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The bactericidal effects of dental ultrasound on *Actinobacillus actinomycetemcomitans* and *Porphyromonas gingivalis*

An in vitro investigation

R. O'Leary[1], A. M. Sved[2], E. H. Davies[3], T. G. Leighton[4], M. Wilson[5] and J. B. Kieser[6]

[1]Department of Periodontology, Eastman Dental Institute, London, UK, [2]Private practice, Sydney, Australia, [3]Department of Biomaterials, Eastman Dental Institute, London, UK, [4]Institute of Sound and Vibration Research, University of Southampton, UK, [5]Department of Microbiology, Eastman Dental Institute, London UK, [6]Department of Continuining Education, Eastman Dental Institute, London UK

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Abstract. This study investigated the possible bactericidal acoustic effects of the dental ultrasonic scaler. *Actinobacillus actinomycetemcomitans* and *Porphyromonas gingivalis* suspensions, were subjected to the vibrations of a Cavitron P1 insert for 2.5 and 5.0 min in an acoustically-simulated pocket model and the survivors enumerated. The extent of any cavitation occurring within the pocket model to which the statistically significant bactericidal activity observed might be attributed, was determined by 'sonoluminescence', which was then investigated by photomultiplication techniques. However, these failed to detect any sonoluminescence within the pocket space and, moreover, the necessary deflection of the water coolant away from the insert tip, to avoid flooding of the experimental pocket, proved to result in temperatures of 47.6°C and 52.3°C at the respective time intervals, and thereby constituted an alternative possible bactericidal mechanism. Examination of the effects of such temperature changes on the target bacteria then revealed statistically significant differences in the viable counts of both microorganisms after 5.0-min periods, and as such were comparable to those previously detected in relation to the pocket model. Whilst it must be presumed that the bacteriolytic effect observed in the main investigation was due to the incidental temperature changes, in the absence of acoustic cavitation the influence of any associated acoustic microstreaming cannot be discounted. Further investigations to assess the bactericidal potential of acoustic phenomena using a modified experimental to exclude any hyperthermic effects are therefore necessary.

Key words: *A actinomycetemcomitans*; *P gingivalis*; ultrasonic scalers; cavitation; sonoluminescence

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Ultrasonic scaling devices are now widely used in clinical practice and have been shown to be as effective as traditional hand instrumentation in the removal of subgingival plaque (Thornton & Garnick 1982, Breininger et al. 1987, Leon & Vogel 1987, Oosterwaal et al. 1987), and calculus (Torfason et al. 1979, Hunter et al. 1984, Gellin et al. 1986, Breininger et al. 1987) and in prospective clinical healing responses (Torfason et al. 1979, Badersten et al. 1981, 1984, Leon & Vogel 1987, Oosterwaal et al. 1987, Biagini et al. 1988). The trend in favour of ultrasonic devices that has evolved as a result of ease of use and reduced clinical time (Torfason et al. 1979, Badersten et al. 1981, Oosterwaal et al. 1987), has been strengthened by the superior clinical and microbiological responses following ultrasonic scaling in Class II and Class III furcations (Leon & Vogel 1987). This has been attributed to the improved access to the furcal walls and may be enhanced by the use of specially designed (Oda & Ishikawa 1989) or modified tips (Checchi et al. 1991). Recently developed ball ended ultrasonic tips have also resulted in more efficient removal of root surface deposits than both conventional ultrasonic tips or hand scalers/curets (Takacs et al. 1993).

Subgingival instrumentation with both hand (Listgarten et al. 1978, Mousquès et al. 1980, Magnusson et al.

1984) and ultrasonic (Oosterwaal et al. 1987, Loos et al. 1988, Baehni et al. 1992) scalers has also been shown to lead to profound shifts in the subgingival microflora from a predominantly Gram-negative composition to one which is Gram-positive and facultative. Similarly, the techniques are equally effective in reducing counts of black pigmented species (Renvert et al. 1990a, Oosterwaal et al. 1987, Loos et al. 1988) and spirochaetes (Listgarten et al. 1978, Mousquès et al. 1980, Magnusson et al. 1984, Thilo & Baehni 1987, Baehni et al. 1992) although Renvert et al. (1990a, b) revealed that neither method affected significantly in vivo levels of *Actinobacillus actinomycetemcomitans*.

