Fluoride Assay Methodology for Carbonated Beverages

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ABSTRACT

Purpose: The purpose of this paper was to review different methodological techniques used for the assessment of fluoride in carbonated beverages, and compare results using a fluoride ion electrode direct read method with and without a prior decarbonation treatment.
Methods: The carbonated beverages in this study were either purchased locally at grocery stores in Iowa City, Iowa, or purchased as part of a national representative sampling approach included in the National Fluoride Database and Intake Assessment Study (NFDIAS). The samples were compared with and without a decarbonating process. Soda pop and beer samples were analyzed by removing a 1-ml sample and adding a 1-ml buffer solution. The fluoride concentration of the sample and buffer combination was then determined using a fluoride ion specific electrode.
Results: There was no significant difference in the fluoride concentration of the samples with or without prior decarbonation. The mean absolute difference between the soda pop group with and without decarbonation was 0.01 ppm F, while results from the beer samples showed variation of 0.00 to 0.02 parts per million fluoride (ppm F). These differences were not statistically significant for the soda pop or beer groups (P=.50 and P=.74, respectively).
Conclusion: Whether or not decarbonation was conducted prior to analysis, the fluoride assay results were the same. Therefore, decarbonation of soda pop and beer was deemed unnecessary prior to fluoride analysis. (J Dent Child 2006;73:136-139)
KEYWORDS: METHODOLOGY, FLUORIDE, CARBONATED BEVERAGES

The role of fluoride in the prevention of dental caries is well documented.[1] Studies have also shown an increased prevalence of dental fluorosis in children, however, as well as adverse effects on bone health from very high fluoride levels.[2] Because of this, it is important to examine fluoride intake from all sources and for all age groups. This applies not only to intake dentifrices, rinses, and gels, but also from water and various types of beverages and foods. A major challenge in assessing need and appropriate dosage for dietary fluoride supplementation for children is estimating the total fluoride intake amount. One main challenge is the lack of information available on fluoride levels of foods and beverages. The fluoride content of waters, beverages, and foods is not required on labels and is not readily available otherwise.

Since soda pop consumption now plays a bigger part in the diets of children and adolescents[3] and these beverages replace more nutritious drinks such as milk and fruit juices, concern over their impact on dental health has increased. This increased trend in soda pop consumption may also lead to excessive energy intake, and may contribute to childhood obesity[3] and dental health problems such as caries. Fluoride intake from this increased soda pop consumption could be an important part of total fluoride intake, so ongoing assessment of fluoride content of these beverages is warranted.

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Because commercially available nutrition databases/software have not featured a fluoride component, researchers interested in assessment of fluoride intake have had to develop their own systems for purchase and analysis of fluoride levels in foods and beverages. To address this gap, the National Fluoride Database and Intake Assessment Study (NFDIAS) was designed.[4] Its goal was to provide estimates of fluoride levels of selected beverages and foods from a national sample of such products and to design tools for estimation of nondietary fluoride intake. NFDIAS data includes fluoride assay levels found in waters, juices, soda pops, beers, other beverages, infant foods, and other foods.

As a part of this study, carbonated soda pops and beers were collected according to a nationally representative sampling approach.[5] This approach was modeled after an existing stratified sampling approach successfully implemented at the US Department of Agriculture (USDA) for several years for sampling of retail foods and beverages. The number of sample pickup locations was expanded to address variability among geographic areas and from different distribution points. These samples were then analyzed for fluoride content, and the results from these beverages were incorporated into the national fluoride database.[4] This database is available online at www.nal.usda.gov/fnic/foodcomp and now allows access to national fluoride estimates for different beverages and foods that were not previously available.[6] This is important not only to the dental community, but also to medical practitioners and research investigators.

Current methods of measuring fluoride involve the use of an ion-specific electrode. For liquid samples, a direct read method is used instead of microdiffusion followed by electrode measurement, which is necessary with solid samples. With this direct read method, it is essential that the solution to be measured falls within a pH range of 5 to 6.[7] The use of one buffer—Total Ionic Strength Adjustment Buffer, for example—to adjust the pH and ionic strength is addressed in a paper by Frant and Ross.[8]

