Protective effects of two food hydrocolloids on dental erosion: Nanomechanical properties and microtribological behavior study

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Abstract: In this study, the protective effects of two food hydrocolloids, Xanthan gum and Arabic gum, on dental erosion are investigated from the perspective of the nanomechanical properties and microtribological behavior of acid-eroded enamel. Enamel specimens prepared from extracted human teeth were immersed in citric acid solution (CAS), CAS with 0.03% w/v Xanthan gum and CAS with 0.03% w/v Arabic gum, respectively, for 10 min to obtain three groups of eroded specimens. The nanomechanical properties and microtribological behavior of enamel were examined using nanoindentation/scratch techniques. The results show that compared with Arabic gum, Xanthan gum inhibits enamel surface demineralization and acid permeation more effectively because of a more uniform and denser adsorption on the surface of the enamel. The impairment of the nanomechanical and microtribological properties of the enamel surface by acid erosion is mitigated more significantly by adding trace amounts of Xanthan gum than Arabic gum. In summary, adding trace food hydrocolloids reduces enamel surface demineralization and inhibits acid permeation to mitigate the influence of erosion on the mechanical and tribological properties of enamel. The adsorption state of food hydrocolloids is the determining factor in the permeability of acid agents into the enamel and plays a significant role in preventing dental erosion.

Keywords: dental erosion; Xanthan gum; Arabic gum; nanomechanical properties; microtribological behavior

1 Introduction

Dental friction and wear is an inevitable lifetime process due to normal masticatory function, but excessive wear of teeth can cause many problems, such as unacceptable damage to the occluding surfaces, alteration of the functional path of masticatory movement, dentine hypersensitivity, and even pulpal pathology [1]. Due to an increased life expectancy, tooth wear has received considerable attention.

Enamel, the outmost covering of a tooth, is exposed to the occlusal surface and chemical environment within the mouth. It contains 92–96 wt% hydroxyapatite (HAP) and has a compact alternate arrangement of keyhole-like rods and inter-rod enamel [1]. HAP crystals are organized and glued together and then assembled into nano-fibrils and fibers in enamel rods. The enamel is particularly vulnerable to acid attack because HAP generally dissolves in acidic media. The surface loss of teeth caused by chemical or electrochemical action without the involvement of microorganisms is widely called “erosion” in dentistry.

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Erosion not only results in the direct erosive substance loss of enamel but also decreases the hardness and elastic modulus of the enamel surface [2, 3]. Hence, acid-eroded enamel has a softened surface, and therefore becomes more susceptible to abrasion and attrition [4]. With the increasing consumption of acidic beverages, including soft drinks, fruit juices, and sports drinks, erosion has gradually become the main cause of tooth wear [5].

Two types of solutions are proposed and used to prevent dental erosion caused by acidic beverages. One solution is to administer a mineralizing agent, such as mouth rinses containing SnCl₂ or NaF, immediately following an acidic challenge to mitigate the acid erosion through an enhanced rapid mineralization [6]. The other solution is to reduce the erosive potential of beverages by product modification [7]. One effective method is to lower the acid content of beverages. However, the special tangy taste of such beverages is associated with their low pH value [8]. Recently, efforts have been focused on reducing the erosive potential of acidic beverages by the addition of food-approved additives.

It was reported that some small molecular additives, such as citrate, calcium and phosphate ions [9], and fluoride [6], could inhibit HAP dissolution in acidic media; however, these additives might deteriorate drink flavors [10] and cause problems with drink formulation and stability, especially at a high concentration. Moreover, the addition of fluoride to drinks is forbidden in the European Union (EU) and in several non-EU countries. Recently, significant attention has been paid to food-approved polymer additives. Many studies have indicated that the addition of trace food hydrocolloids, such as Xanthan gum and Arabic gum, has the potential to reduce the mineral loss in enamel caused by acidic beverages and has no obvious influence on beverage taste [11–14]. However, the anti-erosion mechanisms of food hydrocolloids are unclear, and previous studies scarcely attempted to investigate the effects of anti-erosion measures on dental anti-wear performance.