A further potential advantage of ultrasonic instrumentation is the facility for the supplementary removal of root surface plaque (Walmsley et al. 1988) and/or surface constituents by cavitational activity (Walmsley et al. 1990, Byrne 1990). Transient cavitation describes the violent oscillations of air bubbles in a liquid following ultrasonic exposure and their subsequent implosion, which generates shock waves throughout the liquid medium (Williams 1983). The detection of sonoluminescence, a low level light emitted as a result of the recombination of the free radicals generated within the compressed gas during bubble collapse, indicates the presence of transient cavitation in vitro (Walton & Reynolds 1984). The reported destructive effect of ultrasonic waves on microorganisms (Hamre 1948, Leadbetter & Holt 1974) has been variously attributed to cavitation (Davies 1959), the associated acoustic streaming (Hughes & Nyborg 1962) and to chemical activity, i.e., the production of free radicals, although this has not been demonstrated to be a significant factor experimentally (Thacker 1973).

The sensitivity of Gram-negative organisms and spirochaetes to ultrasound (Robrish et al. 1976, Thilo & Baehni 1987, Loesche & Laughon 1982) is of particular interest periodontally. The relative resistance of Gram-positive bacteria, spores and viruses (Thacker 1973, Hughes 1961), can be explained by differences in the size, rigidity and strength of their cell walls (Thacker 1973), such that Thilo & Baehni (1987) have postulated that the lack of such features in Gram-negative species render them susceptible to the shearing forces of ultrasonic acoustic phenomena (Robrish et al. 1976), and thereby contribute to the post-ultrasonic instrumentational changes in the microbial composition of dental plaque, (Oosterwaal et al. 1987, Loos et al. 1988, Baehni et al. 1992). This contention is supported by the in vitro bactericidal effects of dental ultrasonic activation on spirochaetes (Thilo & Baehni 1987), and more recently, on suspensions of *Actinobacillus actinomycetemcomitans* (Aa) and *Porphyromonas gingivalis* (Pg) (Sved 1991). However in the latter investigation, the observed sonoluminescence appeared concurrently with temperature rises which may have contributed to the bacteriolytic effects. In addition, the volume of the experimental tubes containing the bacterial suspensions far exceeded that of any clinical pocket space.

The aim of this study was therefore to investigate the acoustic and any accompanying thermal effects of ultrasonic activation upon pure cultures of periodontally associated microorganisms, in an acoustically simulated periodontal pocket model.

Material and Methods

Acoustic impedance and the development of an acoustically-simulated pocket

Acoustic waves are reflected or refracted at the interface between different media in a fashion similar to light waves (Daly & Wheeler 1971). The speed of transmission of light waves through a medium is determined by its refractive index, and in the case of mechanical waves by *"Specific Acoustic Impedance"* (Z), expressed as kg/m^2s representing the ratio of the amplitude of the reflected and refracted wave. Specific acoustic impedance is the product of the density (ρ) of a material and the velocity (V) of sound through it. If an acoustic wave, travelling in one medium, impinges upon the boundary between the 1st medium and a 2nd one, in general, the greater the difference in acoustic impedance between the 2 media, the larger the portion of the wave that is reflected back to the 1st medium and the smaller the proportion of the wave that is transmitted into the 2nd medium.

The acoustically simulated pocket model was constructed from materials with acoustic impedance (Z) values equivalent to those of pocket wall tissue and dentine. Thus silicone rubber was selected to simulate the soft tissue pocket wall as its impedance value of $Z=1.34\times10^6$ kg/m^2s is similar to that of soft tissues ($Z=1.55\times10^6$ kg/m^2) (Lees 1971). Furthermore, being available in tubular form, it could be readily adapted to the model base. Dentine, with an impedance value of 8×10^6 kg/m^2s (Lees 1971), was in turn reproduced by a blend of "Araldite" resin and tungsten powder ($Z=8.11\times10^6$ kg/m^2s) (Selfridge 1985), prepared in proportions of 8 g Araldite resin: 1.6 g hardener: 1.92 g tungsten powder. The constituents were mixed thoroughly, placed in an incubator at 37°C, and as the mixture became viscous, poured into silicone rubber (Xantopren, Bayer Dental, Berkshire) 'tooth' moulds. These had been prepared from a replica of the experimental model hard tissue component, milled from a polycarbonate block to produce a root length of 10 mm and a pocket width of 2 mm (Fig. 1). The resin/tungsten mixture was left to harden in an incubator (a. 16 h). Following the appropriate trimming of excess material, the simulated pocket space with a volume of 0.3 ml was created by fitting the silicone rubber tubing over the tooth model (Fig. 1). However despite the tight fit between the tubing and the model base, a complete seal could not be ensured, necessitating additional fixation with "Locktite" cyanoacrylate adhesive (Locktite UK, Hertfordshire).

