Effect of sodium hypochlorite on mechanical properties of dentine and tooth surface strain

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Abstract

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Aim The aim of this study was to test the null hypothesis that sodium hypochlorite irrigation of root canals does not alter the properties of dentine and contribute to the weakening of root-treated teeth.

Methodology The effect of two concentrations (0.5%, 5.25%) of sodium hypochlorite (NaOCl) and saline on (i) the elastic modulus and flexural strength of machined dentine bars, and (ii) changes in strain of 'whole' extracted human teeth were evaluated. One hundred standardized plano-parallel dentine bars ($> 11.7 \times 0.8 \times 0.8$ mm) were randomly divided into the three groups, immersed for 2 h in the respective solutions and then subjected to a three-point bend test. Changes in strain of each of 10 teeth on cyclical nondestructive occlusal loading were measured using electrical resistance strain gauges bonded to the cervical aspects. Each tooth had its crown and enamel reduced and root canal prepared. These were irrigated sequentially in a series of four separate, 30-minute regimes; initial-saline, 0.5% NaOCl, 5.25% NaOCl and final-saline. The changes in strains after each irrigation regime were compared.

Results There was a significant decrease in elastic modulus of the dentine bars immersed in 5.25% NaOCl compared with the saline group ($P < 0.01$). There was also a significant decrease in flexural strength of the dentine bars in the 5.25% NaOCl group compared to both the saline and 0.5% NaOCl groups ($P < 0.01$). The strain data from the nondestructive tooth loading tests revealed significant increases in tensile strain between the initial-saline and the final-saline stages ($P < 0.01$). Significant increases in compressive strains were also found between initial-saline and 5.25% NaOCl; and between 0.5% NaOCl and 5.25% NaOCl stages ($P < 0.01$).

Conclusions The null hypothesis was rejected, 5.25% NaOCl reduced the elastic modulus and flexural strength of dentine. Irrigation of root canals of single, mature rooted premolars with 5.25% NaOCl affected their properties sufficiently to alter their strain characteristics when no enamel was present.

Keywords: dentine, irrigant, sodium hypochlorite, strain.

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Introduction

There is a widely held belief that root-treated teeth are weakened and more susceptible to fracture than vital teeth (Rosen 1961, Johnson *et al.* 1976, Gher *et al.* 1987), yet conclusive epidemiological evidence is still lacking. Nevertheless, there is convincing circumstantial evidence for the putative causes of nonvital and root-treated teeth to fracture (Burke 1992). The main causes may be divided broadly into three areas; loss of tooth tissue, altered physical properties of dentine and altered proprioception/nociception (Gutmann 1992, Gulabivala 1995). It is probable that these factors interact cumulatively to influence tooth loading and distribution of stresses, ultimately increasing the possibility of catastrophic failure.

Tooth tissue loss reduces the force required to strain and ultimately fracture teeth *in vitro* with the pattern of loss influencing the magnitudes of the induced strains (Vale 1956, Mondelli *et al.* 1980, Larson *et al.* 1981, Panitvisai & Messer 1995). Evidence from clinical

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studies confirms these observations (Cavel *et al.* 1985, Hansen *et al.* 1990). The relative importance of disruption of the marginal ridge and the width and depth of cavities continues to be debated, but it is also probable that tooth anatomy plays an important part (Khera *et al.* 1990). The presence of an endodontic access cavity may weaken teeth further, but the extent of this contribution is unresolved (Reeh *et al.* 1989, Howe & McKendry 1990, Panitvisai & Messer 1995). Wide coronal flaring of canals has been implicated as an additional factor in fracture of root-treated teeth (Hansen & Asmussen 1993).

It has been proposed that loss of pulp vitality alters the properties of dentine. This has been assessed by changes in moisture content (Helfer *et al.* 1972, Huang *et al.* 1992, Jameson *et al.* 1993, 1994, Papa *et al.* 1994), nature of collagen (Rivéra *et al.* 1988) and standard laboratory physical properties (Baraban 1967, Lewinstein & Grajower 1981, Carter *et al.* 1983, Huang *et al.* 1992, Sedgley & Messer 1992). Unfortunately, the findings have been contradictory or equivocal and as yet no definitive proof of mechanical weakening of dentine exists. One fundamental problem is that since all tests are carried out in the laboratory, the dentine tested is by definition nonvital.

Two studies (Löewenstein & Rathkamp 1955, Randow & Glántz 1986) have inferred that loss of pulp tissue compromises the tooth's proprioceptive/nociceptive function. This may predispose the tooth to greater loading in function with consequent increased probability of fracture (Randow & Glántz 1986).

