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Levels of Fluoride in the Ethiopian and Imported Black Tea (*Camellia sinensis*) Infusions Prepared in Tap and Fluoride-Rich Natural Waters

Abstract: In this study, the possible effect of original fluoride (F) concentration in six natural waters on the fluoride release from nine brands of tea leaves (whose origin was from Ethiopia, Sri Lanka, and China) during the infusion has been investigated. The capacity of tea leaves (commercially available tea) to absorb F from high-fluoride ground waters which were collected from the Ethiopia Rift Valley has also been investigated in this study. Infused F content of black (either powder or bag form) and green (bag form) teas and water from different sources were assayed. The fluoride content of waters used in this study ranged from 0.254 to 30.2 mg/L, while the fluoride in tea infusion of nine brands of tea ranged from 0.51 to 20.57 mg/L. The F content in moderately F-rich water was reduced from 12.5 to < 5 mg/L in tea infusion while F content in highly F-rich water was reduced from 30.2 to < 10 mg/L in tea infusion of most of the tea brands even with 3 min infusion time. Thus one can reduce the daily intake of F level by drinking tea rather than F-rich water itself.

Keywords: *Camellia sinensis*, Ethiopian tea, imported tea, fluoride, fluoride-rich natural water, tea infusion, brewing time

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1 Introduction

Tea is one of the most widely consumed beverages second to water [1–4]. Tea (*Camellia sinensis*) is a perennial plant capable of taking up fluoride (F) from soil and accumulating it in leaves [1, 2, 5, 6]. A substantial amount of F is released during tea infusion and nearly all (about 95%) of the released F is available to consumers [7–10]. Thus the tea leaves contain nearly 98% of total F content of the whole plant.

Many researchers have reported the concentration of F in tea infusion in different brands of tea of different origins [1, 6, 11]. The F concentration of several brands of tea powders and tea bags available in Ethiopian supermarkets and their tea infusions in tap water was studied by Zerabruk et al. [12].

Fluoride is found naturally in low concentration in drinking water and foods. In Ethiopia the concentration of F in surface water is generally below 1.5 mg/L in highland areas except in some locations, whereas in lowlands mainly in the Rift Valley Regions, the F content of drinking water from ground water is as high as 33 mg/L [13]. Tekle-Haimanot et al. [13] reported that water as epidemiologically the most important source of F (75–90% of daily intake) in most areas of the Rift Valley of Ethiopia.

Even though the F concentration of different types of tea, their infusion in deionized water, and the F absorbing capacity from F added deionized water (20 mg/L) have been studied [14], the significance of the present study was: to determine the daily intake of F from tea infusion prepared by naturally F-rich water which was sampled from Ethiopian Rift Valley, since naturally F-rich water has a high concentration of different metal ions which may increase or decrease the F absorbing capacity of tea leaves from naturally F-rich water by forming F metal complex; so, to determine the amount of F absorbing capacity of tea leaves from naturally F-rich water; and to determine amount of F increase or decrease in daily intake from consumption of tea prepared by naturally F-rich water.

2 Materials and methods

2.1 Sampling area description

Naturally F-rich water samples were collected from five different places in the Ethiopian Rift Valley namely Koka,

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Table 1 List of sampled teas and their details

No.	Tea brand name	Specific form	Amount per box	Country of origin	Number of box taken
Green tea bags					
1	Ethiopian green tea	Powder	25 bags	Ethiopia	3
2	Hyson green tea	Powder	25 bags	Sri Lanka	3
3	Quality green tea	Powder	25 bags	Sri Lanka	3
Black tea powder					
4	Abyssinia	Powder	80 g	Ethiopia	3
5	Black lion	Powder	80 g	Ethiopia	3
6	Lipton	Powder	200 g	Sri Lanka	3
Black tea bags					
7	Addis	Powder	25 bags	Ethiopia	3
8	Mervin strawberry	Powder	25 bags	Sri Lanka	3
9	Lipton flavored	Powder	25 bags	China	3

Wonji Shoa, Alemtena, Abomsa, and Meki in 10-L polyethylene containers. They are located in East Shoa Zone of Oromia Regional State. These sampling areas have water with F content ranged from 1.5 to 33 mg/L [13] which were used to prepare infusion of tea with low, medium, and high F content water that have been used to determine the F binding capacity of tea leaves with different levels of naturally F-rich water. The people who live in this area are affected by dental and skeletal fluorosis due to high F intake mainly from water. Furthermore, tap water having F concentration of 0.254 mg/L was collected from the Science Faculty of Addis Ababa University, Addis Ababa.

2.2 Sample collection description

Naturally F-rich water samples were collected from ground water sources used for all domestic purposes. From each site, about 4 L of water was collected, mixed, and formed one bulk sample in a pre-cleaned, dried 5-L plastic polyethylene container (zerican) rinsed with the sample water and was brought to the laboratory for immediate measurement of fluoride concentration without any pre-treatment.

