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SCIENTIFIC articles

Effects of various acids on the buccal surface of human permanent teeth: a study using scanning electron microscopy*, †

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Abstract

Contradictory evidence exists as to the acid necessary for producing optimum enamel etching on buccal tooth surfaces. The aims of this study were to (1) compare the etching abilities of four different acids; (2) identify the acid concentrations required for optimum etching; and (3) determine the concentration stability of phosphoric acid solutions. Crowns of caries-free molar teeth were pumiced, sectioned mesiodistally, washed, and dried. Study groups of 10 specimens were etched with either phosphoric, lactic, citric, or pyruvic acid prepared in concentrations from 5 to 70%. Etching was performed for 1, 1½, or 5 minutes. Specimens were prepared and examined with scanning electron microscopy. To simulate clinical conditions, the final part of the experiment exposed 37% H_3PO_4 to room air for various time periods. Stability of the acid concentration was determined. Phosphoric acid produced the most desirable etching effects when applied for either 60 or 90 seconds. Investigations of different concentrations of phosphoric acid suggested that a superior etching result is obtained within the range of 30 to 40%. Titration experiments revealed that 37% phosphoric acid was reasonably stable under normal clinical conditions.

Introduction

Using 85% phosphoric acid, Buonocore[1] demonstrated that treating enamel surfaces with acid enhanced adhesion of resins to tooth surfaces. The adhe sion was improved by etching the enamel surface with the acid. However, as various resin materials were introduced to the commercial market, other acids were suggested as etchants.[2-4] This has led to controversy as to the type of acid necessary for producing optimum enamel etching on buccal tooth surfaces. Therefore, the initial aim of this investigation was to compare the etching abilities of four different acids on human buccal enamel surfaces using scanning electron microscopy (SEM).

Phosphoric acid has been used most widely by clinicians and researchers. Controversy has existed, however, as to the concentrations to be used. Despite a paucity of information, 37% H_3PO_4 has become most popular. Thus, a second aim of this study has been to determine with SEM which concentration of phosphoric acid yields optimum etching on human buccal surface enamel.

Clinicians may recall that when using silicate cements, they were cautioned about changes in the concentration of the liquid. The liquid, an orthophosphoric acid, was hygroscopic. Since currently used phosphoric acids are similar in chemical structure, the final objective of this investigation was to determine phosphoric acid stability by simulating clinical conditions and performing a series of titration experiments.

Materials and Methods

To compare the effects of four different acids, 1040 human permanent premolar and molar teeth were polished with a fine abrasive pumice and then washed thoroughly. The teeth were sectioned using a Gillings and Bronwill sectioning machine, and their buccal surfaces were mounted on glass slides. A minimum of 10 specimens was assigned to test each acid type and concentration. The acids for study were selected either because they currently are used in clinical practice or because they were suggested by previous researchers. Accordingly, the buccal surfaces were treated with the following solutions [numerical concentrations, weight/weight (W/W) percentage expressions]: (1) phosphoric acid (30, 40, 50, 60, 70); (2) lactic acid (20, 30, 40, 50, 60, 70); (3) citric acid (30, 40, 50, 60, 70); and (4) pyruvic acid (5, 10, 15, 20, 25, 30, 35, 40, 45, 50).

Solutions were applied to the specimens using a sable hair brush for different periods of time (1, 1½, 2,

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or 5 minutes). Following acid treatment, teeth were washed thoroughly and dried with compressed air. Specimens were mounted on stubs, dessicated, and vacuum coated with a 200-A layer of gold palladium for examination with a Hitachi 2R scanning electron microscope. Fine sections were taken then from specimens and examined with the light microscope for quantitative analysis.

The quality of the acid etching was determined with a light microscope by the etching depth. Etched surfaces were classified as follows: "1" referred to a 0 to 10 μm depth; "2" represented etching of 10 to 25 μm; and "3" indicated the depth of etching ranged between 26 and 50 μm.