In addition to the cumulative effect of these factors, intracanal irrigants, medicaments and materials may also play a part in influencing the physical and mechanical properties of dentine; however, these have not been studied extensively. Eugenol-containing root canal sealers, for example, can harden intracanal dentine (Biven *et al.* 1972), whilst chloroform, xylene and halothane soften dentine (Rotstein *et al.* 1999). Sodium hypochlorite (NaOCl) is widely recommended as a root canal irrigant because of its antibacterial (Shih *et al.* 1970, Harrison & Hand 1981, Byström & Sundqvist 1983) and organic tissue disintegration properties (Hand *et al.* 1978, Rosenfeld *et al.* 1978, Thé 1979, Gordon *et al.* 1981, Moorer & Wesselink 1982, Baumgartner & Cuenin 1992). Since dentine is composed of 22% organic material by weight, NaOCl could potentially weaken it (Sakae *et al.* 1988, Barbosa *et al.* 1994, Haikel *et al.* 1994). The recommended concentration of sodium hypochlorite ranges from 0.5% to 5.25% (Spångberg *et al.* 1973, Harrison 1984, Baumgartner & Cuenin 1992), with no consensus on the ideal concentration. The greater effectiveness of NaOCl at higher concentrations (Moorer & Wesselink 1982) has prompted some clinicians to adopt the higher end of the range. It is possible that whilst this makes tissue disintegration more effective (Baumgartner & Cuenin 1992), it may also have a deleterious effect on the properties of dentine.

It was hypothesized that exposure of dentine to a high concentration of sodium hypochlorite would reduce its strength and resilience. It was proposed to test this effect at two levels. First, the local effect on dentine bars and secondly, the effect of root canal irrigation on 'whole' tooth strain.

The specific aims of the two parts of the study were:

1 To measure and compare elastic modulus and flexural strength of standardized bars of dentine immersed in saline, 0.5% NaOCl and 5.25% NaOCl using a three-point bend test.

2 To compare change in tooth surface strain of extracted human teeth upon cyclic nondestructive occlusal loading as a result of sequential irrigation of prepared root canals in a series of four separate 30-minute regimens (initial-saline, 0.5% NaOCl, 5.25% NaOCl and final-saline).

Materials and methods

Measurement of elastic modulus and flexural strength of dentine bars

Fifty intact, caries and crack-free, human teeth with mature roots stored in 4% formal-saline immediately after extraction were sectioned longitudinally using a motorized saw (Exakt, GmbH, Nordestedt, Germany) into 100 plano-parallel dentine bars ($0.8 \times 0.8 \times > 11.7$ mm). The bars were randomly allocated to be soaked in one of three solutions (37 in saline, 31 in 0.5% NaOCl, 32 in 5.25% NaOCl).

The bars were soaked in 200 mL of the respective solutions in a beaker that was constantly agitated. The solutions were changed every 10 min. The total immersion time was 2 h. At the end of this time the test solutions were neutralized and the bars maintained in water. The bars were then subjected to a three-point bend test using a custom-made jig mounted in a load testing machine (H25K, Hounsfield, London, UK). A cross-head speed of 0.5 mm min^{-1} and a 0–20 N load range with 200 N load cell were used. The cross-head and test data were used to calculate the elastic modulus and flexural strength.

Means and standard deviations were calculated and the one-factor, independent measures analysis of variance (ANOVA) test was used to compare the three groups. The level of significance was set at 0.01 and the calculated *P*-value would have to be less than 0.01 before the results were considered significant.

Table 1 Groups and number of teeth for cyclic nondestructive occlusal loading tests

	Control group					
	Positive 1	Positive 2	Negative 1	Negative 2	Test group	Configuration tooth
No. of teeth	1	1	1	1	10	1
Total			15			

Measurement of change in strain of teeth

Test specimens

Fifteen intact human mandibular second premolars with single roots and canals with mature roots were used after temporary storage in 4% formal-saline. The precise age of the teeth was unknown, but all were caries and crack-free when examined under a stereomicroscope. The crowns were removed 4 mm above the cemento-enamel junction (CEJ), leaving a flat surface perpendicular to the long-axis of the tooth. In 14 teeth, the remaining enamel around the circumference of the tooth was also removed. Four teeth acted as positive and negative controls and 10 as the experimental teeth (Table 1). The fifteenth tooth acted as a control with intact enamel. An oval access preparation was made and the canals prepared to a standard flare with an apical size 40 with apical control zone size 80 as described by Roane (1991). A Gates Glidden size 2 was introduced to 3–5 mm from the apex, the remaining preparation being flared sequentially with larger Gates Glidden drills. The apices of the prepared teeth were sealed with two coats of nail varnish.

Strain gauge attachment

The teeth were mounted in acrylic resin to within 2 mm of the CEJ, contained inside brass moulds and allowed to cure and stabilize over several days. Stacked, three-element, rosette, foil electrical resistance strain gauges (gauge element resistance – 120 Ω, gauge factor – 2.06, with integral 1 m leads, TML, Tokyo, Japan) were then bonded to the mesial surfaces of the teeth to measure strains. The surface of the tooth was prepared by etching with 10% maleic acid (Scotchbond Multi-purpose, 3M, MN, USA) for 60 s, washing with water for 20 s and drying with a stream of air. The backings of the strain gauges were cleaned and bonded with 'CN' quick-setting cyanoacrylate adhesive (TML, Tokyo, Japan) onto the prepared area of the tooth. The gauge was mounted with the 'x' gauge vertical so that it was parallel to the direction of loading. The wire leads from the strain gauges were embedded into the resin blocks to prevent accidental detachment. The exposed wire leads and rosette gauges were protected with three coats of 'M-Coat D' air-drying acrylic varnish (Measurements Group (UK) Ltd, Basingstoke, UK). The teeth were stored in 100% humidity for 1 week to allow the resin, adhesives and varnish to set completely.

