Determination of fluoride in black, green and herbal teas by ion-selective electrode using a standard-addition method

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ABSTRACT

Tea leaves are very rich in fluoride, since tea plants take up fluoride from the soil and accumulate in its leaves. Some of this fluoride is released into the infusion, which is drunk as tea. Fluoride in tea could be beneficial for the prevention of dental caries, but it may result in excessive intake and lead to enamel fluorosis. The purpose of this work was to determine the fluoride levels in 12 different brands and types of tea by means of a computer-controlled ion-selective electrode potentiometry using a standard-addition method. It is a rapid method which showed good accuracy and precision. Fluoride contents of tea infusions after 5 min ranged from 0.95 to 4.73 mg/l for black teas; from 0.70 to 1.00 mg/l for green teas, and from 0.26 to 0.27 mg/l for herbal teas. It was concluded that black teas and green teas examined may be important contributors to the total daily fluoride intake. However, the ingestion of some black teas that were found to have high fluoride content by children at the age of risk to dental fluorosis should be avoided.

Key words*: fluoride, tea, ion-selective electrode, potentiometry*

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INTRODUCTION

Fluoride is the anionic form of fluorine. It is an important anion, since a small amount of fluoride has beneficial effects on the teeth by reducing the incidence of caries.[1] Fluoride interacts with hydroxyapatite by replacing the hydroxyl ions to form a new more crystalline phase. This phase, fluoroapatite, is more resistant to errosion by plaque acid and demonstrates a lower surface energy thus making plaque adhesion more difficult.[1,2] Fluoride may also increase the rate of enamel remineralization, so that calcium and phosphate ions are protected and not lost during demineralization. Moreover, fluoride may reduce oral concentration of cariogenic bacteria or reduce the metabolism of bacteria in plaque.[1,3] For this reason, the intake of fluoride is needed to promote good dental health.

In some countries fluoridated drinking water seems to be the main dietary source of fluoride, and the optimal amount of fluoride for prevention of dental caries has been suggested as 1.0 ppm. Tea is also considered a major source of fluoride, since tea plants *Camellia sinensis* take up fluoride from the soil and accumulate in its leaves. Epidemiological surveys have reported that some populations who drink tea on a regular basis have a reduced number of carious teeth.[4–6] Due to the fluoride content, tea should be an effective vehicle for delivering fluoride to the oral cavity helping to prevent dental decay.

During recent years several papers have been published on the fluoride content of tea leaves.[7–10] However, the results obtained are often in poor agreement. The accuracy and the precision of the analytical method used may give the contribution on the variation of the results. Feldheim and Miehe[11] reported the total fluoride content in tea leaves to range from 40 to 330 ppm. Fung *et al*.[12] reported that the highest fluoride contents are found in fallen leaves. Fluoride accumulated in old leaves over 2000 mg/kg and young leaves ranged from 250 to 300 mg/kg. Some of this fluoride is released into the infusion, which is drunk as tea.[4,5] Chan and Koh[13] reported the fluoride concentration in caffeinated and decaffeinated tea infusion ranged from 0.34 to 3.71 ppm and 1.01 to 5.20 mg/l, respectively. Based on the reported fluoride content from black teas, the benefits of fluoride in tea especially for children have been debated. Some authors agree that fluoride in tea is beneficial since it could account for a significant portion of total dietary fluoride intake. However, the fluoride in tea may result in excessive intake and lead to enamel fluorosis.[1,4] Fluoride may cause mottled teeth at around 1 ppm when it is present in water. For that reason, the determination of fluoride content as a trace element in tea infusions is important in order to assess any possible health hazards.

Determination of fluoride in tea is usually carried out by direct potentiometric methods using an ion-selective electrode. The calculation of the results is obtained using a calibration curve, which shows that the electrode potential is linear to the logarithmic of the ionic acitivity. It is a simple procedure without any sample preparation for measuring aqueous samples. However, any problems appear when the sample matrix is complex, such as tea that contains polysaccharides, volatile oils, vitamins,

minerals, purines, alkaloids, and polyphenols. In this case, the electrode potential is often not linear to the logarithmic of the ionic acitivity due to the interfering molecules and the fluoride contents may be found to be somewhat lower than the levels in real samples.[14,15] To overcome this problem, a standard-addition method should give good accuracy and precision.[16] Therefore, the purpose of this work was to develop an accurate method for the determination of fluoride in infusions of black, green and herbal teas by ion-selective electrode using a computer-controlled standard-addition technique.

