

A Study on the Amount of Fluoride that Remains in the Oral Cavity based on the Number of Times that Rinsing Is Done as Determined Using Nuclear Magnetic Resonance Spectroscopy

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The most common method for children's oral health management is brushing the teeth, and for this, toothpaste is often used. Most of the toothpaste brands that are currently in the market, however, contain fluoride, which children should not swallow. This poses a problem for children as they have difficulty regulating their swallowing reflex and may thus end up swallowing the toothpaste with its fluoride content. Therefore, using ¹⁹F NMR spectroscopy, this study analyzed the amount of fluoride left in the oral cavity after brushing the teeth depending on the number of times that rinsing is done. It was shown that brushing the teeth using toothpaste containing 0.24 % NaF does not pose any risk and is safe even when rinsing just two-times.

Keywords : Nuclear magnetic resonance (NMR) spectroscopy, toothpaste, fluoride, rinsing, brushing

1. Introduction

Baby teeth start emerging at around 6-7 months old and are expected to be replaced by permanent teeth after about 6 years of mixed dentition from 6 to 12 years old [1]. Early childhood caries, used as a comprehensive term of late indicating nursing bottle caries and multiple caries in toddlers and children, starts at the same time as the eruption of the teeth, and progresses very fast [2].

There are various ways of applying fluoride in the oral cavity for the protection of the teeth, including drinking water fluoridation, specialist fluoride local application, and the use of a fluoride mouth rinsing solution, a fluoride tablet, and fluoride toothpaste. Among these, the use of fluoride toothpaste is known worldwide to be significantly effective in decreasing dental caries [3]. It has been reported that the use of fluoride toothpaste decreases dental caries experience by 15-30 % and the incidence of dental caries by about 24 % [4, 5]. It has also been reported, however, that toothpaste containing sodium fluoride (NaF) provides much more effective dental caries prevention [6]. Based on the study results reporting that

the use of fluoride toothpaste is effective for dental caries prevention, fluoride toothpaste has been widely used. Also, based on the regulation on indicating a product's fluoride content, the total fluoride contents of different fluoride toothpaste brands are indicated [7].

When fluoride exists as an ion (F⁻) on a biofilm, it prevents demineralization and promotes remineralization. Therefore, fluoride in an ion state, or soluble fluoride, has a dental caries prevention effect when included as an ingredient in toothpaste [8]. For these reasons, while the total fluoride content of toothpaste is important, so is the degree of ionization. Therefore, a dental caries prevention effect can be expected when the toothpaste used for brushing the teeth contains a sufficient amount of soluble fluoride [9, 10].

It has been reported, however, that side effects might occur when the use of fluoride is abused while the appropriate use of fluoride can show a dental caries prevention effect [11]. It was reported that 72 % of the toothpaste that was used for brushing the teeth of a 30-month-old child remained in the child's oral cavity and was swallowed by the child. It was also suggested, based on a study that was conducted, that pea-sized toothpaste be used for brushing the teeth of children less than 7 years old, and that care be exerted to prevent children from swallowing the toothpaste [12]. Moreover, it was reported

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that there is a risk involved in using fluoride toothpaste for brushing the teeth of children less than 6 years old due to their poorly regulated swallowing reflex. Specifically, it can become a risk factor for spotted teeth in children less than 3 years old [13].

The most commonly used method of measuring the amount of fluoride ion is that using fluoride ion electrode [14]. Such method, however, responds to the activity of the ion rather than to the fluoride ion concentration, and has a limitation in that measurement is difficult when the fluoride ion is less than 10^{-6} M [15]. Nuclear magnetic resonance (NMR) spectroscopy is a method that allows non-invasive quantitative analysis and has been generally used of late [16]. Fluorine (^{19}F) NMR spectroscopy can be employed as a powerful tool for analyzing the selective and sensitive fluorinated species [17].

