# SONOCHEMICAL EFFECTS OF DESCALER-PRODUCED ULTRASOUND IN VITRO

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A b s t r a c t

The aim of our study was to quantify, by means of a chemical dosimeter, the chemical effects of cavitation ultrasound generated by a descaling tool. These effects are associated with the formation of cavitation bubbles that produce free radicals. Quantification (dosimetry) of chemical cavitation effects due to ultrasonic descalers has not been reported yet. In this study, a Piezon Master 400 ultrasonic unit with different working tips was used. A mixture of chloroform and distilled water, at a 1:1 ratio, served as a chemical dosimeter. $Cl^{z}$ ions, which are liberated during exposure to ultrasound, give rise to a weak hydrochloric acid (HCl). The pH values of this solution were measured in each sample at 11 different exposure times. Using a logarithmic scale, the linear dependence of pH values on exposure times was obtained. This approach also simplified statistical evaluation of the results. Our study showed that, in addition to mechanical cavitation effects, ultrasonic treatment for tooth descaling also resulted in formation of sonochemical products. Because the safety of patients is involved, the clinical importance of these reactions should be further studied.

K e y w o r d s

Ultrasound, Descaling, Cavitation, Chemical dosimetry

## INTRODUCTION

The main aim of our study was to demonstrate and quantify the chemical effects of cavitation ultrasound generated by a commonly used descaling tool in order to contribute to safety assessment of ultrasonic descaling. The measurement was carried out by means of a liquid chemical dosimeter.

The chemical action of intense ultrasound, which is similar to the action of ionising radiation, has extensively been studied in biomedical research for many years (*1*). The basic mechanism of chemical effects of ultrasound can be explained on the basis of the formation of cavitation bubbles that, during their vigorous oscillation, are able to produce free radicals. This phenomenon is called cavitation. Oscillating bubbles behave like heat engines. Therefore, very high temperatures (several thousand °C) can be achieved at the moment of adiabatic compression of bubble content (*2*).

When ultrasonic cavitation (similar to ionising radiation) acts on aqueous solutions of certain compounds, including dissolved air oxygen and nitrogen, free radicals produced due to water molecule decomposition react with these compounds or gases. Both free radicals and other compounds formed inside the solution (e.g., hydrogen peroxide or nitrous and nitric acids) are of particular biological importance considering their chemical reactivity or even toxicity.

The chemical effects of low-frequency ultrasound produced by laboratory disintegrators have been described in our previous papers (*3,4*). In these studies, we investigated, using spectrophotometry, the effects of low-frequency ultrasound cavitation on concentrated solutions of selected simple organic compounds, i.e., liberation of iodine from potassium iodide as well as the production of heat in these solutions. The methods used in our experiments are described in the radiochemical literature (*5*). The simplest way of how to quantify the amount of HCl produced is based on measurements of a decrease in pH value. Chromatographic assessment of a decrease in chloroform concentration in the presence of hydrogen peroxide, as reported by Chen (*6*), is another option.

The effects of low-frequency ultrasound on cell lysis have also been studied in our laboratory (*7*). However, according to the present knowledge (*8,9*), bacterial cells do not seem to provide an appropriate model to study cavitation-conditioned cell lysis. *O'Leary et al.* did not investigate cavitation and thermal effects separately in their study (*8*). *Schenk et al.* found that cavitation alone had no statistically significant anti-microbial effects (*9*). In our opinion, this was caused by small dimensions of bacterial cells, mechanical rigidity of their cellular walls and also by the non-homogeneity of a low-frequency ultrasonic field.

It is a well-known fact that cavitation phenomena play a role in ultrasonic descaling (*10, 11,12,13,14,15,16,17,18*). Main attention has so far been paid to the mechanical effect of cavitation, which is the most important descaling mechanism. Photomicrographs have revealed microscopic spots of the destructive action of oscillating cavitation bubbles directly in the plaque layer (*15,16*). *Williams and Walmsley* estimated exposure of teeth to ultrasound on the basis of an increase in temperature in the tooth. They also measured amplitudes of oscillation of descaler working tips (*17*).