MATERIAL AND METHODS

The analytical method used in the present work incorporates a fluoride ion-selective electrode (Metrohm AG, Switzerland). The membrane of the electrode is lanthanum fluoride doped with europeum fluoride that interacts specifically with fluoride ion in the sample, allowing the electrical potential. A double junction electrode Ag/AgCl (Metrohm Model 90–02) was used as reference electrode. For the measurement of the potentials the ion-meter with millivolt readability to ± 0.1 mV (WsA, Würzburg Germany) was used combined with an automatic burette (Dosimat model E655) with stirrer unit model E649 and titration tube (Metrohm AG). The potential values were transmitted every second to a work station.[16] The mean value over a 10s period was calculated and displayed in monitor. The difference between the two mean potential values over a period of 10 s was successively monitored. The equilibrium potential was defined as the mean potential value when the potential difference decreased to within 0.10 mV.

The standard fluoride solutions were made from NaF supra pure reagent. TISAB (Total Ionic Strength Adjustment Buffer) solution contains 58.5 g of sodium chloride, 57.0 ml of glacial acetic acid, 61.5 g of sodium acetate and 5.0 g of CDTA (trans-1,2-cyclohexanediamino N,N, N',N'-tetra acetic acid) which were dissolved in Milli-Q water and diluted to 500.0 ml. pH of this solution was adjusted to 5.0–5.5 with 6 M of sodium hydroxide and diluted to 1000.0 ml with Milli-Q water. The solutions were stored in screw top polyethylene containers.

Stock solutions were made up as 500 ppm from sodium fluoride. Standard solutions of 10 ppm and 100 ppm were prepared by sequential dilution of the stock solution. Both standard and stock solutions were placed in polyethylene bottles.

Twelve different brands and types of tea (see table 2) were purchased from supermarkets in Wuerzburg, Germany. The samples were obtained in the form of a tea bag and fine powder. A sample bag of tea (1.75–2.25 g) or 1.0 g of fine powder was infused in about 90 ml of boiling Milli-Q water for 5 minute. The flask was then gently swirled and the solution was filtered using a 0.45 μm Millipore filter. The solution was then diluted with Milli-Q water up to 100.0 ml and bottled in polyethylene containers. The sample solutions were analyzed immediately by ion-selective electrode potentiometry at 25 ° C. All measurements were done in six replicates.

A 10.0 ml of sample solution was transferred to the titration tube thermostatted at (25.0 ± 0.1) ° C and mixed with 10.00 ml of TISAB solution. The solution was stirred during the analysis. The potential (E_{xo}) was recorded after 3 minute. To the sample solution was then added 1/10 volume of about 10 times more concentrated, buffered natrium fluoride standard solution and the new potential (E_{x1}) was recorded. The addition of the standard solution was repeated six times. The fluoride concentration of the sample was then calculated by Gran's plot.[16–18] By this computer-controlled instrument the standard addition was done in such a way that equidistant intervals of the potential steps to be measured were achieved. The analysis was performed automatically using a computer-controlled potentiometry equipped with WsA software (self developed by Department of Pharmacy, University of Würzburg, Germany).

RESULTS

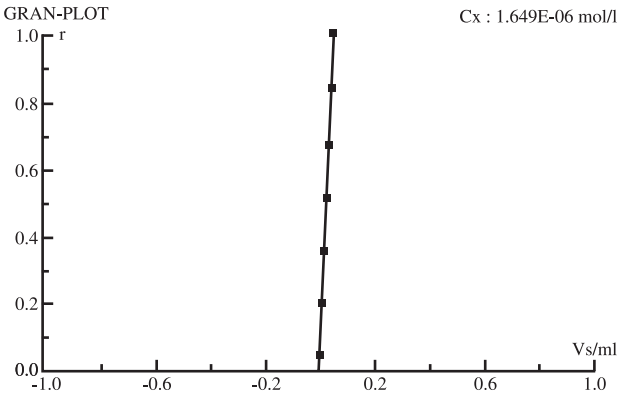

Figure 1. A typical Gran's plot for the analysis of fluoride

In order to investigate the accuracy and precision of the method the samples were spiked with standard solutions and then analyzed by the described method. The accuracy expressed as percent recovery was obtained by comparing the results between the fluoride found and the fluoride added. Table 1 presents the results of the accuracy and precision experiment (n = 6).