A total of nine different brands of tea which were either imported or produced in Ethiopia were collected from different supermarkets in Addis Ababa. From nine brands, three were green tea bags, three were black tea powders, and the other three were black tea bags. For each of the nine brands, three packets of the same brand were randomly collected, mixed well, and put in plastic bags. A total of 27 samples were collected. List of sampled tea brands and their details are given in Table 1.

2.3 Preparation of tea infusion samples

Though there is no universal agreement on how to prepare tea beverage, the common practice in Ethiopia is adding the required amount of tea powder in boiling water and continue boiling usually for 3–5 min. In this study, tea infusion was prepared simulating home tea preparation as follows: 100 mL of water (deionized water, tap water having F concentration of 0.254 mg/L or naturally fluoride-rich water) was added to three different beakers and was brought to boil at constant temperature using hot plate, and separate 1 g tea bag was added to each of the three beakers. After 3, 5, and 10 min of infusion, it was filtered to remove the tea bag using plastic mesh and cooled to room temperature. Finally, the infusion was filtered using Whatman No. 70 filter paper, deionized water was added in a volumetric flask to make up 100 mL volume, and finally the liquor was stored in plastic beaker and its F concentration was determined. Whereas for powder form 1 g of tea was added to the boiling water at constant temperature (92°C) in three separate beakers, and boiling was continued for different infusion time of 3, 5, and 10 min, after their infusion time it was filtered using plastic mesh. The infusion was cooled to room temperature (22 $\pm$ 2°C) and filtered using Whatman No. 70 filter paper, deionized water was added in a volumetric flask to make up 100 mL volume, and finally the liquor was stored in a plastic beaker. Ready-to-drink teas were analyzed by taking samples directly from plastic beaker.

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2.4 Determination of F in tea infusion, tap water, and F-rich water

F determination in tea infusion, tap water, and F-rich water were made using a pH/ISE meter (Orion model, EA 940 Expandable Ion Analyzer, USA) equipped with combination F selective electrode (Orion Model 96-09, USA). Plastic beakers were used to store tea infusions and sampling water.

F stock solution (1,000 mg/L) was prepared from sodium fluoride (99.0% NaF, BDH Chemicals, England) and stored in 1-L volumetric flask. Total ionic strength adjustment buffer (TISAB) solution containing 58 g of sodium chloride, 57 mL of glacial acetic acid, 2 g of ethylene diamine tetra-acetate, 7 g of sodium citrate, and ~500 mL of 6 M NaOH in a volume of 1,000 mL was prepared (to adjust pH 5.0–5.5).

2.5 Method validation for F analysis

Calibration of the fluoride ion selective electrode for fluoride measurement of tea infusion and naturally fluoride-rich water was done by preparing a series of 1, 5, 10, 20, and 30 mg/L F standard solutions from stock solution of 1,000 mg/L F, whereas the calibration of the electrode for measurement of fluoride concentration of tap water was done by preparing a series of 0.1, 0.5, 1, 5, and 10 mg/L F standard solutions. An aliquot of 10 mL of F standard solution from each series was taken and mixed to 10 mL of TISAB in a 50-mL plastic beaker, the mixture were stirred (uniformly) thoroughly using a magnetic stirrer, the fluoride selective electrode was immersed in each beaker, and the calibration was done. The slope and correlation coefficient (r) of the calibration curve were found to be −58.6 mV/dec and −0.9999 for tap water, −58 mV/dec and −0.9997 for fluoride determination in naturally fluoride-rich water and tea infusion, respectively. The detection limit of the method was found to be 0.01 mg/L F.

Moreover, the precision of the results was evaluated by the standard deviation of the results of three readings of samples. To check method validity, the samples were spiked with known amount of F, and percent recovery was obtained. Table 2 shows the results of percent recovery which was ranged from 93 to 105%.

2.6 Statistical analysis

To test for significant difference between groups, one-way analysis of variance was used. The data are expressed as the mean ± standard deviation and a probability of less than 0.05 ($p < 0.05$) for the determination of significant differences between means.

3 Results and discussion

3.1 Levels of F in water and in tea infusions

The F concentrations of the waters used for preparation of tea infusions are given in Table 3. The F concentrations

Table 2 The recovery of the spiked standard and its precision for the method using tap water for tea preparation

Types of tea	F added[a] (mg/L)	F in un-spiked tea[b] (mg/L)	F in spiked tea[c] (mg/L)	% Recovery
ABTP	0.40	6.85 ± 0.16	7.24 ± 0.12	97 ± 3
BBTP	0.50	4.79 ± 0.29	5.32 ± 0.16	105 ± 3
LBTP	0.60	1.23 ± 0.10	1.78 ± 0.14	93 ± 2
HGTB	0.35	1.02 ± 0.08	1.38 ± 0.12	103 ± 2
EGTB	0.40	0.85 ± 0.04	1.25 ± 0.02	101 ± 8
QGTB	0.45	0.66 ± 0.07	1.10 ± 0.02	99 ± 2
ABTB	0.25	2.90 ± 0.13	3.14 ± 0.02	96 ± 2
FBTB	0.35	0.70 ± 0.08	1.05 ± 0.01	101 ± 5
SBTB	0.30	0.52 ± 0.08	0.82 ± 0.01	100 ± 3