Ultrasonic activation

A Dentsply Cavitron 2002 ultrasonic scaling unit (Cavitron IIA, Dentsply Corp., York, PA.) with a 'P' type (P-1) insert was used. The inevitable dilution of the microbial suspensions from the Cavitron spray and loss thereof through overflow, was overcome by bending the metal water coolant tube away from the long axis of the insert and then fitting a flexible 30 cm length plastic tube to divert the water into an adjacent container.

The periodontal pathogens, *Actino-*

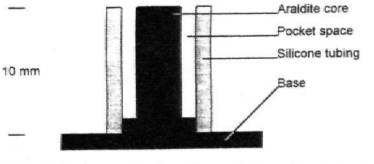

Fig. 1. Schematic drawing of the simulated pocket model.

bacillus actinomycetemcomitans Y4 and *Porphyromonas gingivalis* W50 were cultured on Wilkins – Chalgrens (W-C) anaerobe agar at 37°C in a Scientific Compact Work Station (Don Witley, U.K.) for 3 days. The organisms were then harvested from W-C plates and transferred into a sterile bottle containing 3ml sterile horse serum. Following vortexing for 30 s, 0.2 ml was placed in the experimental pocket reservoir immediately prior to ultrasonic activation. 5 test and 5 non-activated control samples of each bacterial species were used in each experiment, which was then repeated.

The insert was held manually in a vertical position within the pocket space and the entire pocket space evenly circumscribed during the experimental 2.5- or 5.0-min periods of activation, at a medium power setting. A similar procedure was carried out on the control samples but without ultrasonic activation. The experimental technique of circumscribing the whole pocket space ensured an even exposure of all parts of the microbial suspension to the sonication activity. On completion, the insert was removed and each 0.2 ml bacterial suspension transferred to sterile bottles containing 0.8 ml W-C broth and vortexed for 30 s. Serial dilutions (10^{-1}–10^{-5}) were carried out in 0.9 ml W-C broth, plated out on W-C agar plates. Pg was incubated anaerobically for 5–7 days, whilst Aa was incubated in candle jars for increased CO_2 production. Viable counts were performed with the aid of a colony counter.

The insert was autoclaved following each group of ten experimental sonication/control sequences of a single bacterial species. However a compromise of disinfection of the pocket models, by immersing them in a water bath at 60°C for 10 min, was resorted to as the Locktite adhesive proved unstable at higher temperatures.

Sonoluminescence analysis

The scaler insert (P1) was fixed into position within the pocket space (containing 0.2 ml of water) of the simulated pocket model in a light-tight box, approximately 4 cm above the (1 cm diameter) photocathode of a vertically directed photomultiplier (EMI 9893B/74044). The pocket model was fixed at an angle using plasticine (Fig. 2). The photomultiplier was supplied with a 2000V power supply (Brandenburg 475R) and its output

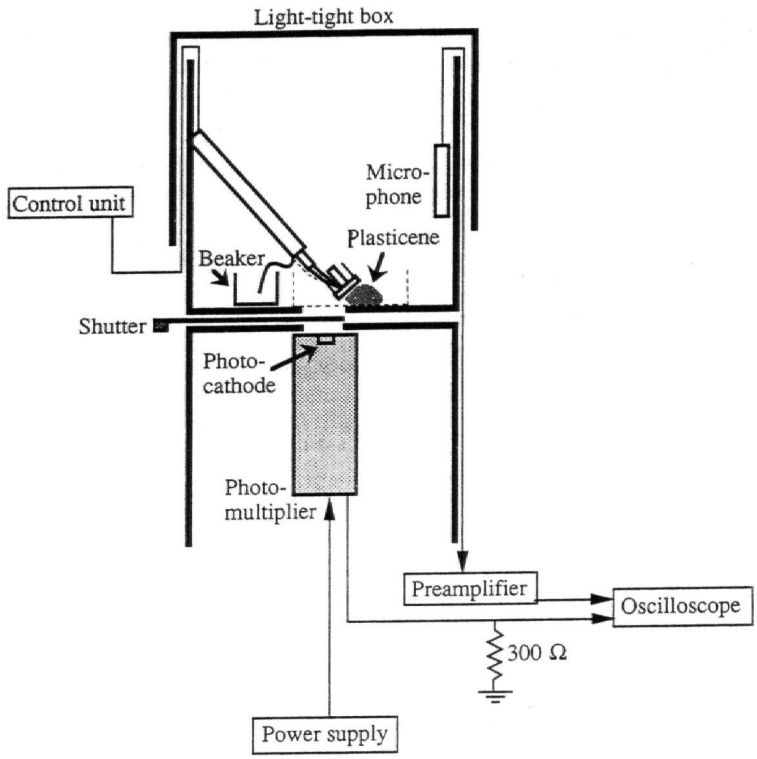

Fig. 2. Diagramatic representatoin of the sonoluminescence experimental set-up. Broken lines show alternative positioning of water spray directed into beaker and over insert tip during irrigation component of the investigation.