Table 1. The accuracy and precision of the method

Tea	Fluoride added (mg/l)	Added fluoride found (mg/l)	Recovery (%)	Precision (RSD)
Hochland Mischung	0.5	0.4923	98.46	0.52
Hochland Mischung	2.0	1.9820	99.10	0.11
China Chun Mee	0.2	0.2020	101.62	3.68
Kamillentee	0.1	0.1010	101.02	2.41

The effect of brewing time on the fluoride release from three different brands of tea was also studied in this work and the results are shown in figure 2.

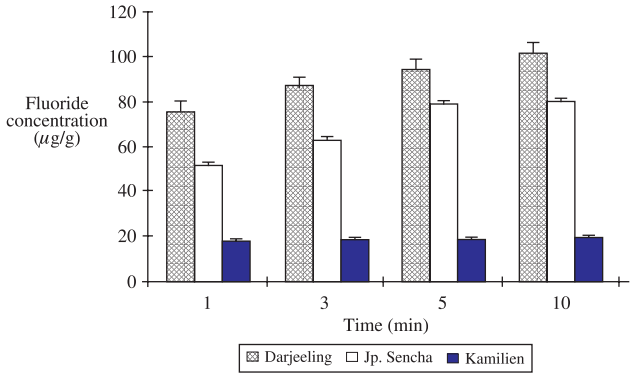

Figure 2. Effect of brewing time of the fluoride release from tea (n = 6).

The amount of fluoride in 12 brands and type of tea expressed as mg/ml and μg/g sample is summarized in table 2.

DISCUSSION

Tea is a popular beverage, which is made from the leaves of the plant species, *Camellia sinensis*. Black tea is produced from leaves that are withered, rolled, fermented and dried. When the withered leaves are steamed and rolled before the process of drying and firing to prevent the fermentation of the leaves, the product is called green tea. Recently, herbal tea is also available that is simply the combination of boiling water and dried fruit, flower or herb. The fluoride content in three different kinds of tea was presented in the present work.

The method used for the determination of fluoride was a potentiometry using an ion-selective electrode. The electrode responds selectively to fluoride ion in the presense of other ions due to the crystal of LaF_3 doped with EuF_2 as membrane of the electrode. The analysis was done automatically using a computer-controlled potentiometry for providing the Gran's plot and calculating the result, which is based on known additions of standard fluoride ion. The values of unknown fluoride concentration can be obtained by Gran's plot, in which the practical values of ion-selective electrode slopes must be known prior or by computer-controlled, especially for the multiple standard-addition. When all points of the Gran's plot lie on the straight line, the intercept on the abscissa yields the concentration of the unknown sample[16] (see figure 1).

As shown in Table 1, the accuracy of the method calculated as recovery of the standard fluoride found in three different brands of teas ranged from 98.46 to 101.62%, while the precision of the method expressed by RSD (relative standard deviation) was between 0.11 and 3.68%. The results showed that the described method demonstrated a good accuracy and precision. With increase

Table 2. Fluoride content in tea infusions, expressed in μg/l and μg/g sample

Variety of Tea	Form	Weight (g)	Fluoride Content	
			mg/l	μg/g
Black tea				
Darjeeling tea	Open	1.00	0.95 ± 0.07	93.50 ± 7.17
Ceylon Hochland	Bag	1.75	3.91 ± 0.05	223.66 ± 2.92
Assam Nr. 4 (148)	Open	1.04	1.09 ± 0.01	104.45 ± 9.50
Lipton-Best English	Bag	1.75	4.73 ± 0.15	270.23 ± 8.45
Ceylon-Indien Mischung	Bag	2.25	4.43 ± 0.12	252.97 ± 6.57
Ceylon-Assam Mischung	Bag	2.25	4.10 ± 0.17	234.37 ± 9.87
Darjeeling, Messmer	Bag	1.75	1.74 ± 0.07	99.41 ± 3.86
Green tea				
Japan Sencha	Open	1.50	0.70 ± 0.02	78.11 ± 0.77
China Chun Mee Nr. 24	Open	1.06	1.00 ± 0.07	51.40 ± 0.93
Herbal Tea				
Pfefferminze	Bag	1.75	0.27 ± 0.00	15.60 ± 0.15
Hagebuttentee	Bag	2.85	0.26 ± 0.00	9.08 ± 0.30
Kamillentee	Bag	1.50	0.27 ± 0.01	18.16 ± 0.57