Mastication is the most important physiological function of human teeth, and dental mechanical and tribological properties cannot be ignored in the anti-erosion strategies. Given that erosion weakens the mechanical and tribological properties of enamel as a result of surface demineralization [1–3], it is reasonable to deduce that as effective anti-erosion agents, food hydrocolloids should have the potential to decrease the influence of acid attack on the anti-wear performance of enamel. Thus, in this study, the protective effects of two food hydrocolloids, Xanthan gum and Arabic gum, on enamel erosion are investigated from the perspective of the nanomechanical and microtribological properties of acid-eroded enamel. According to the morphology, nanomechanical properties, and microtribological behavior of the enamel surface, the anti-erosion mechanisms of the two hydrocolloids are analyzed.

2 Materials and methods

2.1 Specimen preparation

All enamel specimens used in this study were prepared from freshly extracted human mandibular third permanent molars, aged from 18 to 35 y. All the teeth, which were caries-free and had no obvious wear or microcracks on their surfaces, were collected from dental clinics in Chengdu, China, in conformity with the ethical standards of the Chinese Psychological Society, and were kept in deionized water at 4 °C to avoid dehydration before use.

Enamel specimens were prepared according to the method described in Ref. [15]. After tooth root was removed, each tooth was cut into 3–6 parts along its division lines using a diamond saw, and then each part was embedded vertically into a stainless steel mold with denture acrylic resin. The embedded blocks were ground and polished with water cooling to obtain flat testing surfaces with an approximately 2 mm × 2 mm enamel window, and the surface roughness Rₐ was controlled to be less than 0.1 µm with a profilometer (TALYSURF6, England). For each specimen, only 0.2–0.3 mm of height was removed by grinding and polishing, and the obtained testing surface was similar to the original enamel surface. Enamel crystals and prisms have a different orientation on the cusps and in the inner part of the teeth, and thus efforts were made to keep the exposed testing surface in the outer zone of the enamel.
A citric acid solution (CAS, pH = 3.20) was used as a control acidic medium. Citric acid solutions modified with food hydrocolloids were obtained by adding 0.03% \( w/v \) Xanthan gum (CAS-XG) and 0.03% \( w/v \) Arabic gum (CAS-AG), respectively. The pH values of the two modified acidic solutions were adjusted to 3.20 with sodium hydroxide.

Enamel specimens were divided into four groups according to different erosion treatments. The first group was obtained without any treatments, which was referred to as the original enamel. The other three groups were immersed in 100 ml of CAS, CAS-XG, or CAS-AG, and slowly stirred for 10 min at 37 °C, which are referred to as the CAS eroded enamel, the CAS-XG eroded enamel, and the CAS-AG eroded enamel, respectively. For each group, three specimens were selected to conduct surface morphology examination using atomic force microscopy (AFM) (Cypher, Oxford Instruments Asylum Research Inc., England) and scanning electron microscopy (SEM) (QUANTA200, FEI Corp., England), eleven specimens were used to do nanoindentation tests, and eleven specimens were used to do nanoscratch tests. Efforts were made to keep the age, class, type, wear rank, and test region of enamel specimens the same or similar to minimize the individual difference among specimens.

2.2 Characterization of nanomechanical and microtribiological properties

The nanomechanical properties of enamel surfaces were measured using a nanoindentation tester (G200, Agilent Technologies Inc., USA). All indentations were made using a Berkovich diamond tip with a radius of 20 nm. For each specimen, 16 indentations were made in constant load mode under a normal load of 10 mN to measure the surface hardness and elastic modulus. Three indentations were also made in continuous stiffness mode to examine the thickness of the softened layer, and the indentation depth where the rapid increase in hardness ended was defined as the softened layer thickness [16]. The space between indentations was 30 \( \mu \)m. The choice of these parameters was based on previously publish studies. Generally, the indent spacing should be at least 10 times the indentation depth for the Berkovich tip to avoid interference between indents [17].

Unidirectional microtribological tests were conducted in constant load mode on the surfaces of enamel specimens using a nano-scratch tester (G200, Agilent Technologies Inc., USA). A conical diamond tip with a radius of 5 \( \mu \)m was used. The applied normal load was 10 mN, the scratch length was 200 \( \mu \)m, and the scratching speed was 6 \( \mu \)m/s. Three scratches were made on each specimen, and the space between scratches was 50 \( \mu \)m. The profiles of the scratch grooves were measured by a three-dimensional (3D) surface profilometer (NanoMap-D, AEP Technology, USA). Wear loss was calculated according to the scratch profile. All the indentation and scratch tests were conducted at 55% RH and 25 °C.