passed across a 300 resistor to generate the voltage signal for the input to a digital oscilloscope (Le Croy 9314L). The experimental set up was based on that described by Byrne et al. (1991). Sampling was carried out at 10 megasamples/s, triggered by the audible acoustic output from scaler activation, as detected by the microphone and preamplifier (Bruel Kjaer 2609). Light output resulted in the production of photon pulses which were then recorded on computer and subsequently printed out. The sampling rate was consistent with the detection of the pulses, which represented optical emissions, since the duration of a single event at the input to the oscilloscope was roughly 0.15–0.2 ms. Each sequence was repeated 3×.

Experimentation to detect sonoluminescence was first carried out with the water coolant deflected into an adjacent water beaker, and then with the water spray directed conventionally over the insert tip (Fig. 2). In the latter instance, the pocket model was positioned within a large beaker which allowed the water to collect. Recordings were made immediately following activation and after 5- and 10-s sonication, each sequence being repeated 3×.

Temperature changes following ultrasonic activation

The temperature rises during sonication were recorded with a digital thermometer (STK No 610-067 R.S. (Corby, UK)) within 0.2 ml samples of horse serum in the acoustically simulated pocket at 0.5, 1.0, 2.0, 2.5, 3.5 and 5.0-min time-periods. This was repeated a further three times and the average temperatures computed. The values of 47.6°C (44.5°C→50.9°C) and 52.3°C (51.1°C→53.7°C) at 2.5 min and 5.0 min, respectively, were then used for the subsequent experimental sequence with respect to viable bacterial cell counts.

Temperature effects on bacterial viability

The pattern of temperature rises occurring in the experimental model was re-

Table 1. Effect of sonication on the viability of suspensions of *Aa* and *Pg* in a simulated pocket model (critical value of $t_{(0.05,8)}$=2.31)

Organism	Time (min)	Survivors (cfu) test: geometric mean (range)	Survivors (cfu) control: geometric mean (range)	*t*-values (8 d. f.)	95 % CI ratio (test versus control)	Significance
Aa Experiment 1	2.5	9.82×10^7 (9.0×10^7–11.4×10^7)	6.92×10^7 (4.1×10^7–10.0×10^7)	2.07	0.96–2.1	N/S
Experiment 2		0.06×10^4 (0–0.31×10^4)	1.06×10^4 (0.34×10^4–2.95×10^4)	1.86	0.49–7.3×10^2	N/S
Aa Experiment 1	5.0	5.49×10^2 (0–4.9×10^4)	5.37×10^5 (1.67×10^5–8.6×10^5)	2.64	4.79–3.98×10^5	$P<0.01$
Experiment 2		2.64 (0–1.3×10^2)	5.13×10^3 (4.05×10^3–6.3×10^3)	7.8	6.02×10^2–6.31×10^3	$P<0.001$
Pg Experiment 1	2.5	1.45×10^3 (0–1.25×10^6)	2.0×10^8 (1.86×10^8–2.0×10^8)	27.41	2.62×10^{34}–4.81×10^{40}	$P<0.001$
Experiment 2		1.74×10^3 (0–5.5×10^5)	4.79×10^7 (0.5×10^7–19.5×10^7)	24.74	4.34×10^{30}–8.82×10^{36}	$P<0.001$
Pg Experiment 1	5.0	0 (0–0)	1.55×10^8 (0.65×10^8–2.0×10^8)	86.67	9.36×10^7–2.56×10^8	$P<0.001$
Experiment 2		0 (0–0)	1.95×10^7 (0.14×10^7–9.55×10^7)	20.83	3.01×10^6–1.25×10^8	$P<0.001$

produced in a water bath. A total of 12 aliquots (0.2 ml) of horse serum suspensions of each of Aa and Pg were placed in individual test tubes. 8 tubes were then positioned in the water bath and half heated to 47°C over 2.5 min and the others to 52°C over a 5-min period. The remaining four test tubes were retained as unheated controls. This sequence was then repeated, providing a total of 16 experimental and 8 control samples for each organism. Finally, each of the 48 bacterial suspensions were transferred to individual sterile bottles containing 0.8 ml. W-C broth, vortexed for 30 s, serially diluted 10^{-1} to 10^{-5}, plated out, incubated and viable counts performed.