The exposed ends of the leads were soldered to shielded multicore cables (RS Components Ltd, Corby, UK) of 1 m length to prevent electro-magnetic interference from other machinery in the vicinity. The ends of the multicore cables were connected to a Data Logger (Solartron 3530D Orion *delta*, Schlumberger Technologies, Bognor Regis, UK) at the time of testing every tooth.

Preparation for cyclic loading test

Configuration of testing equipment One tooth was used to configure the test system (MTS 830 Elastomer, MTS Systems Corp, Eden Prairie, MN, USA) with a 10-kN load cell attached to the data logger. Tuning of the control system for the test machine was necessary to ensure that the readings were stable and reproducible for the entire experiment. Programmed instruction sets were written for both the test machine and data logger.

Test system (MTS 830 Elastomer) The test machine was programmed to apply the following loading, under force control. A preload of 10 N was applied, followed by a cyclic load between 10 and 110 N at 33.33 Nsec^{-1} for six cycles, then unloading. The range 10–110 N was considered to be within the physiological range (De Boever *et al.* 1978). Data from each test stage were saved as a Microsoft Excel file (Microsoft Corp, Redmond, WA, USA).

Data logger (Solartron 3530D Orion delta) The leads from the rosette gauges were connected to the data logger in a 'quarter-bridge' configuration. Temperature compensation was achieved via the data logger's in-built 'dummy' compensation gauge. The data logger was programmed to excite and record signals received from the strain gauges at a rate of five readings per s per strain gauge. The signals were automatically converted into microstrain ($\mu\varepsilon$) by the data logger and a data file (ASCII format) was created for each test stage.

Table 2 Sequence of irrigation and load testing for each of four controls and the 10 test teeth

Table 2a Sequence of events for positive control 1

Stage	Phase	Treatment
1	Irrigation	Saline (30 min)
	Loading of tooth	
2	Irrigation	5.25% NaOCl (30 min)
	Loading of tooth	
3	Irrigation	5.25% NaOCl (30 min)
	Loading of tooth	
4	Irrigation	5.25% NaOCl (30 min)
	Loading of tooth	
5	Irrigation	Saline (30 min)
	Loading of tooth	

Table 2b Sequence of events for positive control 2

Stage	Phase	Treatment
1	Irrigation	Saline (30 min)
	Loading of tooth	
2	Irrigation	0.5% NaOCl (30 min)
	Loading of tooth	
3	Irrigation	0.5% NaOCl (30 min)
	Loading of tooth	
4	Irrigation	0.5% NaOCl (30 min)
	Loading of tooth	

Table 2c Sequence of events for negative control 1

Stage	Phase	Treatment
1	Irrigation	Saline (30 min)
	Loading of tooth	
2	Irrigation	Saline (30 min)
	Loading of tooth	
3	Irrigation	Saline (30 min)
	Loading of tooth	
4	Irrigation	Saline (30 min)
	Loading of tooth	

Testing of teeth

A specially machined brass receptacle was clamped to the lower member of the MTS machine. Each test tooth was placed in the receptacle and positioned so that the upper member of the MTS machine was aligned vertically over the centre of the access preparation. A holder with a 6.3-mm ball-bearing for applying the load was attached to the actuator. The teeth in the acrylic blocks were securely fastened by means of four brass screws. The tooth was not moved during the entire testing procedure. The 14 teeth from the control and test groups were each subjected to four or five test stages as shown in Table 2 (a–e), each stage creating a separate data file.

Table 2d Sequence of events for negative control 2 – intact enamel

Stage	Phase	Treatment
1	Irrigation	Saline (30 min)
	Loading of tooth	
2	Irrigation	0.5% NaOCl (30 min)
	Loading of tooth	
3	Irrigation	5.25% NaOCl (30 min)
	Loading of tooth	
4	Irrigation	5.25% NaOCl (30 min)
	Loading of tooth	

Table 2e Sequence of events for test group teeth

Stage	Phase	Treatment
1	Irrigation	Initial-saline (30 min)
	Loading of tooth	
2	Irrigation	0.5% NaOCl (30 min)
	Loading of tooth	
3	Irrigation	5.25% NaOCl (30 min)
	Loading of tooth	
4	Irrigation	Final-saline (30 min)
	Loading of tooth	

The standardized 30-minute irrigation regime for each test stage consisted of rubber dam isolation of the access cavity to protect the gauges from the irrigant, followed by an initial 3 mL flush with the test irrigant over 1 min, using a 27-gauge needle attached to a Monoject syringe, the tip being placed within 1 mm of the 'canal length'. The solution was then agitated in the canal for 10 s using a blunted medium-fine finger-spreader. The solution was left for 50 s (total time 2 min). In the next step, 0.5 mL of the irrigant was introduced into the canal over 20 s. The solution in the canal was agitated for 10 s with the same finger-spreader and left for 1.5 min. This was repeated 14 times for a total of 28 min. There was then a final flush with 9 mL of saline over 3 min.