As of this writing, no studies have been conducted to measure the amount of fluoride present in fluoride toothpaste, the amount of fluoride that can be swallowed when brushing the teeth, and the amount of fluoride that remains in the oral cavity after brushing. Therefore, the purpose of this study was to measure the amount of fluoride that remains in the oral cavity depending on the number of times that rinsing is done after brushing the teeth of a child using children's fluoride toothpaste, through ^{19}F NMR spectroscopy.

2. Materials and Methods

2.1. Sample preparation

In this study, fluoride toothpaste containing 0.24 % NaF (0.15 % w/v F ion) for infants and preschoolers (Colgate-Palmolive Company, New York, NY, USA) was used. The subjects' teeth were brushed for 3 minutes using toothpaste containing 0.1 mg fluoride and children's toothbrushes. Right after brushing, the remaining fluoride toothpaste and saliva in the oral cavity were collected. The 5 ml sterilized distilled water that was used for the first rinsing was collected, followed by the 5 ml sterilized distilled water that was used for the second rinsing. Furthermore, saliva was collected to check the amount of fluoride that remains in the oral cavity after two-time rinsing.

2.2. NMR measurement

^{19}F NMR spectroscopy measurements were carried out with an ECZR NMR spectrometer (400 MHz FT-NMR spectrometer, JNM-ECZ400S/L1, JEOL Ltd., Tokyo, Japan) operating at 376.17 MHz and equipped with a dedicated 5 mm spinning probe (Fig. 1). The probe temperature was 23 °C. The typical spectral parameters for this study were



Fig. 1. (Color online) The 400 MHz FT-NMR spectrometer that was used in this study.

as follows: 90° pulse width, 6.74 μs; relaxation delay, 5 s; and acquisition time, 83.88 s. A known amount of D₂O (100 μl) was added as an internal field frequency lock.

The NMR spectrometer calculated the spectral resonance frequency (ν_0) using the formula below.

$$\nu_0 = \frac{\gamma}{2\pi} B_0$$

γ = gyromagnetic ratio

B_0 = Magnetic Field strength

And Chemical shift (ppm) were used the following formula.

$$\text{Chemical shift (ppm)} = \frac{\nu_i - \nu^{ref}}{\nu_0} \times 1,000,000$$

ν_0 = The resonance frequency of the chemical bonds that are not nuclear

ν_i = The resonance frequency of each element in the molecule

ν^{ref} = The reference frequency

3. Results

3.1. Fluoride analysis via ^{19}F NMR spectroscopy

The analysis of the ^{19}F NMR spectrum showed a range of –122.27 to –120.56 ppm (Fig. 2). These peaks represented the amount of fluoride contained by the toothpaste