It should be admitted that dosimetry of low-frequency ultrasound presents a general research problem. Any method of ultrasound dosimetry based on measuring ultrasound intensity by means of hydrophones cannot be appropriately used in small-volume insonation vessels because of non-homogeneity of the ultrasonic field. Measurement of sonoluminescence (weak light emitted from hot gaseous content of cavitation bubbles) was used to assess cavitation produced by oscillating ultrasonic root débridement files (*19*). *Khambay et al.* measured sonoluminescence enhanced by luminol, a fluorescent dye excitable by hydrogen peroxide, around a descaler working tip. The results were influenced, among other factors, by the shape of a working tip (*20*).

However, it is very difficult to design experimental conditions that will provide reproducible sonoluminescence measurements. Better conditions for reproducible results of dosimetry were provided, for instance, when only the displacement amplitudes of vibrating working tips or probes were measured or, if cavitation was present, chemical dosimetery was used (*17*). We assume that, for comparative dosimetry of low-frequency ultrasound descalers or other ultrasonic tools used in dentistry, the measurement of well-defined chemical effects of cavitation can be employed. According to our knowledge, no direct quantification of chemical effects accompanying cavitation produced by ultrasonic descalers has been made yet.

## MATERIALS AND METHODS

### ULTRASONIC APPARATUS

A multipurpose ultrasonic unit (Piezon Master 400, EMS, Switzerland) equipped with three different descaling tips was used. The instrument was applied at a working frequency of 32 kHz. We used 50% of the maximum descaling power setting and this value was kept constant for every sample investigated because we expected that, at this setting, instrument power and frequency would provide maximum stability even if relatively long exposure times were used. Standard tips A, B and C (as denoted by the producer) were used as working parts of the ultrasonic unit. These tips slightly differed in their shape and also working area. Tip A was pointed and had a smaller working area than tips B and C (*Fig. 1*).

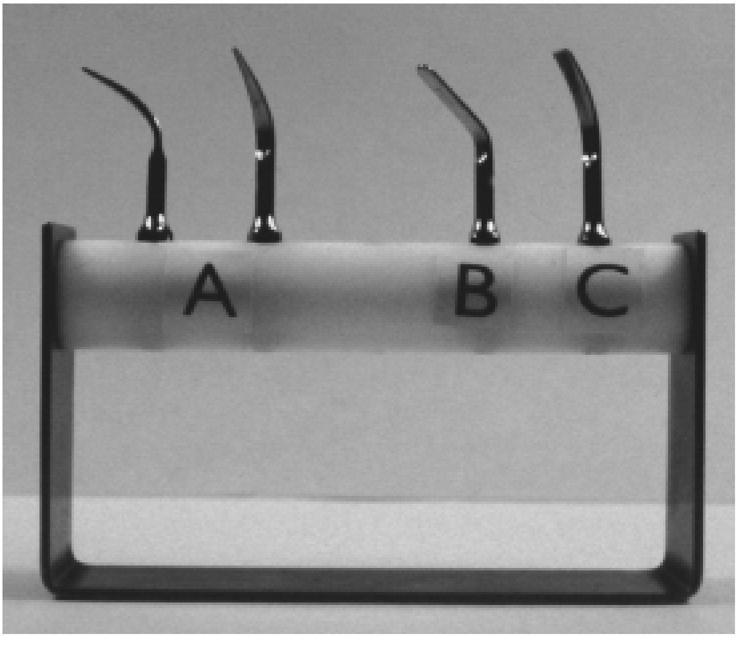


*Fig.1*
Descaling working tips

Dosimetry

Chloroform ($CHCl_3$, MW 119.38, analytical grade, Lachema a.s., Czech Republic), in a 1:1 mixture with redistilled water, was used as a chemical dosimeter. Other radiochemical dosimeters commonly used in sonochemistry (e.g., Fricke dosimeter or oxidation of ferrous to ferric ions) were not sensitive enough, as shown by our preliminary experiments. The volume of each sample treated was always 50 ml (25 ml chloroform and 25 ml redistilled water). Both components were mixed in a glass weighing bottle (8 cm high, 4 cm in diameter). Redistilled water formed the upper (light) phase and chloroform the lower (heavy) phase. During exposure to ultrasound, liberated $Cl^-$ ions reacted with $H^+$ ions, forming hydrochloric acid. With increasing exposure times, more HCl was formed and pH values of the water phase decreased.

Measurement of pH values

An MS 22 laboratory pH-meter (Laboratorní přístroje Praha, Czech Republic), equipped with a common, combined glass electrode, was used for the measurements. After temperature had been measured, the pH value was assessed in each 20-ml water-phase sample at 30 min after ultrasonic exposure. To ensure the constant temperature (20 to 21 °C) throughout the experiment, the samples were kept in a water/ice bath.