Analysis of data

The rosette gauges yielded three sets of data which were resolved into maximum and minimum principal strains (tensile and compressive strains), using a series of simultaneous equations. As each tooth acted as its own control, a Repeated Measures, single-factor analysis of variance was used to compare the data for each treatment stage. Data were also presented as percentage change in strain (compressive or tensile) using the following calculation:

$$[(\text{Initial measured strain} - \text{measured strain})/\text{Initial measured strain}] \times 100$$

Table 3 Means and standard deviations (SD) of modulus of elasticity (Nm^{-2}) for each group, including the statistical analysis

Groups	*n*	Mean	SD
Saline	37	15.1E + 9	2.1E + 9
0.5% NaOCl	31	14.5E + 9	2.8E + 9
5.25% NaOCl	32	12.9E + 9	2.3E + 9

ANOVA

Source of variation	SS	d.f.	MS	F	*P*-value	F crit
Between groups	8.8E + 19	2	4.4E + 19	7.85	< 0.01	4.83
Within groups	5.4E + 20	97	5.6E + 18			
Total	6.4E + 20	99				

Post hoc comparisons

1. Tukey's HSD procedure 2. Scheffe's procedure

Treatment levels	Error d.f.	Multiplier	*n*	1. HSD	2. Scheffe's
3	97	4.3	31	1.8E + 9	1.8E + 9

Differences between means

	Saline	0.5% NaOCl	5.25% NaOCl
Saline		0.6E + 9	2.3E + 9[a]
0.5% NaOCl			1.7E + 9

[a] Significant.

Table 4 Means and standard deviations (SD) of flexural strength (Nm^{-2}) for each group, including the statistical analysis

Groups	*n*	Mean	SD
Saline	37	17.0E + 7	4.5E + 7
0.5% NaOCl	31	14.8E + 7	4.0E + 7
5.25% NaOCl	32	10.3E + 7	3.0E + 7

ANOVA

Source of variation	SS	d.f.	MS	F	*P*-value	F crit
Between groups	7.9E + 16	2	4.0E + 16	26.13	< 0.01	4.83
Within groups	1.5E + 17	97	1.5E + 15			
Total	2.3E + 17	99				

Post hoc comparisons

1. Tukey's HSD procedure 2. Scheffe's procedure

Treatment levels	Error d.f.	Multiplier	*n*	1. HSD	2. Scheffe's
3	97	4.3	31	0.3E + 8	2.9E + 8

Differences between means

	Saline	0.5% NaOCl	5.25% NaOCl
Saline		0.2E + 8	0.7E + 8[a]
0.5% NaOCl			0.5E + 8[a]

[a] Significant.

Results

Modulus of elasticity and flexural strength of dentine bars

The means and standard deviations of the modulus of elasticity (Table 3) and flexural strength (Table 4) are presented in the respective tables. The one-factor, independent measure ANOVA showed that there were significant differences ($P < 0.01$) between the three groups for both the modulus of elasticity and flexural strength.

Post hoc comparisons using Tukey's HSD (Tukey 1977) and Schéffe's procedures (Schéffe 1959) showed

Table 5 Means and standard deviations (SD) of principle maximum (tensile) and minimum (compressive) strains for each treatment stage in the positive and negative control groups

Table 5a Positive control 1

	Tensile strains		Compressive strains	
	Mean	SD	Mean	SD
Stage 1 (Saline)	157.3	2.77	–132.7	0.92
Stage 2 (5.25%)	209.0	2.18	–212.6	2.06
Stage 3 (5.25%)	239.4	2.68	–257.1	1.78
Stage 4 (5.25%)	240.0	4.03	–242.8	3.17
Stage 5 (Saline)	246.4	2.05	258.2	4.04

Table 5b Positive control 2

	Tensile strains		Compressive strains	
	Mean	SD	Mean	SD
Stage 1 (Saline)	364.7	5.58	−444.3	3.35
Stage 2 (0.5%)	359.9	3.06	−449.2	3.36
Stage 3 (0.5%)	373.4	4.43	−435.5	2.12
Stage 4 (0.5%)	385.7	3.39	−432.6	2.94

Table 5c Negative control 1

	Tensile strains		Compressive strains	
	Mean	SD	Mean	SD
Stage 1 (Saline)	75.7	1.77	–70.2	1.81
Stage 2 (Saline)	77.9	0.81	–68.0	0.89
Stage 3 (Saline)	77.4	1.24	–71.4	1.79
Stage 4 (Saline)	79.4	1.18	–68.9	1.10

Table 5d Negative control 2 – intact enamel

	Tensile strains		Compressive strains	
	Mean	SD	Mean	SD
Stage 1 (Saline)	47.0	0.61	–65.7	0.21
Stage 2 (0.5%)	52.4	1.64	–68.5	0.71
Stage 3 (5.25%)	42.2	0.71	–57.2	1.65
Stage 4 (5.25%)	54.4	1.01	–57.8	2.29

that the mean elastic modulus value for the 5.25% sodium hypochlorite group (12.9 GNm^{-2}) was significantly less than that of the saline group (15.1 GNm^{-2}). Differences between the 0.5% sodium hypochlorite group (14.6 GNm^{-2}) and the saline and 5.25% sodium hypochlorite groups were not found to be statistically significant (Table 3).

Post hoc comparisons showed a statistically significant difference in flexural strength between the saline group (170.2 MNm^{-2}) and the 5.25% sodium hypochlorite group (103.0 MNm^{-2}). The difference between the 0.5% sodium hypochlorite group (148.3 MNm^{-2}) and the 5.25% sodium hypochlorite group was also significant (Table 4).