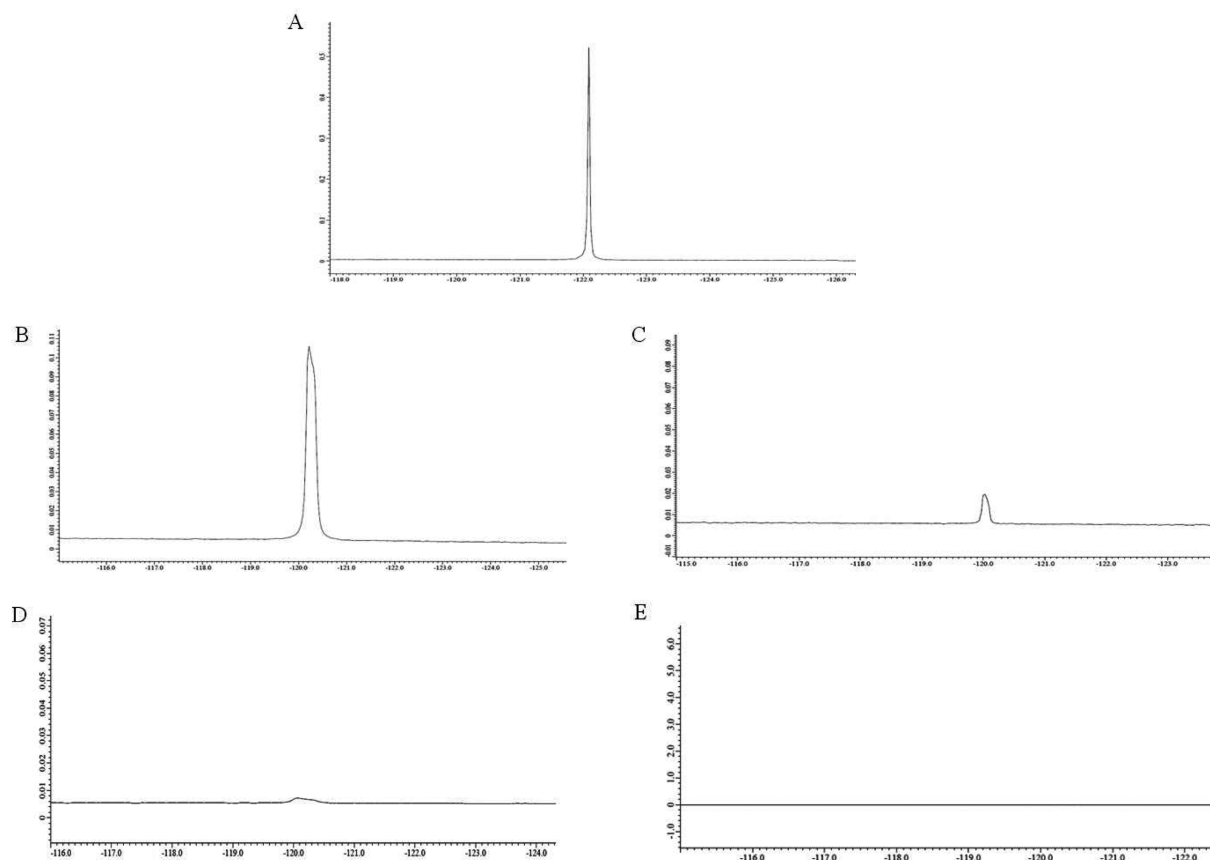


Fig. 2. (A) Toothpaste containing 0.24 % NaF (0.15 % w/v F ion). (B) Amount of fluoride in the oral cavity right after 3-minute brushing. (C) Amount of fluoride in the oral cavity after one-time rinsing. (D) Amount of fluoride in the oral cavity after two-time rinsing. (E) Amount of remaining fluoride in the oral cavity after the second rinsing.

that was used for brushing the subjects' teeth, and the amount of fluoride that remained in the oral cavity based on the number of times that rinsing was done. The total areas represent the quantified number that was a range of

areas by peak (Fig. 3). It was confirmed that the fluoride concentration was almost maintained when brushing was done for 3 minutes using fluoride toothpaste, and it was measured as 0 % for both one- and two-time rinsing. The amount of fluoride that remained in the oral cavity after two-time rinsing following the brushing of the teeth was 0 %. Thus, it was confirmed that there was no remaining fluoride in the oral cavity after one- and two-time rinsing following the brushing of the teeth.

4. Discussion

The most commonly used oral health management method for children is brushing the teeth. For this, toothpaste is used, and most toothpaste brands contain fluoride [18]. It is well known that fluoride is the most important anti-cavity ingredient of toothpaste. The amount of fluoride in the toothpaste varies, and it is usually included in the form of sodium fluoride and sodium monofluorophosphate [19]. According to a report by White [20], it was shown that NaF-containing toothpaste prevents the development