The possibility of localized heating about the instrument tip influencing the findings, was also considered. Thus during subgingival root surface instrumentation, that part of the instrument located subgingivally would be shielded from the direct coolant effect of the water spray by the overlying soft tissues. Although the temperature rise expected in the immediate vicinity of these submerged tips would obviously be countered by heat conduction towards the cooled supragingival segment of the instrument, any residual heating might still have some bactericidal effect. The temperature at the instrument tip within the experimental model was therefore measured with a thermocouple placed at the base of the 10 mm pocket space adjacent to the sonicating tip, operating normally with optimal water coolant and also as a control with the water deflected, when the model space was first filled with tap water at room temperature. This was repeated three times for sonication periods of 2.5 and 5.0 min and the average temperatures computed.

Table 2. Survival of *Aa* and *Pg* at various temperatures

Organism	Temp (°C)	Time (min)	Survivors (cfu) test: geometric mean (range)	Survivors (cfu) control: geometric mean (range)	*t*-values (6 d. f.)	95% CI ratio (test v control)	Significance
Aa Experiment 1	49	2.5	2.48×10^4 (2.15×10^4–3.05×10^4)	2.4×10^4 (1.22×10^4–3.9×10^4)	0.13	0.55–1.95	N/S
Experiment 2	49		4.37×10^5 (5.6×10^4–6.15×10^5)	8.9×10^5 (8.65×10^5–9.2×10^5)	3.10	1.16–3.44	$P<0.05$
Aa Experiment 1	52	5.0	2.44×10^3 (1.25×10^3–4.0×10^3)	2.39×10^4 (1.22×10^4–3.9×10^4)	6.52	4.13–23.13	$P<0.001$
Experiment 2	52		5.62×10^2 (0–1.02×10^4)	8.86×10^5 (8.65×10^5–9.2×10^5)	3.19	8.12–3.01×10^5	$P<0.05$
Pg Experiment 1	47	2.5	8.13×10^5 (1.31×10^5–6.15×10^6)	3.89×10^7 (2.0×10^6–1.24×10^8)	3.04	2.3–1.08×10^3	$P<0.05$
Experiment 2	47		8.91×10^5 (5.85×10^5–1.21×10^6)	1.15×10^8 (9.6×10^5–1.42×10^8)	24.84	7.98×10^1–2.08×10^2	$P<0.001$
Pg Experiment 1	53	5.0	4.65×10^1 (0–3.7×10^3)	3.9×10^7 (2.0×10^6–1.24×10^8)	5.62	1.58×10^3–4.37×10^8	$P<0.01$
Experiment 2	53		6.03×10^1 (0–1.7×10^4)	1.1×10^8 (9.6×10^7–1.42×10^8)	6.04	5.39×10^3–2.38×10^4	$P<0.001$

The temperatures chosen were those achieved within the pocket model during sonication for the times indicated (critical value of $t_{(0.05,6)}$= 2.45).