3 Results

Enamel specimens subjected to different erosion treatments were first examined by AFM, and the typical three-dimensional micrographs of eroded surfaces before and after a deionized water wash are shown in Fig. 1. An apparent honeycomb-like structure is evident on the surface of the CAS-eroded enamel, and the water wash has no influence on the surface morphology. A honeycomb-like structure is lightly visible on the surface of the CAS-AG-eroded enamel, and it becomes clear after the water wash. The CAS-XG-eroded enamel surface is uniform and compact before the water wash, while a very light honeycomb-like structure appears after water wash. Clearly, both the Arabic gum and Xanthan gum that were added to the CAS were adsorbed onto the enamel surface, but the adsorption of Xanthan gum was more uniform and denser.

The water-washed eroded enamel surfaces were also examined by SEM. The original enamel surface without erosion treatment is used as a control. As shown in Fig. 2, the CAS-eroded enamel surface is characterized by an apparent honeycomb-like structure. In addition, the HAP nanofibers in the enamel rods are unordered and marked lacunae appear between them. Both the honeycomb-like structure and the lacunae between the nanofibers are reduced with the addition of the two food hydrocolloids, especially Xanthan gum,
and the nanofibers have an orderly alignment along the rod axis. The nanofibers on the CAS-AG-eroded enamel surface are loosely arranged, while they tend to be tightly aligned on the CAS-XG-eroded enamel surface. In addition, the diameters of the nanofibers have different distributions on the two eroded enamel surfaces, as shown in Fig. 3. The average fiber diameter is approximately 157 nm on the CAS-AG-eroded enamel, which is much larger than that on the CAS-XG-eroded enamel (approximately 56 nm).

The mean values of the nanoindentation hardness and elastic modulus of the original and the three eroded enamel surfaces with their standard deviations and one-way analysis of variance (ANOVA) result are listed in Table 1. Each mean value was obtained by the average of eleven specimens under a normal load of 10 mN. One-way ANOVA reveals significant differences in the hardness and elastic modulus for the four surfaces ($P < 0.005$). Erosion for 10 min in the CAS causes a substantial decrease in the hardness and elastic modulus of the enamel surface, and the decrease is abated with the addition of either Arabic gum or Xanthan gum, but especially Xanthan gum. Paired t-test analysis reveals no significant difference between the elastic modulus of the CAS- and CAS-AG-eroded enamel surfaces ($P < 0.005$), as shown in Table 2.

For the original and the three eroded enamel surfaces, typical curves describing the variation of nanoindentation hardness with indentation depth are shown in Fig. 4. As the indentation depth
Table 1  Nanoindentation hardness and elastic modulus of enamel surfaces subjected to different erosion treatments and one-way ANOVA result.

| Surface                  | Original   | CAS eroded | CAS-AG eroded | CAS-XG eroded | F     | P        |
|--------------------------|------------|------------|---------------|---------------|-------|----------|
| Hardness (GPa)           | 5.21 ± 0.21| 1.04 ± 0.15| 1.26 ± 0.13   | 2.15 ± 0.36   | 752.6 | <0.005   |
| Elastic modulus (GPa)    | 109.3 ± 5.0| 76.5 ± 5.9 | 80.7 ± 4.2    | 90.3 ± 5.9    | 83.07 | <0.005   |

Table 2  Comparison of the hardness and elasticity modulus of enamel surfaces subjected to different erosion treatments by paired t-test (P < 0.005).

| Surface                  | Original   | CAS eroded | CAS-AG eroded | CAS-XG eroded | Hardness | Modulus depth | Hardness | Modulus |
|--------------------------|------------|------------|---------------|---------------|----------|---------------|----------|---------|
| Original                 | 53.60a     | 14.03a     |               |               | 24.16a   |               | 6.649a   | 7.661a  |
| CAS eroded               | 53.60a     | 14.03a     |               |               | 52.57a   | 14.51a        | -3.557a  | -1.915b |
| CAS-AG eroded            | 52.57a     | 14.51a     | -3.557a       | -1.915b       | 24.16a   | 8.131a        | -9.367a  | -5.475a |
| CAS-XG eroded            | 24.16a     | 8.131a     | -9.367a       | -5.475a       | 52.57a   | 14.51a        | 6.649a   | 7.661a  |