Cyclic nondestructive loading tests of teeth

For each treatment stage, the raw strain data from the three elements of the rosette gauge were plotted against time. The transformed principal maximum (tensile) and principal minimum (compressive) strains calculated from the raw strains for each treatment stage were also plotted against time.

Positive control 1 – 5.25% sodium hypochlorite (Table 5a) – There was a marked increase in both tensile and compressive strain from baseline values with 5.25% sodium hypochlorite treatment. The increase in tensile strain plateaued at treatment stage 3 (after 1 h of 5.25% NaOCl).

Beyond stage 3, there was minimal increase in strain. A similar pattern was observed for compressive strain.

Positive control 2 – 0.5% sodium hypochlorite (Table 5b) – The tensile and compressive strain values appeared to remain relatively unchanged from baseline.

Negative control 1 Saline irrigation only (Table 5c) – The tensile and compressive strain remained relatively low and unchanged from baseline even after four irrigation stages using saline.

Negative control 2 Tooth with intact enamel margins (Table 5d) – The tensile and compressive strain remained relatively unchanged from baseline even after two irrigation stages using 5.25% sodium hypochlorite. The strain values were also the lowest of all teeth tested.

Test group The means and standard deviations of the peak strains were calculated for every treatment stage for each test tooth. The means of the tensile strain values for all the treatment stages for the 10 test teeth are presented in Table 6 and the percentage change in strain in Figure 1 and those of the compressive strains in Table 7 and Figure 2.

There appears to be an increase in mean tensile strain values from the initial-saline baseline (15.7% change) after irrigation with high concentration sodium hypochlorite (Fig. 1). There was minimal change in strain values between 5.25% sodium hypochlorite and final-saline (1.8% change) and between initial-saline and 0.5% sodium hypochlorite (2.3% change).

Table 6 Mean principal maximum (tensile) strains [$\hat{I}_P$] against treatment for all test teeth

	Treatment stage			
Test tooth	1 Saline	2 0.5% NaOCl	3 5.25% NaOCl	4 Saline
1	226.0	216.6	352.2	346.8
2	353.9	363.9	365.0	379.9
3	123.9	133.6	216.6	226.0
4	360.2	363.5	338.1	335.3
5	209.0	209.2	240.0	227.2
6	146.4	148.2	197.3	208.9
7	433.0	435.4	439.0	434.7
8	274.2	263.7	271.0	272.6
9	412.9	457.9	450.6	499.1
10	171.2	181.6	266.3	262.2
Mean	271.1	277.4	313.6	319.3

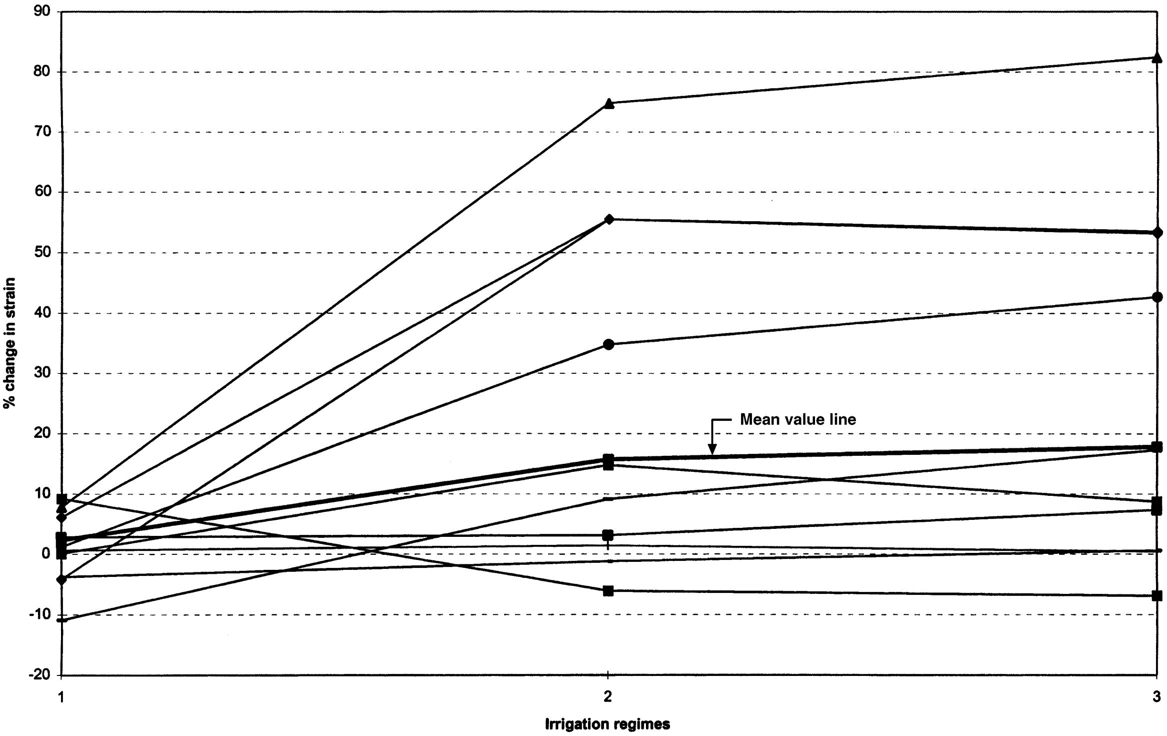

Figure 1 Graph depicting the percentage change in tensile strains for each of the 10 test teeth as well as mean values at different treatment stages (bold line). Under irrigation regimes: 1 = % change between initial-saline and 0.5% NaOCl; 2 = % change between initial-saline and 5.25% NaOCl; 3 = % change between initial-saline and final-saline.