Although phosphoric acid has been used widely, debate has centered on the concentration required for optimum etching. Accordingly, phosphoric acid concentrations (31, 32, 33, 34, 35, 36, 37, 38, 39, and 40%; W/W) were prepared. Specimens were prepared as described previously. A minimum of 10 specimens was treated with each concentration of H_3Po_4 for 1 minute. Similar to the previously outlined procedures, specimens were examined with the scanning electron microscope.

With 37% H_3PO_4 (W/W), the third part of this investigation attempted to simulate clinical conditions to determine concentration changes in the acid. The phosphoric acid was exposed to air in three ways. (1) Acid was placed in two wide-mouthed 15-ml vials and left uncovered for 51 hours at room temperature. If a clinician were using the acid for 15 minutes a day, this would approximate 10 months of clinical usage. (2) A 500-ml bottle was exposed to air for 2½ hours, each time period simulating 2 weeks of clinical exposure. This was repeated 20 times, again approximating 10 months of clinical usage. Following each exposure, two 10-ml samples were drawn for titration. (3) Acid was placed in 40 wide-mouthed 15-ml vials and left uncovered for 2½ hours each period. The concentration of two vials was tested following each exposure.

The indicator used for titrations consisted of two parts phenolphthalein and one part napholphthalein in 45% ethanol. Calculation of the concentrations of the samples was made (Appendix 1), and mean values were taken to express results.

Results

The acids examined in this study produced a wide range of effects on buccal enamel. The results of the four different acids with differing concentrations and varying application times are summarized in Table 1. Since 10 specimens were used to test each parameter, these data represent the consensus of findings for each group. In almost all instances, there were exceptions as buccal enamel surfaces do not react uniformly over their entire areas.

The data from Table 1 reveal that three acids, *i.e.*, lactic, citric, and pyruvic, did not produce high-quality etching patterns. Regardless of their application times or concentrations, the etching was poor as compared to the phosphoric acid groupings.

Teeth treated with 20% lactic acid for 1 minute offered the best results for this acid. However, these etches displayed only an outline of shallow, superficial pits (Fig. 1*A*). This reaction was mild, and it was often difficult to distinguish the etched surfaces from non-treated enamel. When lactic acid concentrations were increased up to 70%, the etching pattern was not significantly improved.

Citric acid etchings were generally unsatisfactory. Only when the exposure time was increased to 5 minutes was a mild etching observed. However, the depth of the microirregularities was only 6 μm, which is considered unacceptable for retention of resin materials (Fig. 1*B*).

Pyruvic acid at the concentrations of 10 and 30% gave satisfactory etching patterns when applied for 1

Table 1. Effects of different acid concentrations on enamel

Acid type and concentration (%; W/W)		Time (min) 1	1½	2	5
Phosphoric	20	2*	2	1	1
	30	3	3	1	1
	40	3	3	1	1
	50	3	3	1	1
	60	1	1	1	1
	70	1	1	1	1
Lactic	10	1	1	1	1
	20	2	2	1	1
	30	2	2	1	1
	40	1	1	1	1
	50	1	1	1	1
	60	1	1	1	1
Citric	10	1	1	1	1
	20	1	1	1	1
	30	1	1	1	1
	40	1	1	1	1
	50	1	1	1	1
	60	1	1	1	1
Pyruvic	5	1	1	1	1
	10	2	2	1	1
	15	1	1	1	1
	20	1	1	1	1
	25	1	1	1	1
	30	2	2	1	1
	35	1	1	1	1
	40	1	1	1	1
	45	1	1	1	1
	50	1	1	1	1

* Numerators, the findings for each group of 10 specimens studied using the light microscope. Results are a consensus for each group with exceptions to the rule in most cases. 1, 0- to 10-μm etch; 2, 11- to 25-μm etch (acceptable); 3, 26- to 50-μm etch (excellent).