Note: a—Significant difference; b—Non-significant difference

As shown in Fig. 3, the hardness of the original surface decreases initially and then becomes almost constant. For the three eroded surfaces, the hardness is extremely low (near zero) at the early stage, then increases rapidly, and finally increases slowly to a saturation value. The indentation depth where the rapid increase in hardness ends varies as CAS-XG < CAS-AG < CAS. This means that the softened layer thickness on the three eroded surfaces varies as CAS-XG < CAS-AG < CAS. The saturated hardness value of the CAS-XG-eroded surface is the highest, followed by the CAS-AG-eroded surface and the CAS-eroded surface.

Figure 5 gives the friction curves and average friction coefficients of the four enamel surfaces.
Friction curves and average friction coefficients of the enamel surfaces subjected to different erosion treatments at applied load of 10 mN: (a) variations of friction coefficients versus displacement; (b) average friction coefficient.

under a normal load of 10 mN. Compared with the original surface, 10 min of erosion causes the friction coefficient to fluctuate and increase clearly, while the fluctuation and increase are abated with the addition of either Arabic gum or Xanthan gum, but especially Xanthan gum. One-way ANOVA reveals a significant difference in the mean friction coefficient of the four enamel surfaces ($P < 0.005$).

Figure 6 illustrates the profiles and wear losses of scratches on the four enamel surfaces. Both the width and depth of the scratches increase after 10 min of erosion. However, compared with the CAS-eroded enamel surface, the CAS-AG- and CAS-XG-eroded surfaces have narrower and shallower scratches, especially the CAS-XG-eroded enamel surface.

Figure 7 gives the typical SEM morphologies of the scratches on the four enamel surfaces. The scratch on the original enamel surface is very shallow, and no obvious wear particles are observed. The scratch is clearly exacerbated on the enamel surfaces subjected to the 10 min erosion treatment, and wear particles appear on the edges of the scratches. Compared with the CAS-eroded enamel surface, the scratches on both the CAS-AG- and CAS-XG-eroded enamel surfaces are slighter, especially the CAS-XG-eroded enamel surface.

4 Discussion

The aim of this in vitro study is to investigate the protective effects of food hydrocolloids on the anti-wear performance of acid-eroded enamel. The pH values of most commercially available acidic
beverages range from 2.7 to 4.2 [18]. Citric acid is commonly used in the beverages to provide fruit tastes and flavors; however, its potential to erode dental hard tissue has been affirmed repeatedly in the past decade [15, 16, 19]. Numerous previous studies concerning dental erosion used a citric acid solution (pH = 3.20) as the demineralizing agent. Arabic gum and Xanthan gum are common food-approved polymer additives and widely used as stabilizers or thickeners in food products. High concentrations of additives are often associated with a change in the taste of drinks. In the present study, therefore, a citric acid solution (pH = 3.20) is used as an acidic medium, while 0.03% w/v Xanthan gum and Arabic gum, which is far lower than the maximum addition level in commercially available soft drinks in China, are used as additives. Considering that the thickness of softened layer caused by 10 min of erosion is around 1–2 µm on the surface of the enamel [16], the nanomechanical properties and microtribological behavior of enamel surface were investigated by the nano-indentation/scratch technique, which is a useful experimental method for assessing the mechanical properties and wear resistance of surface layers [20].

Acid attack can result in enamel surface demineralization through HAP dissolution, and thus the surface morphology of acid-eroded enamel is generally characterized by a honeycomb-like structure. The more significant the surface demineralization is, the more obvious the honeycomb-like structure is. It is evident that under the same erosion condition, both the CAS-XG- and CAS-AG-eroded enamel surfaces have a less visible honeycomb-like structure as compared to the CAS-eroded enamel surface (Fig. 2), and the CAS-XG-eroded surface is much more compact than the CAS-AG-eroded surface. Additionally, compared with the CAS-eroded enamel surface, both the CAS-XG- and CAS-AG-eroded enamel surfaces have higher mechanical properties, lower friction coefficients with smaller fluctuations, and slighter wear, especially the CAS-XG-eroded enamel surface (Table 1, Figs. 5–7). Clearly, compared with Arabic gum, Xanthan gum inhibits surface demineralization more effectively to significantly reduce the influence of erosion on the mechanical properties and tribological behavior of enamel.

