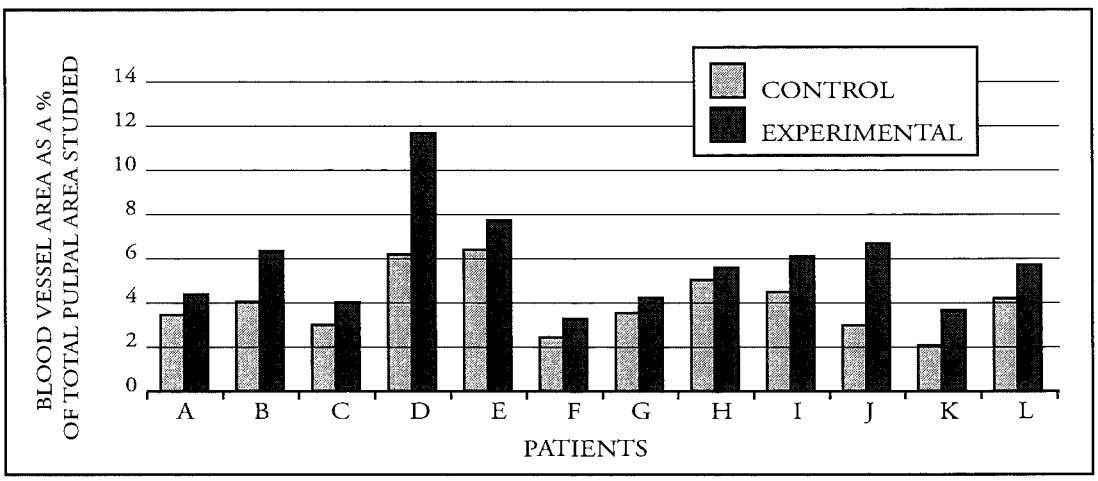

Figure 2. Comparison of blood vessel area between control and experimental teeth.

mesially tipped rat molars. Similar work by Vandevska-Radunovic *et al.*[17] on rats and by McDonald and Pitt Ford[18] on human subjects also suggested a similar picture, noting an initial decrease in blood flow to the dental pulp following orthodontic force application. However, blood flow then increased until it reached a peak, seven days after the application of the force. This pattern of blood flow change may correspond to the rate of tooth movement experienced. Lee[19] noted that, in general, tooth movement is biphasic: the first phase consists of rapid movement occurring within the first week of force application, the timing corresponding with the timing of the blood flow increase noted by Vandevska-Radunovic *et al.*;[17] this is then followed by a second phase with a slower rate of movement.

In contrast, Melsen[19] noted little or no difference between the control and intruded teeth in monkeys. This study involved the use of low forces, which could account for the lack of response. Forces in the magnitude of 10 g were used in Melsen's study. Forces of 170 g were used in the present study.

Kvinnsland[14] has commented that vasodilation represents the initial stages of inflammation and is the result of an increased blood flow to the affected area. Subsequent inflammatory events, such as increased vascular permeability and extravasation of inflammatory cells, will occur only if the inflammatory process progresses. If the orthodontic force levels in the present study had been increased or a different movement utilised, greater signs of inflammation might have been noted in the pulps of the experimental teeth. Duration of force exposure may also be an important factor that should be considered. In this study, the teeth were extracted five weeks after initial force application.

Conclusion

This study has shown that orthodontic tooth movement does have an effect upon the vascularity of the dental pulp in human subjects. Simple tipping of a tooth 2 mm buccally results in an increase in the vascular area in the pulp of that tooth.

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References

1. Cwyk F, Saint-Pierre F, Tronstad L. Endodontic implications of orthodontic tooth movement. IADR Abstracts 1984; #1039.
2. Popp TW, Artun J, Linge L. Pulpal response to orthodontic tooth movement in adolescents: a radiographic study. Am J Orthod Dentofac Orthop 1992; 101: 228-33.
3. Graber TM, Vanarsdall RL. Orthodontics: Current principles and techniques. Mosby, 2nd Edition. 1994; 96-192.
4. Avery JK. Essentials of oral histology and embryology: a clinical approach. Mosby, 1992; 105-22.
5. Bhaskar SN. Orban's Oral histology and embryology 10th Edition. The C.V. Mosby Company, 1986; 135-74.
6. Ten Cate AR. Oral histology: development, structure and function. 3rd Edition. The C.V. Mosby Company, 1989; 157-96.
7. Stuteville OH. A summary review of tissue changes incident to tooth movement. Angle Orthod 1938; 8: 1-20.
8. Orban B. Biologic problems in orthodontia. J Am Dent Assoc 1936; 23: 1849-70.
9. Oppenheim A. Human tissue responses to orthodontic intervention of short and long duration. Am J Orthod 1942; 28: 263-301.
10. Astendig HS, Kronman JH. A histologic study of pulpal reaction to orthodontic tooth movement in dogs. Angle Orthod 1972; 42: 50-5.
11. Stenvik A. Pulp and dentine reactions to experimental tooth intrusion (A histologic study: long-term effects). Rep Cong Eur Orthod Soc 1969; 449-64.
12. Stenvik A, Mjor IA. Pulp and dentine reactions to experimental tooth intrusion. A histologic study of initial changes. Am J Orthod 1970; 57: 379-85.
13. Stanley HR, Weissman MI, Michanowizc AE, Billizzi R. Ischaemic infarction of the pulp: sequential degenerative changes of the pulp after traumatic injury. J Endo 1978; 4: 325-35.
14. Kvinnsland S, Heyeraas K, Ofjord KS. Effect of experimental tooth movement on periodontal and pulpal blood flow. Eur J Orthod 1989; 11: 200-5.
15. Nixon CE, Saviano JA, King GJ, Keeling SD. Histomorphometric study of dental pulp during orthodontic tooth movement. J Endod 1993; 19: 13-16.
16. Hammersky PA, Weimer AD, Traintor JF. The effect of orthodontic force application on the pulpal tissue respiration rate in the human premolar. Am J Orthod 1980; 77: 368-78.
17. Vandevska-Radunovic V, Kristiansen A, Heyeraas KJ, Kvinnsland S. Changes in blood circulation in teeth and supporting tissues incident to experimental tooth movement. Eur J Orthod 1994; 16: 361-9.
18. McDonald F, Pitt Ford TR. Blood flow changes in permanent maxillary canines during retraction. Eur J Orthod 1994; 16: 1-9.
19. Lee BW. The force requirements for tooth movement. Part I: Tipping and bodily movement. Aust J Orthod 1995; 13: 238-48.
20. Melsen B. Tissue reaction following application of extrusive and intrusive forces to teeth in adult monkeys. Am J Orthod 1986; 89: 469-75.