Table 7 Mean principal minimum (compressive) strains $[\hat{I}_Q]$ against treatment for all test teeth

	Treatment stage			
Test tooth	1 Saline	2 0.5% NaOCl	3 5.25% NaOCl	4 Saline
1	−158.2	−162.6	−255.1	−254.8
2	−297.0	−318.3	−368.2	−371.4
3	−87.2	−94.2	−162.6	−158.2
4	−204.0	−208.8	−272.1	−277.5
5	−212.6	−224.0	−242.8	−247.1
6	−180.8	−184.5	−212.0	−202.8
7	−250.3	−248.9	−380.6	−379.5
8	−252.8	−274.5	−306.6	−306.5
9	−389.8	−401.5	−461.2	−465.3
10	−104.7	−107.9	−191.3	−193.1
Mean	−213.7	−222.5	−285.2	−285.6

Statistical analysis using the Repeated Measures ANOVA revealed that there were significant differences ($P < 0.01$) between the four treatment stages (Table 8). *Post hoc* comparisons with Tukey's HSD procedure revealed that the source of differences between the means were the initial-saline and the final-saline measurements.

There was an increase in mean compressive strain values from baseline (33.5% change) after irrigation with 5.25% sodium hypochlorite (Fig. 2). There was minimal change in strain between initial-saline and low concentration sodium hypochlorite (4.1% change) and between 5.25% sodium hypochlorite and final-saline (0.14% change). The Repeated Measures ANOVA revealed significant differences ($P < 0.01$) between the four treatment stages (Table 9). *Post hoc* comparisons revealed that the differences were between: (i) initial-saline and 5.25% sodium hypochlorite, (ii) initial-saline and final-saline, (iii) 0.5% sodium hypochlorite and 5.25% sodium hypochlorite and (iv) 0.5% sodium hypochlorite and final-saline. There were no statistically significant differences between (i) initial-saline and 0.5% sodium hypochlorite and (ii) 5.25% sodium hypochlorite and final-saline.

Discussion

This study was divided into two quite separate but related parts. The first part evaluated the effect of sodium hypochlorite on dentine (bars) at the 'local' level and the second part allowed observation of the effect of such 'local' dentine weakening on strain in 'whole' teeth.

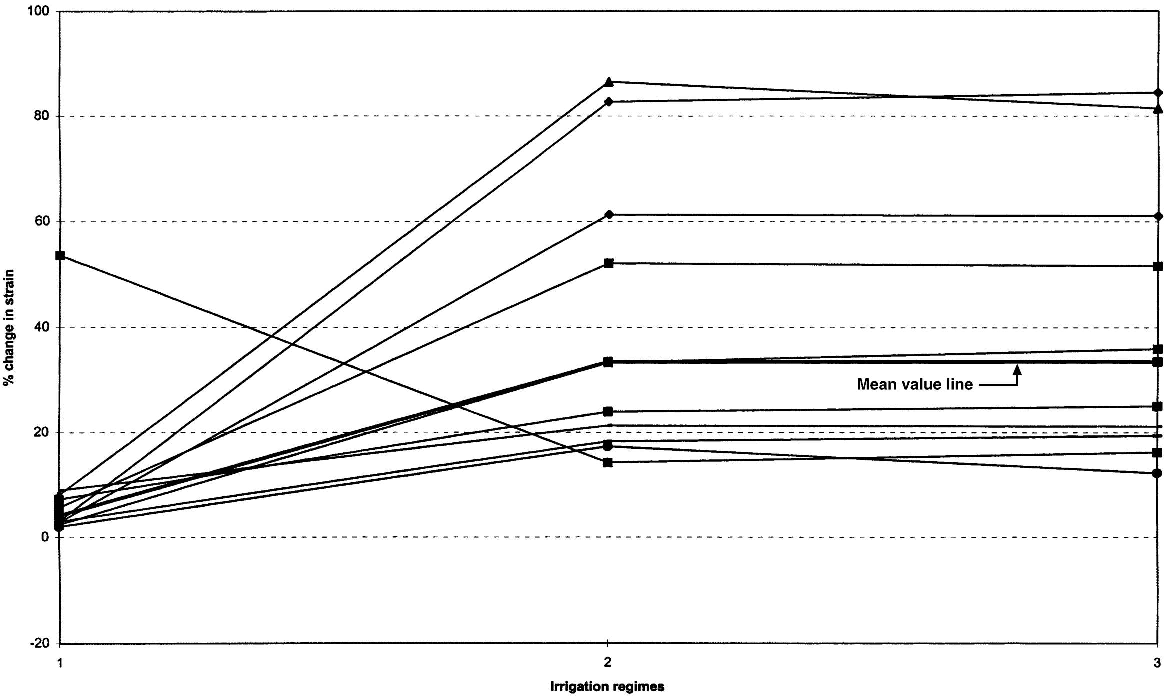

Figure 2 Graph depicting the percentage change in compressive strains for each of 10 test teeth as well as mean values at different treatment stages (bold line). Under irrigation regimes: 1 = % change between initial-saline and 0.5% NaOCl; 2 = % change between initial-saline and 5.25% NaOCl; 3 = % change between initial-saline and final-saline.