Once the enamel contacts acids for a few minutes, a multilayer structure consisting of an outer softened layer and inner transition layer will form on the enamel surface due to HAP dissolution [16]. The softened layer contains a completely demineralized layer and a partially demineralized layer. The completely demineralized layer is primarily composed of the residual organic substances, and its hardness is near zero, while the hardness of the partially demineralized layer increases rapidly with indentation depth (Fig. 4). The transition layer is characterized by less demineralization and a slight reduction in the mechanical properties, and its hardness increases slowly to a saturation value as the indentation depth increases. Generally, a thin softened layer and high saturated hardness correspond to a weak impairment of surface mechanical and tribological properties. Under the same erosion conditions, the softened layer thickness and saturated hardness are strongly associated with the demineralization potential and permeability of acidic media [15, 16]. In the present study, the three erosive agents, CAS, CAS-AG, and CAS-XG, have the same acid type and concentration, and thus their demineralization potentials should be approximately the same or similar. However, on the three eroded enamel surfaces, the softened
layer thickness is different (CAS-XG < CAS-AG < CAS), as is the saturated hardness (CAS-XG > CAS-AG > CAS) (Fig. 4), suggesting that the permeability of the three erosive agents into the enamel is different. Therefore, apart from inhibiting surface demineralization, the anti-erosion mechanisms of Arabic gum and Xanthan gum are also related to inhibiting the permeation of citric acid into the enamel.

In human tooth enamel, the enamel rod is the basic structural unit, and it consists of tightly packed HAP nanofibers which are assembled by HAP crystals [1]. The adsorption of molecules onto HAP usually proceeds via ion exchange [14], and the charge distribution on the adsorbed molecules and HAP surface may play an important role in determining which molecules are adsorbed and which are not. HAP is accepted to have a positive or near-neutral surface charge at neutral pH. It was reported that pH affects the charges on the food hydrocolloids (or food gums) and the HAP and hence, affects the adsorption of hydrocolloids [14]. A low pH increases the positive charge on the HAP surface and advances the adsorption of gums. Thus, in the present study, both the Arabic gum and Xanthan gum, which were added to a citric acid solution with a pH of 3.2, were adsorbed onto the enamel surface (Fig. 1). However, the adsorption of Xanthan gum was more uniform and denser than that of Arabic gum. The differential adsorption of the two food hydrocolloids is responsible for their different protective effects on dental erosion.

The Xanthan gum molecule has a very high molecular weight and a spiral structure; therefore, its aqueous solution has a high viscosity, even at a low concentration. Xanthan gum can bind to the cations (Ca\(^{2+}\), Mg\(^{2+}\), Mn\(^{2+}\), Fe\(^{3+}\), Zn\(^{2+}\), etc.) in an aqueous solution and form a semirigid net-like structure of chelate-like complexes [21]. The calcium ions in HAP have a stronger tendency to be dissolved in an aqueous solution than the phosphate anions. Thus, the calcium ions at the interface of the enamel and aqueous solution can act as a bridge not only between the enamel surface and Xanthan gum molecules, but also between the Xanthan gum molecules. When the enamel contacts the citric acid solution modified with Xanthan gum, a uniform and dense colloidal layer, which consists of the chelate-like complex with a net-like structure, is formed on the surface of enamel. The colloidal layer acts as a barrier to prevent surface demineralization and the permeation of citric acid into the enamel, and thus protects the enamel surface from acid attack. Apart from this, the reduction in the fluidity of the citric acid solution caused by the high viscosity of Xanthan gum also contributes to this protection [22]. It was reported that decreasing the mobility of an acidic solution reduces HAP dissolution [23]. Therefore, adding a trace amount of Xanthan gum can effectively reduce the erosive potential of a citric acid solution, and thus reduce the influence of erosion on the mechanical and tribological properties of enamel.