Due to a lack of clarity in the literature concerning appropriate methods for the measurement of carbonated beverages, different analytical approaches were compared. Previous studies on fluoride concentrations of carbonated soda pop generally have included a decarbonating process prior to fluoride analysis. Some of the different preparations include partial decarbonating by exposure to air for at least 6 hours,[9] or loosely covering 30 ml of the beverage and leaving the sample for 12 hours before analysis.[10] Shannon measured 0.5 oz of the product and placed the sample in a test tube for 45 minutes at 70ºC.[11] These methods have been compared to a more complex procedure of heating the beverages to 70ºC and then bubbling nitrogen gas through the sample for 5 minutes.[12] The simpler method, when compared by Schultz et al without prior heating and bubbling nitrogen gas, resulted in the same pH and fluoride concentration measurements found when using the heating and nitrogen bubbling technique.[10] Heilman et al[13] decarbonated soda pop by placing approximately 5 ml of sample into a plastic vial and loosely covering the vial for at least 12 hours prior to analysis, while Pang et al[14] used a modified method originally described by Taves[15] to analyze several different types of beverages, including soda pop.

Similar techniques were used with beer samples. A study by Warnakulasuriya et al[16] on fluoride levels found in beers placed 5 ml of beer into 5 ml tubes, allowing the solution to stand for 30 minutes prior to analysis (Warnakulasuriya, 2-23-2004, personal communication via e-mail). Martin Delgado et al[17] also used different fluoride methodologies to compare fluoride concentrations in beers as well as soda pops.

PURPOSE

This study was undertaken to evaluate the necessity of decarbonating beverage samples prior to fluoride analysis. A more streamlined analysis without decarbonation would save time, reduce costs, and increase efficiency when a large number of samples need to be analyzed, as with the NFDIAS project that required representative samples from around the country. Therefore, different methodological approaches were undertaken to compare fluoride results with and without a prior decarbonating process.

METHODS

For the purpose of this paper, the authors use the terms "carbonating" and "decarbonating." This refers to the carbonating/decarbonating of soda pops and carbonating/decarbonating or gassing/degassing for beers.

Sixteen different soda pop brands were purchased locally at grocery stores in Iowa City, Iowa, for this study. In addition, 4 blind samples of soda pop, part of the NFDIAS project which had been purchased according to a nationally representative sampling approach, were also included. These samples were:

1. collected in plastic bottles;
2. shipped frozen to Iowa;
3. refrigerated overnight;
4. allowed to thaw;
5. removed; and
6. left at room temperature for 30 minutes before analysis.

The locally purchased soda pop and the blind soda pop samples were then analyzed together. Immediately after opening the 16 soda pop containers and the 4 bottles containing the blind samples, 2 10-ml aliquots from each were placed into separate 15-ml culture tubes. One of the samples from each beverage was read directly and the other sample was left with the cap loosely placed on top of the culture tube and read the following day, thus allowing a 24-hour period for decarbonation. Decarbonation was determined to have occurred after the solution was shaken, and no bubbling was observed.

Although few studies were available on the fluoride content of beer, methodologies similar to soda pop were used for decarbonating beer and, therefore, this study includes a limited number of beer samples.

A 6-pack of beer (Miller High Life, Miller Brewing Co, Milwaukee, Wis) was purchased. From each can, aliquots were removed and decarbonated. The decarbonating was performed by bubbling nitrogen gas through the sample until the emitted bubbles changed texture, from slightly foamy to a "blowing bubbles in milk" texture, after approximately 5 to 10 minutes. Subsequently, aliquots from the same cans were removed and not decarbonated. These samples, with and without decarbonation, were placed into separate 30-ml bottles and frozen until analysis. The thawing process of the samples was handled in the same manner as the soda pop.

Soda pop and beer samples were analyzed by removing 1 ml of sample and adding 1 ml of Total Ionic Strength Adjustment Buffer (TISAB, product no. 94-09-09, Orion Research Inc, Beverly, Mass) to maintain the pH and adjust the total ionic strength. The sample and buffer combination then was stirred by hand prior to analysis. The fluoride concentration was determined using a fluoride ion-specific electrode (model no. 9609 ionplus electrode, Orion Research, Inc) in conjunction with an expandable and programmable Ionanalyzer (model no. 920A+, Orion Research, Inc). Results were reported in parts per million fluoride (ppm F).

Electrode calibration was performed using standards prepared from a sodium fluoride 100-ppm F stock solution (Orion Research, Inc) and deionized water. A serial dilution was used to prepare the standards, which ranged from 0.05 to 2 ppm fluoride. A minimum of 10% of all samples were duplicated and analyzed along with a certified freeze-dried urine reference material (National Institute of Standards and Technology, Gaithersburg, Md). Electrode calibrations were also checked after 1 hour. In addition, different electrodes were used when assaying the duplicate samples.