in length of infusion time there was steady, slight increase in fluoride release from black and green teas. The herbal tea showed a constant fluoride release (see figure 2). The usual practice of tea preparation is to infuse tea for 5 min, which produces the best flavor with a little extraction of tannin. For this reason, the infusion time of the sample for 5 min was used in the present work. The fluoride concentration of tea infusions prepared from black teas ranged from 0.95 to 4.73 mg/l. The fluoride contents of infusion prepared from green tea and herbal tea ranged from 0.70 to 1.00 mg/l and 0.26 to 0.27 mg/l, respectively. The fluoride contents expressed as μg/g of sample were found to be (93.50–270.23) μg/g for black tea, (51.40–78.11) μg/g for green tea and (9.08–18.16) ìg/g for herbal teas. The values resulted from the present study are comparable to those from previous reports.[7,8,13] Results obtained in this study showed that depending on the brand, the kind of tea, the fluoride content in tea infusions may vary over severalfold. The variation may be due to the difference of the sample sources such as leaf age, maturity and genetics of the plant, type of soil etc. The low fluoride content of herbal tea is a reflection of its ingredient not from regular tea but from herb, flowers or fruit. It seems that most plants are found to be poor sources of fluoride, even when grown in fluoride-rich soils. Tea plants, which grow in relatively acidic soils, are exceptional for taking up fluoride. From the present study it is evident that black and green tea infusions are rich in fluoride. The mean fluoride content from black tea is higher than of green tea.

Drinking water appears to be the main dietary source of fluoride. The optimal of fluoride for prevention of dental caries in fluoridated drinking water has been suggested as 1.0 ppm. In extremely hot climate, recommended fluoride concentration is as high as 0.8 ppm whilst in cold climate, a concentration of 1.2 ppm is recommended.[19,20] On other hand, mottled teeth have occurred where the water supply is above 1.0 ppm[21] or above 1.5 ppm.[19] This is possibly caused by an inadequate diet or by the inclusion in the diet of some contributor of fluoride, such as fish or tea. A survey performed in India[22] reported that mildest grade of mottling in 10% of the population was reported due to the constant use of fluoridated water to the extent of 1 ppm. As the fluoride level increases, the effects get worse. The mildest grade of mottling in 100% of the population was observed when the fluoride concentration reached 6 ppm. The optimum fluoride intake in human is 0.07mg/kg body weight/day.[19,23]

Based on the fluoride content, the amount of fluoride in black tea and green tea may significantly contribute to the daily fluoride intake. If it is considered that a black tea bag containing 1.75–2.25 g of finely powdered black tea is brewed with 250 ml of boiled Milli-Q water for 5 minutes, the fluoride content varies from 0.4 mg to 1.18 mg. This may be no great harm for human adults when just one or two tea bags are consumed per day. Some concern has been raised over excessive intakes of fluoride from black tea infusion that contains high fluoride content such as Lipton-Best English. The ingestion of some black teas that were found to have high fluoride content by children at the age of risk to dental fluorosis should be avoided. For example, the tolerable upper limits (UL) for children aged one to three years is 1.3 mg fluoride. In this case, two cups of a black tea consumed just one a day may provide over the upper limit of the ranges of estimated believed to be associated with risk of enamel fluorosis. In addition, the level of fluoride may be much higher, when other fluoride-containing food, beverage or toothpastes are consumed. Concerning with any possible health hazards especially for children while still growing, the fluoride content in black tea products should be informed by tea manufacturing companies on their labels of the products.

It was concluded that fluoride contents of tea infusions after 5 min ranged from 0.95 to 4.73 mg/l for black teas, from 0.70 to 1.00 mg/l for green teas, and from 0.26 to 0.27 mg/l for herbal teas. Black teas and green teas examined in this study may be important contributors to the total daily fluoride intake. However, the ingestion of some black teas that were found to have high fluoride content by children at the age of risk to dental fluorosis should be avoided.

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