Table 8 Means, standard deviations (SD) and statistical analyses of tensile strains for the different treatment stages

Groups	*n*	Mean (mL)	Variance	SD
Initial-saline	10	271.1	12685.9	112.6
0.5% NaOCl	10	277.4	14173.5	119.0
5.25% NaOCl	10	313.6	7956.9	89.2
Final-saline	10	319.3	9460.6	97.3

ANOVA (Repeated measures design, one-factor)

Source of variation	SS	d.f.	MS	F	*P*-value	F crit
Between conditions	18190.2	3	6063.4	6.99	< 0.01	4.60
Within conditions	398491.0	36				
Between subjects	375075.3	9				
Error	23415.7	27	867.3			
Total	416681.2	39				

Post hoc comparisons

Tukey's HSD procedure

Treatment levels	Error d.f.	Multiplier (4,27)	*n*	HSD
4	27	4.9	10	45.2

Differences between means

	0.5% NaOCl	5.25% NaOCl	Final-saline
Initial-saline	6.3	42.6	48.2^{a}
0.5% NaOCl		36.3	41.9
5.25% NaOCl			5.7

[a] Significant.

Table 9 Means, standard deviations (SD) and statistical analyses of compressive strains for the different treatment stages

Groups	*n*	Mean (mL)	Variance	SD
Initial-saline	10	–213.7	8105.6	90.0
0.5% NaOCl	10	–222.5	8822.3	94.0
5.25% NaOCl	10	–285.2	8832.3	94.0
Final-saline	10	–285.6	9225.4	96.0

ANOVA (Repeated measures design – one-factor)

Source of variation	SS	d.f.	MS	F	*P*-value	F crit
Between conditions	45694.0	3	15231.3	43.2	< 0.01	4.60
Within conditions	314871.0	36				
Between subjects	305345.5	9				
Error	9525.3	27	352.8			
Total	360564.8	39				

Post hoc comparisons

Tukey's HSD procedure

Treatment levels	Error d.f.	Multiplier (4,27)	*n*	HSD
4	27	4.9	10	28.8

Differences between means

	0.5% NaOCl	5.25% NaOCl	Final-saline
Initial-saline	8.8	71.5^{a}	71.9^{a}
0.5% NaOCl		62.7^{a}	63.1^{a}
5.25% NaOCl			0.4

[a] Significant.

The concentrations of sodium hypochlorite solutions used in this study were verified by iodometric titration (British Pharmacopoeia 1973) and were used within 48 h of preparation. The concentrations selected (0.5%, 5.25%) represent extreme concentrations commonly reported as used clinically in the literature. Saline was used as a standard neutral solution, which is not considered to be different in its effect on dentine from that exerted by distilled or tap water (Waters 1980).

The dentine bars used in the three-point bend test behaved as brittle materials during testing. Thus, it was important that the cross-head speed of the load testing machine was set as low as possible so as to record the load-displacement curves accurately. Rates of between 0.1 mm to 1.0 mm min^{-1} (Sedgley & Messer 1992, Jameson *et al.* 1993) have been reported. The Hounsfield H25K machine used in this study was set at its slowest cross-head speed of 0.5 mm min^{-1}. The ratio of the span between the supports and the depth of the dentine bars was maintained at 16 (tolerance, +4 or −2) to ensure that the formulae used to calculate the flexural strength and modulus of elasticity were valid and that correction factors would not be needed (American Society for Testing & Materials 1989).

The dentine bars which had been immersed in 5.25% sodium hypochlorite appeared bleached and 'chalky' in texture. This appearance translated into a significant decrease in flexural strength and rigidity compared to the other two groups. The change in physical properties could be explained by the loss of the organic matrix within the dentine. Work on the physical properties of deproteinized bone by Wright *et al.* (1981) has revealed a decrease in ultimate stress values to 55% of that of normal bone. The elastic modulus of the deproteinized bone was also decreased to 55% of the control value. They concluded that the change in mechanical behaviour of the altered bone must result from dissolution of the collagen or the collagen-mineral bond. It would seem reasonable to speculate that the same occurs in dentine.