RESULTS

The fluoride concentrations of the 20 soda pops, with and without decarbonation, and the absolute differences of the 2 are listed in Table 1 along with the means, standard deviations, and medians. When each sample was analyzed with and without decarbonation, the absolute difference for the 9 lower-range fluoride samples (0.05-0.15 ppm F) varied up to 0.01 ppm F. Two mid-range samples (0.31-0.37 ppm F) varied by 0.02 ppm F. The absolute difference for the 9 higher-range fluoride samples (0.49-0.83 ppm F) was 0.01 to 0.03 ppm F. All individual differences were less than or equal to 0.03 ppm F, and all but 2 were less than or equal to 0.02 ppm fluoride. There was no absolute difference (0.00 ppm F) between the means or medians of the 2 groups. A paired *t* test showed no statistically significant differences (P=.50) between the carbonated and decarbonated soda pop.

Similarly, beer sample results showed no significant difference (P=.74) in the mean fluoride concentration between the samples with or without prior decarbonating. The 6 beer samples (fluoride levels=0.28-0.30 ppm F) with decarbonating showed variation of 0.00 to 0.02 ppm F. The decarbonating mean was 0.29 (±SD=0.010) ppm F compared to 0.29 (±0.008) ppm F for the samples without prior decarbonating.

DISCUSSION

Since the results showed no substantial differences between soda pop samples or beer samples that were assayed with or without decarbonating procedures, these treatments were deemed unnecessary prior to analysis. Whether or not these processes were conducted did not have an impact on the assayed fluoride concentrations. Therefore, after reaching this conclusion, the large numbers of samples of soda pop and beer

Table 1. Comparison of Fluoride Assay Results Between Carbonated and Decarbonated Soda Pop

	Results in ppm fluoride		
Sample	**Carbonated**	**Decarbonated**	**Absolute difference**
Ginger ale (Hy-Vee)	0.10	0.09	0.01
Orange (Hy-Vee)	0.11	0.10	0.01
Sierra Mist (Pepsi)	0.71	0.73	0.02
A&W Root Beer (Dr. Pepper/7-Up)	0.15	0.14	0.01
Mountain Dew (Pepsi)	0.83	0.86	0.03
Sierra Mist (Pepsi)	0.71	0.73	0.02
Cola*	0.56	0.54	0.02
Cola*	0.56	0.55	0.01
Cola*	0.62	0.59	0.03
Cola*	0.31	0.33	0.02
Pepsi Twist (Pepsi)	0.56	0.55	0.01
Vanilla Diet Coke (Coca Cola)	0.49	0.48	0.01
Squirt (Dr. Pepper/7-Up)	0.05	0.06	0.01
Mountain Dew Orange (Pepsi)	0.58	0.56	0.02
Red Fusion(Dr. Pepper/7-Up)	0.03	0.03	0.00
Diet Coke (Coca Cola)	0.09	0.10	0.01
Diet Pepsi (Pepsi)	0.08	0.08	0.00
A&W Cream Soda (Dr. Pepper)	0.37	0.35	0.02
Sunkist Orange (Dr. Pepper/7-Up)	0.07	0.07	0.00
Lemon Lime (Wal-Mart Stores, Inc)	0.09	0.08	0.01
Mean±SD	**0.35±0.27**	**0.35±0.27**	**0.00±0.00**
Median	**0.34**	**0.34**	**0.00**

**Unknown brands of cola (blind samples)* for the NFDIAS were analyzed without prior decarbonating, thus saving time and resources.

As with previous studies analyzing soda pop,[13] a range of fluoride levels was observed in the larger samples of beers assayed for the NFDIAS. The range of fluoride levels from 242 beers was from 0.06 ppm F to 0.92 ppm F, with a mean of 0.45 ppm F. This range was consistent with findings from soda pop where the most important determinant in fluoride content was the production site for these beverages and the fluoride level of the water used during production.[13] The continued monitoring of fluoride from all sources remains of great importance when evaluating total fluoride intake.

CONCLUSIONS

Overall, this study's results showed no significant differences in the fluoride content of soda pop or beer samples when comparisons were made with or without a decarbonating or degassing process. Therefore, decarbonation is not necessary prior to fluoride assay.

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