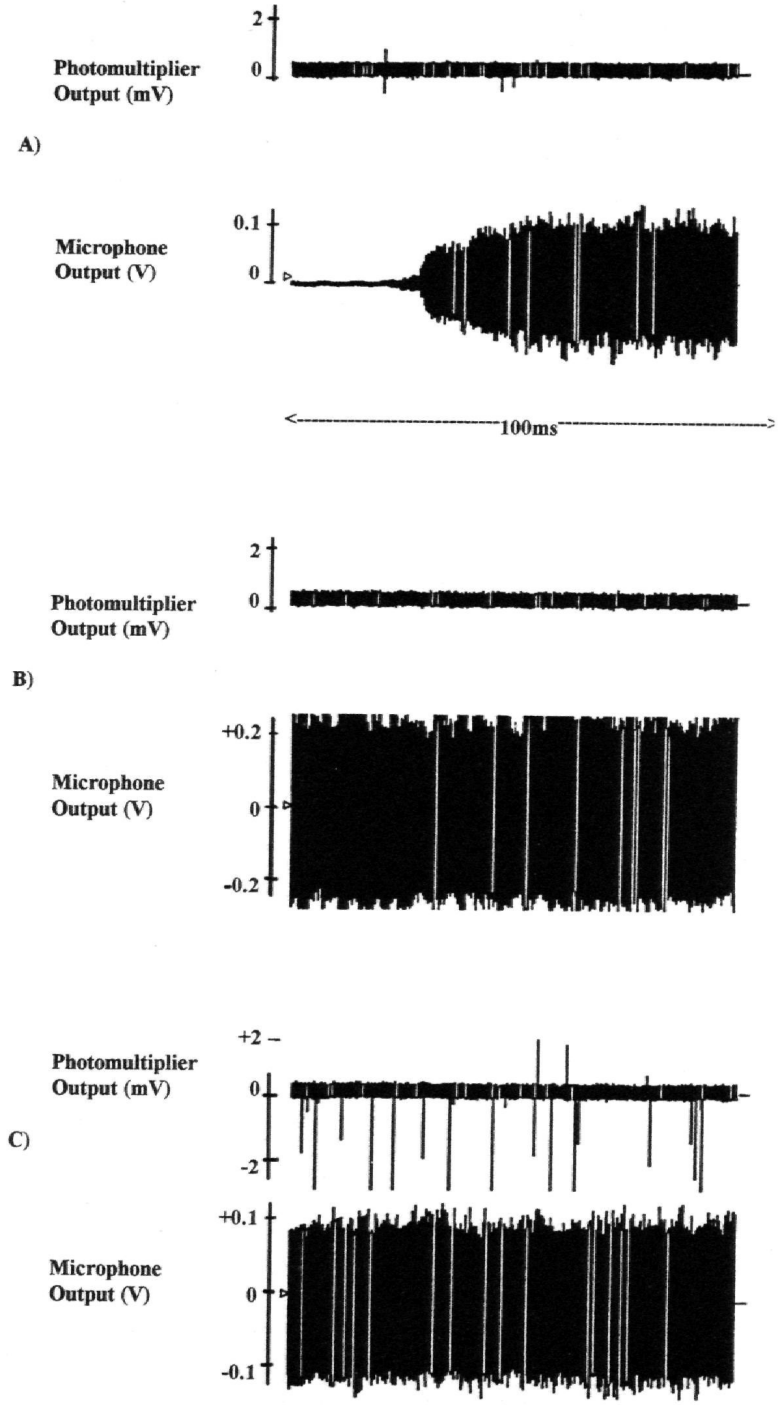

Fig. 3. Simultaneous recordings, each of 100 ms duration, of the outputs of the photomultiplier and the microphone (representing sonoluminescence and audible sound emissions from the scaler), taken: (A) at start of sonication; (B) 5 after start of sonication; (C) 10 s after start of sonication.

Statistical analysis

Microbiological data, in general, tend to be non normally distributed. To normalise the data, log transformation was carried out and geometric means calculated. Comparisons between test and control experiments were made using unpaired *t*–tests following log transformation and 95% confidence intervals calculated. However as log transformation had been used, the resultant confidence intervals are for the ratio of the means rather than the difference between them. Statistical significance is accepted if the confidence interval does not include 1.

Results

Sonication and bacteriolysis

The effect of sonication on the viability of Aa and Pg at 2.5 and 5.0 min are shown in Table 1. Statistically significant differences in colony forming units (cfu) ($P<0.01$ and $P<0.001$), were found between test and control samples for Aa at 5.0 min. This represents bacterial cell kills of 99.89% and 99.99% respectively for experiments 1 and 2 (calculated by: $((\bar{X}_{control}-\bar{X}_{test})\div\bar{X}_{control})\times100\%$). No significant differences in cfu occurred following exposure to ultrasound for 2.5 min. Sonication of Pg for both 2.5 and 5.0 min resulted in significant cell kill, corresponding to almost 100% reduction in each case.

Sonoluminescence production

Activation of the P1 insert in the simulated pocket model failed to generate any sonoluminescence when the water coolant was deflected from the tip. However when used normally, i.e. with water directed at the tip, sonoluminescence was only detected after 10 s activation as shown in Fig. 3. This corresponded to the stage at which the junction of the tip with the metal housing had become immersed by the water that had accumulated in the beaker. This is presumably the location of the cavitation.

Temperature effects on bacterial viability

The effect of temperature alone on bacterial viability and the associated statistical analysis is shown in Table 2. Significant differences ($P<0.05$) in the viable count were demonstrated between test and control samples of Pg at 2.5

and 5.0 min, and Aa at 5.0 min. When suspensions of Aa were held at 47°C for 2.5 min, there was a significant reduction in the viable count in only one of the experiments. However after 5.0 min and 52°C, 89% and 99.8% of the bacteria were killed in successive experiments. The effect of similar temperatures on Pg resulted in 100% kills at both time-periods.

The temperature recorded immediately adjacent to the activated ultrasonic tip located within the experimental pocket model space reached 31.3°C (30.5°C→33.4°C) and 33.6°C (30.8°C→34.7°C) at 2.5 and 5.0 min respectively under conditions of optimal cooling. When the coolant was deflected, the temperatures reached in excess of 50°C.