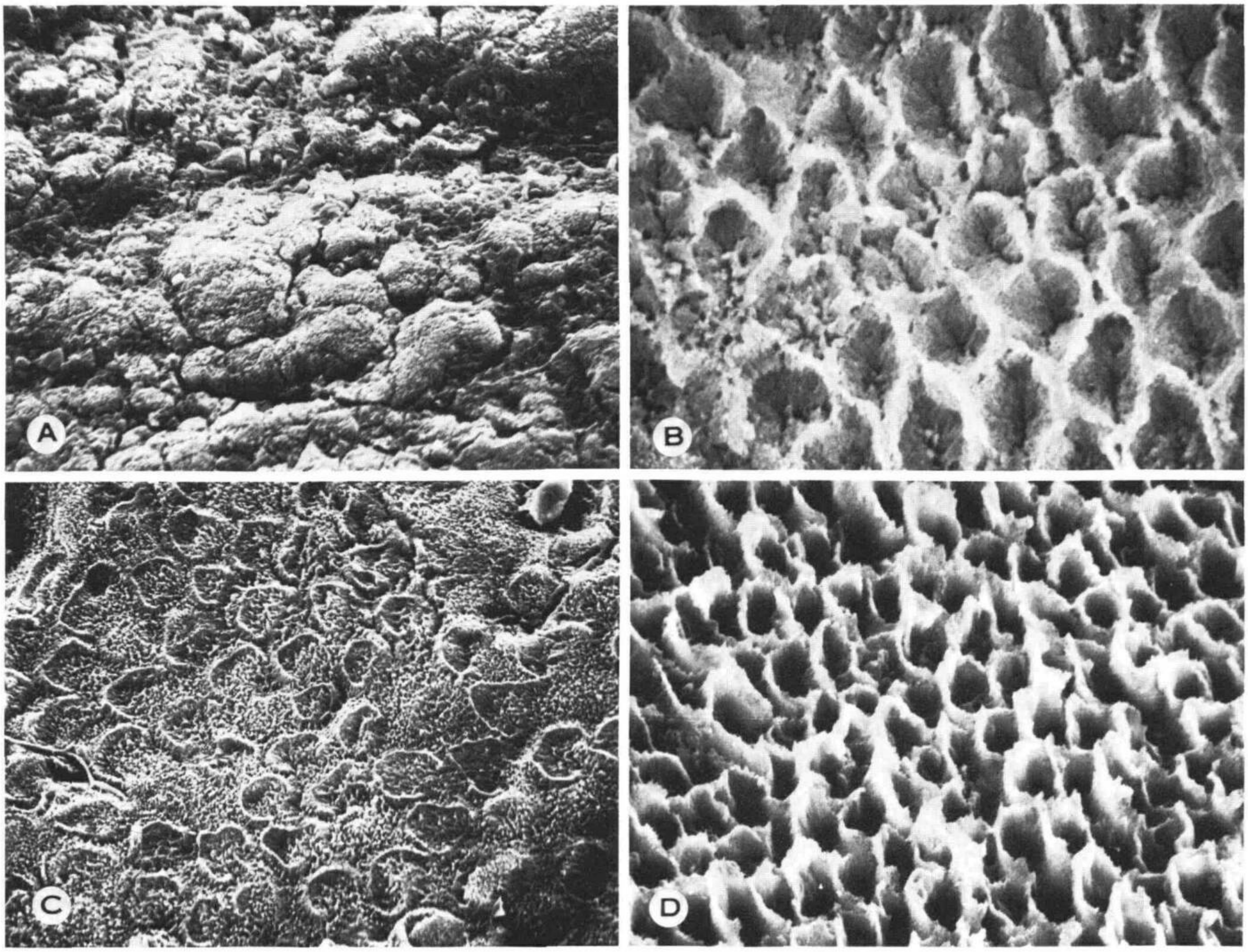

Fig. 1 *A*. Scanning electron micrograph of an enamel surface that has been treated with 20% lactic acid for 1 minute. Notice shallow, superficial pitting. Original magnification × 1000. *B*. Scanning electron micrograph of an enamel surface that has been etched with 20% citric acid for 5 minutes. Original magnification × 1750. *C*. Scanning electron micrograph of an enamel surface that was exposed to 10% pyruvic acid for 1 minute. Notice satisfactory etching of prisms. Original magnification × 1400. *D*. Scanning electron micrograph of an enamel surface that has been etched for 1 minute using 30% phosphoric acid. Original magnification × 850.

minute (Fig. 1*C*). An increase in etching time produced no significant improvement in the quality of the etch. Although the overall pattern of etching was acceptable, examination with the light microscope showed that the maximum depth of microcavities was only 6 μm. Compared with the much greater depth of phosphoric acid etches, it was concluded that the pyruvic acid patterns were unacceptable.