The working tips were fully immersed in the chemical dosimeter liquid, at a constant depth for each measurement, with their longitudinal axes perpendicular to the liquid surface. The times of exposure were linearly increased, and the longest exposure time did not exceed 600 s. The resulting pH values were averaged and plotted on a graph.

## RESULTS

The pH value of the water phase of the chemical dosimeter was measured at eleven different exposure times. At the first six exposure times, measurements were repeated five-times. The pH values obtained at longer exposure times were measured only once because they have limited clinical importance (in dental practice only short treatment times are used). These values are reported only for a better understanding of the chemical process. The values obtained are presented in *Fig. 2*.

A logarithmic scale was used to construct a curve of the dependence of pH values on exposure times (*Fig. 3*). Linear dependence was obtained for all three tips used. The data were statistically evaluated and regression curves and coefficients were calculated as follows:

Tip A: $y = -0.5498\ln(x) + 7.0767$ ($R^2 = 0.9938$)
Tip B: $y = -0.5300\ln(x) + 6.6848$ ($R^2 = 0.9962$)
Tip C: $y = -0.5497\ln(x) + 7.0226$ ($R^2 = 0.9949$)

It can be seen that the line slopes (numbers in the first term on the right sides of these equations) are very similar and the probability that dependence is linear is larger than 99 % in all cases. This is very important for future evaluation and comparison of cavitation effects in different ultrasonic systems.

The results of our measurements show that cavitation effects of ultrasound manifest themselves as an exponential decrease in pH values.

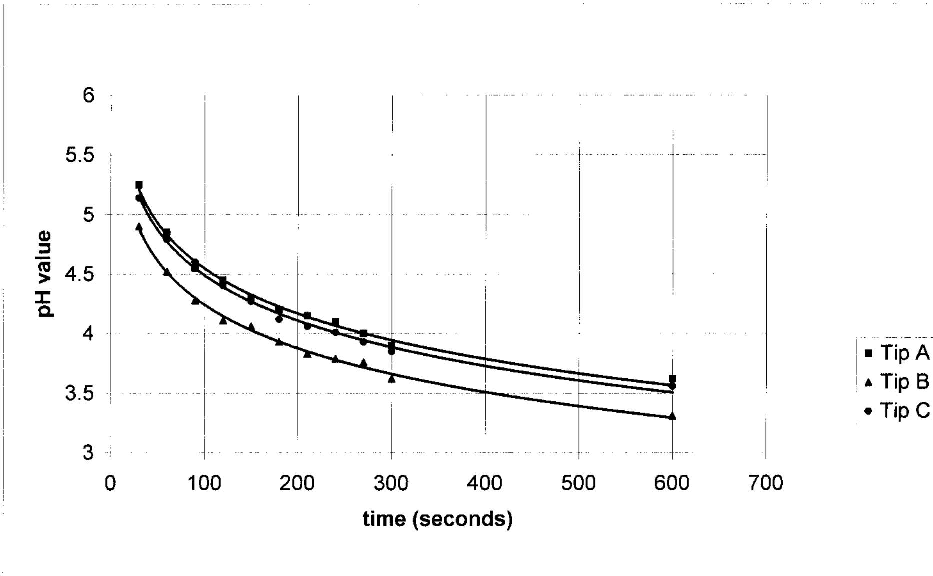


*Fig.2*
A curve showing the dependence of pH values on sonication time for all working tips used

DISCUSSION

Our results demonstrate that ultrasonic tools used for descaling are chemically active, i.e., they are able to give rise to sonochemical products in aqueous media. The amounts of these products depend exponentially on exposure time. The chemical significance of this fact is simple to explain. The curves illustrate a first order chemical reaction, i.e., reaction in which one product molecule is formed from one substrate molecule. Therefore, the reaction rate directly depends on the substrate (chloroform) concentration. It is rather difficult to explain the differences observed among the individual working tips used. The smallest tip (A) appeared to have the lowest chemical activity, i.e., lowest decrease in pH value. However, different effects can also be due to differences in tip geometry, which influences its oscillation modes and amplitudes. These complex physical properties of oscillating tips can be directly related to chemical effectiveness only with difficulty.