Discussion

This investigation was designed to examine further the *in vitro* bactericidal activity of dental ultrasound on the periodontal pathogens Aa and Pg following sonication within plastic tubes (Sved 1991). A more representative simulated pocket model was utilised here, providing a 2.0mm pocket space dimension, to ensure that the insert tip (measuring±1.0 mm in cross-section) would not engage the adjacent surfaces, thereby dampening any sonication activity, or producing an additional possible mechanical bacteriolytic action. While the size of this pocket space might appear unrepresentative, the fact that even modestly inflamed pocket wall tissue is readily displaced, without untoward discomfort by ultrasonic inserts as used in this investigation, suggests that a comparable pocket space also exists *in vivo*. The use of materials with acoustic impedance values similar to those of dentine and gingiva in the model construction, was designed to reproduce the acoustic absorption potential of the sonic activity within the natural pocket space. As the acoustic impedance value for cementum appears not to have been documented, that for dentine was used on the strength of its similar mineral content.

The choice of Aa and Pg pure cultures in this investigation reflects their strong association with chronic inflammatory periodontal disease (Dzink et al. 1988, Moore et al. 1991). The duration of the sonication periods was based upon the average time of 5 min estimated to achieve adequate root surface instrumentation *in vivo* by Badersten et al. (1984) and Gellin et al. (1986), and as such was much more conservative than the nine to twelve min in another study (Badersten et al. 1985). Instrumentation times of 3.9 and 5.9 min have been found to be appropriate in reducing microbial counts for ultrasonic and hand scalers respectively (Copulos et al. 1993). Statistically significant bactericidal effects in terms of viable counts were observed following both 2.5 and 5.0 min sonication exposure times for Pg, but only after 5.0 min for Aa.

The disruption of subgingival flora by ultrasonic instrumentation (Oosterwaal et al. 1987, Leon & Vogel 1987) is generally attributed to the well recognised mechanical chipping action of the insert tip (Walmsley et al. 1986), but the physical effects of the associated acoustic phenomena (Walmsley et al. 1988) and their potential for microbial destruction (Hamre 1948, Leadbetter & Holt 1974, Davies 1959, Robrish et al. 1976), may also be implicated. This would in turn be advantageous during subgingival instrumentation, when the direct contact with the root surface necessary for the mechanical oscillating cleaning action of the instrument tip, is precluded by variations in root form. Although subsequent examination failed to demonstrate transient cavitational activity within the experimental model and to which the bacteriolysis might have been attributed, the influence of the associated microstreaming shear forces, as shown by the disruption of pure cultures of *Escherichia coli* and the protozoa *Tetrahymena pyriformis* (Hughes & Nyborg 1962), cannot be discounted and further investigation thereof is necessary. The absence of cavitation may be related to the limited fluid medium, imposed by the obvious need to deflect the coolant water in the current investigation, coupled with the confined experimental pocket model space restricting cavitation, as suggested by the findings of Byrne et al. (1991). The former situation might indeed also arise clinically during subgingival instrumentation with the P 1 type insert investigated here, by virtue of the shielding effect of the overlying gingival pocket wall. This would however not be applicable to Thru flo type inserts, which would discharge coolant into the pocket space. The relevance of the size of the pocket space upon the facility for cavitation remains unclear and demands further evaluation, using experimental models of the type used in the current investigation.

An inevitable consequence of the necessary deflection of the coolant was heating of the instrument tip. This resulted in temperatures in excess of 50°C after sonication of the horse serum samples for 5.0 min within the experimental model. The subsequent examination of the possible bactericidal effects of such temperature rises alone revealed significant reductions in the viable count of Pg at both time periods, and for Aa at 5.0 min. These proved to be equivalent to those observed following the earlier corresponding sonication experiments, suggesting that an alternative thermolytic activity had operated within the experimental pocket model. However this mechanism would seem of little practical importance since such temperature changes would neither occur, nor be tolerated clinically. Further investigations of the acoustic bacteriolytic potential of ultrasonic scalers, should be directed at the thus far uninvestigated influence of the associated acoustic streaming phenomenon in experimental models which incorporate suitable cooling, to overcome the unwanted compounding influence of incidental hyperthemia upon the findings. In conclusion, whilst the findings of this investigation failed to confirm the existence of a supplementary bactericidal potential of ultrasonic instrumentation, such that the efficacy of these devices in clinical practice remains primarily dependant upon mechanical oscillation, the influence of acoustic phenomena does still warrant further examination.

Acknowledgement

The invaluable assistance of Dr J Bulman in the statistical analysis of the findings is acknowledged.