In order to gauge the effect of sodium hypochlorite-weakened dentine on the 'whole' tooth, nondestructive occlusal loading tests were used. It was decided to remove the remaining enamel from the coronal 4 mm of the prepared teeth as results from negative control tooth 2 (intact enamel) showed very low strain readings. The inference was that the band of enamel around the cervical dentine had a significant 'reinforcing' effect on the stiffness of the tooth. This confirms work by Meredith (1992), in which he found strain values on the buccal surface of molars to be only in the region of 100 $\mu\varepsilon$ when subjected to 100 N loads applied at the central fossae of the teeth. Despite removal of the enamel, there were wide individual variations in strain between tooth samples to the standard cyclic-load regimen in the present study (Tables 6 and 7). In some teeth, there was minimal change, in a small number the reverse, and in others quite large. This may have been partly due to differences in the thickness of the remaining dentine between the canal and the axial surface of the tooth in the area where the rosette gauges were bonded. Measurements of the thickness of dentine for all 14 teeth revealed that the differences were minor (within 0.12 mm between the thinnest and thickest specimen, measured at the flattened dentine surfaces). Therefore, overall architecture or shape and distribution of the dentine is probably quite important. The same type of teeth were used to reduce the effect of variation. The occlusal surface of the tooth was flattened for ease of alignment of the upper component of the MTS machine during repeated loading and irrigation phases. The amount of remaining coronal tooth structure simulated a moderate-severely broken down tooth, frequently encountered in endodontic practice. The results confirmed not only the value of residual dentine but also that of enamel.

A standard canal preparation as described by Roane (1991) was used to keep the dimensions of the root canal preparation as uniform as possible. A size 80 control zone ensured that the tip of the 27-gauge irrigating needle would reach the apical end of the preparation and lie passively within it, as recommended by Ingle & Bakland (1994). This allowed for good exchange of irrigant within the main canal system and helped ensure that lack of irrigant penetration was not a significant variable.

It was important that the actual change in strain of the tooth due to load was transferred without attenuation to the rosette strain gauge. A thin layer of cyanoacrylate, with an elastic modulus as close as possible to that of tooth structure, was used for this purpose. Heat generation during excitation of the strain gauges which may affect the readings was minimized in two ways. First, a dummy gauge was used which was subjected to identical environmental conditions apart from strain. Secondly, the data logger used intermittent gauge excitation, limiting gauge heating to a minimum.

The cervical margin was chosen as the site for bonding of the rosette gauge because it was found by Meredith (1992), using the stress pattern analysis by thermoelastic emission (SPATE) techniques, to be a site of stress concentration during loading of posterior teeth. This has been confirmed using finite element analysis (FEA) methods (Yettram *et al.* 1976, Goel *et al.* 1991) and by clinical observations (Lee & Eakle 1984, Bream *et al.* 1992).

The long recovery period between recordings, i.e. during the irrigation stages, minimized the influence of hysteresis on the strain readings. The advantage of the MTS machine was that it could record the change in displacement during each cycle of the test sequence to confirm that there was elastic recovery during each unloading cycle. There was no evidence of increasing strain caused by slow crack propagation from the positive controls (Table 5).

Analysis of the transformed strains revealed that there were both tensile (positive values) and compressive (negative values) components in the principal strains. The magnitude of change in compressive strain following irrigation appeared to be greater than tensile strain. This was to be expected as the tooth was loaded in compression. The direction of any tensile strains would be perpendicular to the long axis of the load and thus it was expected not to record significant changes in these strains.

It was interesting that the increase in strain value for positive control tooth 1 plateaued after two, 30-minute periods of irrigation with 5.25% sodium hypochlorite. It is possible that the organic portion of the dentine had been removed to such an extent during the first two irrigation stages that any further removal of organic material with a further 30-minute period of 5.25% sodium hypochlorite would not significantly affect the residual strength of the tooth. Alternatively, it may be that some sort of chemical equilibrium prevented further dissolution. It is interesting to speculate on the effect of higher concentrations of sodium hypochlorite being recommended by some clinicians.

Although sodium hypochlorite remains the irrigant of choice in endodontics, this study shows that the 5.25% concentration has a negative effect on the properties of teeth. The study also found a decrease in flexural strength of the dentine bars subjected to high concentration sodium hypochlorite. These results indicate yet another factor that may weaken root-treated teeth, namely sodium hypochlorite irrigation. The change in stiffness of a tooth after root canal treatment (Reeh *et al.* 1989) is clinically relevant as it may predispose the tooth to fracture. The decrease in flexural strength is also clinically relevant as it indicates that far less force is required for the cohesive bonds within dentine to fail. In this study the increase in strain after high concentration sodium hypochlorite irrigation was 15.9% for tensile strain and 33.5% for compressive strain. This change, although statistically very significant, does not automatically indicate an increased risk of fracture due to repeated loading. The results should also be viewed in the context of the irrigation regime which is the worst case scenario and unlikely to match that of many clinicians. It was necessary to select an aggressive regimen in this study in order that the effect was guaranteed.

Studies have shown that lower concentrations of sodium hypochlorite are effective in cleaning and disinfecting root canal systems (Trépagnier *et al.* 1977, Byström & Sundqvist 1985, Baumgartner & Cuenin 1992). It would thus be prudent to select a suitable concentration which has minimal effects on the physical properties of the tooth whilst achieving the desired debridement effect.

The study raises many further questions, including the effect of other concentrations of sodium hypochlorite and their duration of use. The strain gauges indicate tooth deformation at only a small representative point on the tooth (Meredith 1992). It would be valuable to map strain over a larger area using full-field strain measuring techniques. The precise mechanism of mineral depletion of dentine by sodium hypochlorite is also worth examining.

In conclusion, this study has demonstrated that 5.25% sodium hypochlorite compared to saline significantly ($P < 0.01$) reduces the flexural strength and elastic modulus of dentine. This effect is noticeable in a 'whole' tooth (without enamel) as increased strain under standard cyclic nondestructive loading.

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