Phosphoric acid generally yielded better etching results when applied for 60 or 90 seconds within the concentration range of 30 to 40%. The etching consistency, however, was not as satisfactory as the 20% level. Although 50% concentrations yielded excellent results, the etches tended generally to borderline on the acceptable depth range. Optimum etching in the 26- to 50-μm range was most frequently observed with 30 and 40% acid concentrations. An example of this etching is offered (Fig. 1*D*). It is interesting to note that 5-minute applications of acid failed to improve the etch quality.

The second part of this investigation focused on the phosphoric acid concentrations between 30 and 40%. Little difference in the concentration groups was detected by SEM. Further examination with the light microscope showed that 30% phosphoric acid when applied for 1 or 1½ minutes produced microcavities averaging 25 to 35 μm in depth as measured from the surface. Interestingly, the 37% H_3PO_4 solution showed no better results than did other concentrations within this range. When the actual depth of microcavities (*i.e.*, not from the surface) was measured, a more constant depth approximateing 20 microns was observed (Fig. 2).

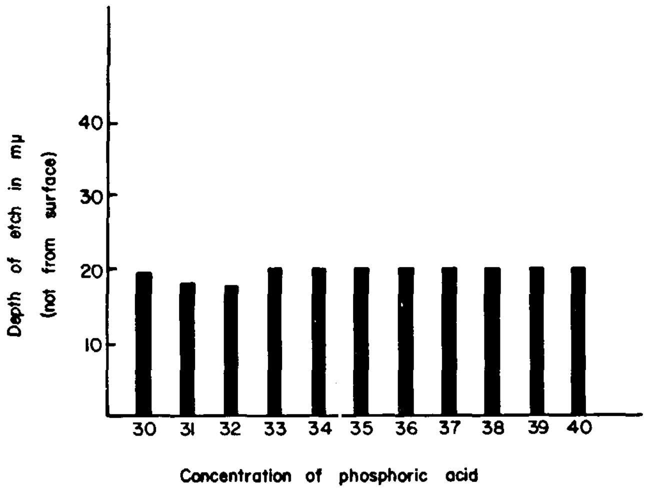

Fig. 2. Effect of varying concentration (30 to 40%) of phosphoric acid on surface of enamel.

When phosphoric acid stability was investigated, only the 51-hour constant exposure created a critical concentration change. The phosphoric acid altered in concentration from 37 to 49%. Subsequent trials had changes of lesser significance, none of which exceeded 2%.

Discussion

The study demonstrated that all acids tested are capable of etching enamel at either high or low concentrations. Since enamel is composed of 96% inorganic material which is susceptible to acid destruction, this was expected. A wide range of patterns of varying depth was produced depending on the type of concentration of acid used as well as the etching time.

SEM and light microscope observations demonstrated that an increase in etching time does not necessarily improve the resultant etch. Rather than simply deepening the microcavities, longer exposures to acid may produce destruction of surrounding enamel. These results concur with the findings of other investigators.[6–8]

Pyruvic, lactic, and citric acids have been cited as producing acceptable etching patterns.[5, 9, 10] It was reasonable, therefore, to compare the etching abilities of these acids with the widely used phosphoric acid. However, buccal enamel surfaces conditioned with these acids in the present study displayed etching patterns considerably inferior to those obtained with phosphoric acid. Moreover, pyruvic acid decomposes at room temperature, and clinicians should be aware that refrigeration is required.