In pure water, saline or even saliva, the same primary sonochemical products (free radicals), as observed in our cavitation experiments, are probably formed. It is possible that they occur at higher concentrations because of the well-known

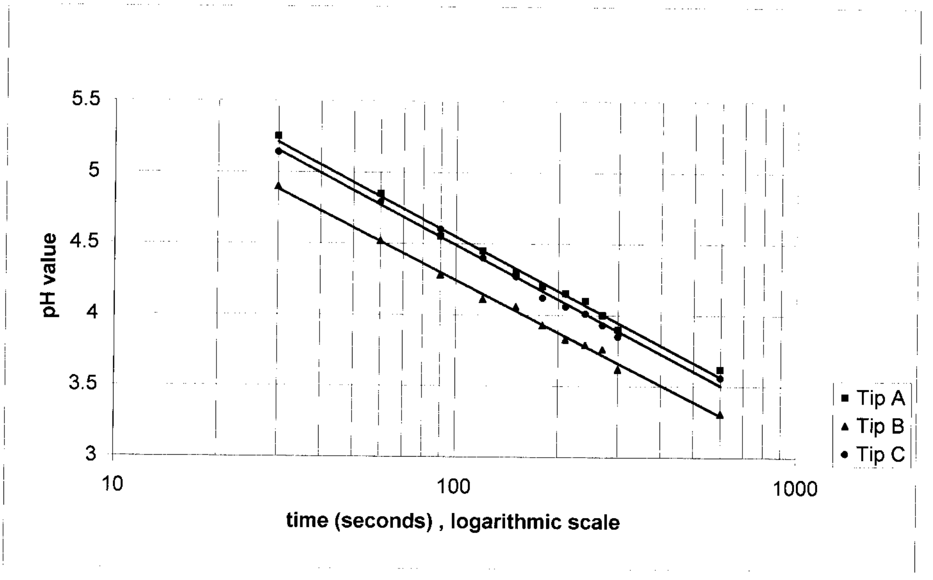


*Fig.3*
A curve showing the dependence of pH values on sonication time. Scale on the abscissa is logarithmic

damping effect of compounds produced by multi-atomic molecules. Consequently, these molecules, especially those with high vapour pressure, reduce the chemical effect of cavitation (*21*). It can be proved, on the basis of thermodynamic considerations or in experiments, that the presence of these compounds decreases the maximum temperature achieved inside collapsing cavitation bubbles. Chloroform can certainly be classified as one of these damping compounds.

Cavitation products may also cause damage to the sensitive gingival mucosa. However, on the basis of our experiments and those reported up to now, it has been difficult to assess the clinical importance of this damage. This should be studied in further experiments and compared with the cytotoxic and lytic effects of ultrasound. Because of the frequent use of anti-bacterial drugs in the descaler cooling water, interactions between primary sonochemical products and these drugs should also be studied.

The results of our study showed that, in addition to the known mechanical cavitation effects, ultrasound used in the process of tooth descaling also gives rise to sonochemical products. The clinical importance of these facts should be further studied with regard to patients' safety.

A c k n o w l e d g m e n t

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## SONOCHEMICKÉ ÚČINKY ULTRAZVUKU GENEROVANÉHO PŘÍSTROJEM PRO ODSTRAŇOVÁNÍ ZUBNÍHO KAZU *IN VITRO*

S o u h r n

Cílem naší práce bylo kvantifikovat pomocí chemického dozimetru chemické účinky ultrazvukové kavitace, generované přístrojem na odstraňování zubního kamene. Chemické účinky ultrazvuku jsou spojeny s vytvářením kavitačních bublin produkujících volné radikály. Kvantifikace (dozimetrie) chemických účinků kavitace u ultrazvukového přístroje pro odstraňování zubního kamene zatím provedena nebyla. Byl použit ultrazvukový přístroj Piezon Master 400 s různými pracovními nástavci. Jako chemický dozimetr byla použita směs chloroformu a redestilované vody v poměru 1:1. Uvolňované ionty $Cl^-$ vytvářejí během expozice roztok HCl, výsledná hodnota pH je měřena u každého jednotlivého vzorku při 11 rozdílných dobách expozice. Pro získání lineární závislosti doby expozice na hodnotách pH a zjednodušení statistického vyhodnocení byla expoziční doba vynesena také v logaritmickém měřítku. Tato studie ukázala, že kromě mechanických účinků ultrazvukové kavitace podílejících se na odstraňování zubního kamene, dochází též k tvorbě sonochemických produktů. Její klinická významnost by měla být posuzována s ohledem na bezpečnost pacientů.

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