In contrast to Xanthan gum, Arabic gum is a complex proteoglycan containing 2 wt% protein components, and it has high hydrophilicity and excellent lipophilicity. Its aqueous solution is generally characterized by low viscosity, even at a high concentration [24]. It has been reported that, in aqueous solution, Arabic gum is adsorbed onto positively charged HAP nanoparticles through electrostatic and hydrogen-bond interactions [25]. Additionally, because of its excellent lipophilicity, Arabic gum is easily adsorbed onto the organic matter in the enamel. Considering that the interface between HAP crystals and organic matter in the enamel acts as a channel for the acid agent during dental erosion [26], adding Arabic gum can prevent the permeation of citric acid into the enamel. However, because of their spherical molecular structure, Arabic gum molecules are difficult to stretch, and therefore will not form a net-like structure through intermolecular cross-bonding. It should be noted that the HAP nanofibers, whose mean diameter is 68 nm in sound human tooth enamel [27, 28], have a smaller diameter on the CAS-XG-eroded surface (a mean diameter of 56 nm) but an larger diameter on the CAS-AG-eroded surface (a mean diameter of 157 nm), as shown in Figs. 2 and 3. The decreased fiber diameter on the CAS-XG-eroded surface is a result of the partial dissolution of HAP fibers, while for the CAS-AG-
eroded surface, the increased fiber diameter results from the adhesion of Arabic gum to the fibers. Therefore, it could be inferred that in contrast to Xanthan gum, Arabic gum tends to be adsorbed on the HAP nanofibers rather than form a uniform and dense colloidal layer on the enamel surface. The less uniform and dense adsorption of Arabic gum results in a weak resistance against the permeation of an acid solution into the enamel, and thus the anti-erosion effect of Arabic gum is inferior to that of Xanthan gum. As a result, the friction coefficient and wear loss of the CAS-AG-eroded surface are higher than that of the CAS-XG-eroded surface (Figs. 5 and 6). It appears that the protective effects of food hydrocolloids on dental erosion largely depend upon their adsorption states.

Tooth wear caused by erosion is becoming more of an issue as life expectancy and acidic beverage consumption increase. Therefore, dental erosion is a prominent clinical concern, and an understanding of the protective mechanisms and measures in enamel erosion is critically important. The results of this study suggest that the impairment of the nanomechanical and microtribological properties of enamel surfaces by acid erosion are mitigated more significantly by adding trace amounts of Xanthan gum than Arabic gum. The protective effects of food hydrocolloids largely depend upon their adsorption states on the enamel surface, and a uniform and dense adsorption benefits erosion prevention. These findings extend the understanding of the anti-erosion mechanism of food hydrocolloids and help to advance their application in dental erosion prevention. It should be noted that the present study does not consider the effect of saliva. Once acid agents are introduced into the mouth, saliva secretion would accelerate to act as a buffer to the acids. In addition, the proteins in saliva can be adsorbed by physisorption onto human teeth and form salivary pellicle. The pellicle is considered to play an important modifying role in enamel surface demineralization because its permselective nature restricts acid diffusion and the transport of ions in and out of the enamel surface [29]. Our future studies will explore the synergistic action of saliva and food hydrocolloids.

5 Conclusions

The protective effects of two food hydrocolloids, Xanthan gum and Arabic gum, on dental erosion were investigated from the perspective of the nanomechanical properties and microtribological behavior of acid-eroded enamel in this paper. Within the limitations of the present study, the main conclusions can be summarized as follows:

1) Compared with Arabic gum, Xanthan gum inhibits enamel surface demineralization and acid permeation more effectively owing to its more uniform and denser adsorption on the surface of enamel. Therefore, the loss of hardness and elastic modulus and the decrease of wear-resistance of enamel surface by acid erosion are mitigated more significantly by adding trace amounts of Xanthan gum than Arabic gum.

2) Adding trace food hydrocolloids reduces the surface demineralization of enamel and inhibits acid permeation, which mitigates the impairment of the mechanical and tribological properties of the enamel surface caused by erosion. The adsorption state of food hydrocolloids is the determining factor in the permeability of acid agents into the enamel and plays a significant role in preventing dental erosion.

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