The experiments supported the conclusions of Silverstone[8] who found that a 1-minute application of any phosphoric acid solution with a 30 to 40% concentration produces acceptable results.

However, the present investigation showed also that a 1½ minute acid treatment produced comparable results and that acid exposures for periods of 2 to 5 minutes led generally to destruction of enamel and unacceptable etching patterns.

Since the findings of the initial part of this study indicated optimum etching results from phosphoric acid in the 30 to 40% range, the second part of the investigation attempted to determine etching differences within this range. No significant differences were detected among specimen groups etched with all the concentrations in this range when applied for either 60 or 90 seconds. While most common commercial etching agents consist of 37% acid phosphoric (W/W), this concentration does not appear to have any distinct advantage over the concentrations in this range.

It is well known that enamel mineralization and maturation affect surface enamel.[11, 12] It follows that surface enamel differs from one tooth to another and from surface to surface. Accordingly, differences in acid etches may result from a multitude of factors which affect mineralization, maturation, and, thus, surface composition of the specimens selected.[13, 14] For these reasons, the investigators are of the opinion that a minimum of 10 specimens is necessary for studying enamel surface morphology with SEM.

The SEM results obtained in the present study should not be considered as necessarily refuting the conclusions of other investigators. Rather, they should be viewed as a recognition of factors which may lead to varying results. The observations were made solely on buccal surfaces of human posterior teeth.

Since concentration of phosphoric acid can be crucial to the quality of acid etching, the third part of this investigation focused on acid stability. From the various tests, it appeared that a continuous exposure of the acid to room air enhances the possibility of changes in H^+ concentration. This change was due to the hygroscopic nature of H_3PO_4; thus, H_2O was absorbed from the ambient relative humidity of the air. As a consequence, the increased H_2O content diluted the H_3PO_4 and, therefore, increased the H^+ concentration due to the increased dissociation of the H_3PO_4, rendering the solution more acidic. However, this aspect of the study suggests that two guidelines be followed: (1) phosphoric acid liquid must be capped when not in use; and (2) containers with small openings to limit evaporation and subsequent concentration changes must be used.

Conclusions

Based on the results of this investigation, the following was concluded: (1) deeper etching was obtained with phosphoric acid when compared to lactic, citric, and pyruvic acids; (2) deepest etch was produced with 30 to 40% phosphoric acid; (3) since the concentration of phosphoric acid can change when exposed to room air for lengthy time periods, containers should be closed when not in use.

Appendix

Sample calculation of H_3PO_4 concentration

1. A 10-ml sample of H_3PO_4 is taken, and the weight of the sample is determined as 12.39 g.
2. To reach the end point of the titration, 46.9 ml of 2 N NaOH were used.
3. Using the equation $N_A V_A = N_B V_B$, where N = normality, V = volume, A = acid, and B = base, we substitute to find N_A.

   $$N_A \times 10 \text{ ml} = 2 \text{ N} \times 46.9 \text{ ml}.$$

   Therefore, $N_A = 9.38$.
4. Since H_3PO_4 is a tribasic acid and we have titrated only to the second end point, we must multiply this value by $^3/_2$ to determine the true normality of the sample. Therefore, $N_{H_3PO_4} = 9.38 \times {}^3/_2 = 14.07$.
5. To determine the molarity of the sample, we divide the normality by 3 (again, since H_3PO_4 is tribasic). Therefore, $M = 14.07/3 = 4.69$.
6. Since the M.W. of H_3PO_4 is 98 g, the weight of H_3PO_4 in our 10-ml sample is:

   $$4.69 \text{ g/liter} \times 98 \text{ g} \times \frac{1000 \text{ ml}}{10 \text{ ml}} = 4.59 \text{ g of } H_3PO_4$$
7. Therefore, % of sample (w/w)

   $$= \frac{\text{weight of } H_3PO_4 \text{ in sample}}{\text{total weight of sample}} = \frac{4.59 \text{ g}}{12.39 \text{ g}} \times 100 = 37.05\%$$

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