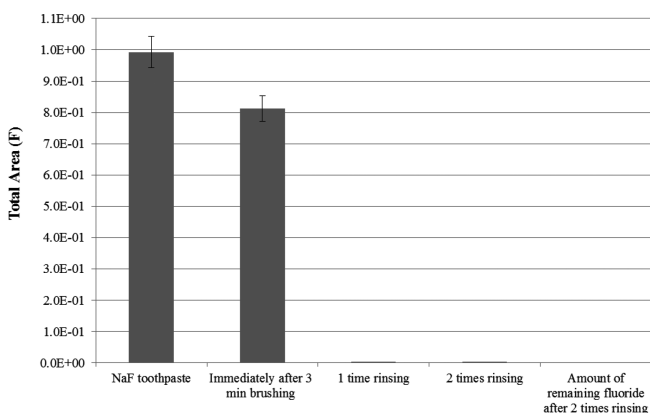


Fig. 3. Mean F levels at the total peak area based on the use of fluoride toothpaste, the number of times that rinsing was done, and the fluoride remaining in the saliva in the oral cavity determined using ¹⁹F NMR spectroscopy.

of dental caries and remineralizes teeth with early caries more than sodium monofluorophosphate does. This study thus used NaF-containing toothpaste.

Fluoride toothpaste can prevent dental caries but can also cause dental fluorosis, and it has been reported that increasing dental fluorosis is mostly associated with the inappropriate use of fluoride toothpaste [21-23]. It has been reported that each time children brush their teeth using toothpaste containing 1 mgF/g fluoride, they swallow 0.12-0.38 mg of the fluoride, which can cause dental fluorosis because much of the swallowed fluoride is absorbed by the body [24]. Although fluoride clearly effectively prevents dental caries at an appropriate concentration, a possibility of children's fluoride exposure exists when they brush their teeth using fluoride toothpaste. Therefore, the number of times that rinsing is done after brushing should be indicated. Thus, in this study, the amount of fluoride that remains in children's oral cavity based on the number of times that rinsing is done, using toothpaste containing 0.24 % NaF, was analyzed, and the amount of fluoride that can be swallowed by children when brushing their teeth was determined by measuring the amount of fluoride that remains in the oral cavity after rinsing.

A study on brushing the teeth reported that 5 % of children less than 2.5 years old, 32 % of children aged between 2.5 and 4 years old, and 27 % of children above 4 years old brushed and rinsed their teeth but swallowed all or almost all of the water that they used for brushing and rinsing [25]. Burt [26] insisted that the number of times that rinsing is done after brushing the teeth should be increased to lessen the amount of toothpaste left in the oral cavity. It was found in this study, however, that 0 % fluoride exists in the oral cavity right after 3-minute brushing without rinsing, showing a decrease in fluoride concentration due to dilution from the new saliva. Therefore, this study showed that there is a possibility of fluoride exposure on the part of children who do not rinse their mouths after brushing their teeth, and who swallow the water that they used for brushing and rinsing their teeth. For the changes in the amount of fluoride that remains in the oral cavity by number of times that rinsing is done, 0 % remained after both one- and two-time rinsing. In other words, no fluoride was detected in the oral cavity after rinsing. This shows that a risk is posed by children's poor management or regulation of the swallowing reflex when brushing their teeth; thus, children should be prevented from swallowing toothpaste. Rinsing at least once reduces the remaining fluoride in the oral cavity to 0 %, which shows that rinsing must be done at least once. This will prevent dental caries despite water and food intake as

no fluoride will remain in the body.

Based on the above study results, it was confirmed that one-time rinsing does not pose a risk of fluoride exposure when brushing the teeth using toothpaste containing 0.24 % NaF, and that there is no risk to the body even when the rinsing water is swallowed after at least two-time rinsing. It is considered that accurately measuring the amount of fluoride that remains in the oral cavity after brushing and rinsing can provide basic data on the number of times that rinsing must be done by children, which can lead to the improvement of children's oral health through safe brushing.

5. Conclusion

Fluoride toothpaste is widely known to have a dental caries prevention effect, but it poses a risk when used for brushing children's teeth. It was found in this study that even just one-time rinsing can reduce the amount of fluoride that remains in the oral cavity by 99.99%. Therefore, simple, safe, and effective brushing using fluoride toothpaste can promote oral health if rinsing is done at least once.

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