Zusammenfassung

Bakterizide Wirkung von detalen Ultraschallgeräten auf Actinobacillus actinomycetemcomitans und *Porphyromonas gingivalis. Eine in-vitro-Untersuchung*

Diese Studie untersuchte die mögliche bakterizide Wirkung dentaler Ultraschallscaler. Suspensionen mit *Actinobacillus actinomycetemcomitans* und *Porphyomonas gingivalis* wurden in einem akustisch simulierten Taschenmodell für 2.5 und 5.0 Minuten den Vibrationen eines Cavitron P1 Ansatzes ausgesetzt und anschließend wurden die lebensfähigen Bakterien ausgezählt. Innerhalb des

Taschenmodells wurde das Ausmaß jeglicher Cavitation, der die beobachtete statistisch signifikante bakterizide Wirkung zugeordnet wurde, mittels 'Sonolumineszenz' bestimmt. Diese wurde dann mittels Fotomultiplikationstechniken untersucht. Es gelang jedoch nicht, irgendeine Sonolumineszenz innerhalb des Taschenraumes nachzuweisen. Um die experimentelle Tasche nicht mit Wasser zu überfluten, wurde eine Ablenkung der Wasserkühlung von der Ansatzspitze notwendig. Dadurch ergaben sich zu den entsprechenden Zeitintervallen Temperaturen von 47.6°C bzw. 52.°C, welche einen alternativen bakteriziden Mechanismus bilden. Die Untersuchung der Wirkung von solchen Temperaturerhöhungen auf den Zielbakterien zeigten dann nach 5.0 Minuten, bei beiden Mikroorganismen, einen statistisch signifikanten Unterschied der Anzahl lebensfähiger Bakterien. Diese Unterschiede waren vergleichbar mit den vorherigen aus dem Taschenmodell. Während vermutlich die beobachtete bakterolytische Wirkung der Hauptuntersuchung durch anfängliche Temperaturveränderung verursacht wurde, darf bei Abwesenheit der akustischen Kavitation nicht der Einfluß jeglicher damit verbundener akustischer Mikroströmungen unterschlagen werden. Eine weitere Untersuchung zur Messung des bakteriziden Potentials von akustischen Phänomenen durch Verwendung eines modifizierten experimentellen Aufsbaus, der alle hyperthermischen Effekte ausschließt, ist daher notwendig.

Résumé

Effets bactéricides des ultrasons dentaires sur Actinobacillus actinomycetemcomitans et Porphyromonas gingivalis. Étude in vitro

Le présent travail est une étude des effets acoustiques bactéricides pouvant être produits par l'appareil à détartrer ultrasonique. Des suspensions d'*Actinobacillus actinomycetemcomitans* et de *Porphyromonas gingivalis* ont été soumises pendant 2.5 et 5.0 minutes aux vibrations causées par l'instrument à détartrer P1 d'un Cavitron dans un modèle simulant la poche parodontale du point de vue acoustique, et les éléments survivants ont été comptés. L'étendue de l'effet de cavitation se produisant éventuellement dans le modèle de poche, et auquel on aurait pu attribuer l'activité bactéricide statistiquement significative observée, a été déterminée par "sonoluminescence", qui a alors été étudiée par des méthodes de photomultiplication. Ces méthodes n'ont cependant pas réussi à mettre en évidence une sonoluminescence à l'intérieur de la poche, et on a de plus constaté que le fait de détourner l'eau de refroidissement de la pointe de l'instrument, nécessaire pour éviter l'inondation de la poche expérimentale, avait respectivement pour résultat des températures de 47.6°C et 52.3°C à 2.5 et 5.0 min, et pouvait ainsi constituer un mécanisme bactéricide alternatif. L'examen des effets produits sur les bactéries cibles par des changements de température de cet ordre a ensuite mis en évidence des différences statistiquement significatives dans les nombres d'éléments viables des deux micro-organismes après des périodes de 5.0 min, différences qui sont donc comparables à celles qui avaient été mises en évidence dans le modèle de la poche. Alors qu'on doit présumer que l'effet bactériolytique observé dans l'étude principale était dû à des changements de température produits accessoirement, on ne peut, en l'absence d'une cavitation acoustique, tenir compte de l'influence d'un microcourant acoustique qui y serait associé. Il est donc nécessaire de pratiquer des études ultérieures pour mesurer le potentiel bactéricide du phénomène acoustique, en employant une modification des procédés expérimentaux afin d'exclure tout effet hyperthermique.

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Address:

Rosalind O'Leary
Department of Periodontology
Eastman Dental Institute
256 Grays Inn Road
London WC1X 8LD
UK