Notes: [a]F (mg/L) standard added to the tea infusion. [b]F (mg/L) mean ± SD ($n = 3$) in tea infusion before standard addition. [c]F (mg/L) mean ± SD ($n = 3$) in tea infusion after standard addition. Tea infusion was prepared by boiling 1 g of tea (for tea powder) and 1 bag (for tea bag) in 100 mL of water. ABTP = Abyssinia black tea powder, BBTP = Black lion black tea powder, LBTP = Lipton black tea powder, EGTB = Ethiopian green tea bag, QGTB = Quality green tea bag, HGTB = Hyson green tea bag, FBTB = Lipton flavored black tea bag, SBTB = Marvin strawberry black tea bag, ABTB = Addis black tea bag.

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Table 3 F level (mg/L mean ± SD) of tea infusion prepared in deionized water, tap water, and different naturally F-rich water for 3, 5, and 10 min infusion

Type of tea	Infusion time (min)	DIW (F = not measurable)	TAW (F = 0.254 mg/L)	MEW (F = 2.46 mg/L)	ABW (F = 6.00 mg/L)	ALW (F = 10.2 mg/L)	WOW (F = 12.5 mg/L)	KOW (F = 30.2 mg/L)
ABTP	3	6.29 ± 0.12	6.83 ± 0.15	6.03 ± 0.04	6.92 ± 0.10	8.40 ± 0.09	8.81 ± 0.09	20.4 ± 0.2
	5	6.32 ± 0.08	7.45 ± 0.27	5.83 ± 0.12	6.63 ± 0.13	8.12 ± 0.24	8.75 ± 0.13	20.2 ± 0.2
	10	6.81 ± 0.22	7.92 ± 0.60	5.59 ± 0.08	6.62 ± 0.05	7.20 ± 0.14	7.82 ± 0.11	18.1 ± 0.1
BBTP	3	4.69 ± 0.09	4.76 ± 0.11	4.81 ± 0.05	6.79 ± 0.10	7.99 ± 0.04	8.57 ± 0.32	20.6 ± 0.1
	5	5.17 ± 0.22	5.68 ± 0.19	4.79 ± 0.05	5.72 ± 0.10	7.83 ± 0.11	8.40 ± 0.10	20.2 ± 0.3
	10	5.22 ± 10.4	5.77 ± 0.10	4.59 ± 0.04	5.68 ± 0.08	7.21 ± 0.11	7.70 ± 0.20	17.7 ± 0.1
LBTP	3	1.08 ± 0.06	1.22 ± 0.01	1.70 ± 0.02	3.33 ± 0.08	5.31 ± 0.09	6.17 ± 0.10	16.9 ± 0.2
	5	1.17 ± 0.01	1.26 ± 0.04	1.67 ± 0.03	3.27 ± 0.06	5.22 ± 0.16	6.16 ± 0.06	16.7 ± 0.2
	10	1.24 ± 0.01	1.46 ± 0.01	1.46 ± 0.02	3.08 ± 0.05	4.91 ± 0.10	5.64 ± 0.08	15.7 ± 0.2
HGTB	3	0.95 ± 0.03	1.01 ± 0.03	1.00 ± 0.01	2.06 ± 0.03	2.60 ± 0.04	3.88 ± 0.05	8.55 ± 0.10
	5	0.98 ± 0.03	1.05 ± 0.06	1.19 ± 0.01	2.07 ± 0.02	2.66 ± 0.04	3.81± 0.03	8.22 ± 0.10
	10	0.99 ± 0.01	1.05 ± 0.04	1.26 ± 0.01	2.15 ± 0.03	2.78 ± 0.02	3.59 ± 0.03	7.78 ± 0.6
QGTB	3	0.62 ± 0.02	0.65 ± 0.01	1.38 ± 0.02	2.48 ± 0.03	2.97 ± 0.02	3.93 ± 0.02	8.87± 0.16
	5	0.64 ± 0.01	0.75 ± 0.02	1.44 ± 0.02	2.62 ± 0.04	3.31 ± 0.03	3.59 ± 0.03	8.57 ± 0.04
	10	0.69 ± 0.01	0.76 ± 0.01	1.57 ± 0.03	2.71 ± 0.03	3.36 ± 0.03	3.40 ± 0.05	8.27 ± 0.15
EGTB	3	0.78 ± 0.01	0.84 ± 0.02	1.44 ± 0.02	2.22 ± 0.02	3.19 ± 0.04	3.85 ± 0.05	9.05 ± 0.05
	5	0.83 ± 0.09	0.88 ± 0.02	1.52 ± 0.02	2.24 ± 0.02	3.20 ± 0.05	3.76 ± 0.02	8.90 ± 0.11
	10	0.87 ± 0.09	0.92 ± 0.01	1.68 ± 0.02	2.31 ± 0.07	3.25 ± 0.03	3.72 ± 0.06	8.87 ± 0.10
ABTB	3	2.74 ± 0.01	2.88 ± 0.06	2.77 ± 0.04	3.42 ± 0.04	4.24 ± 0.04	4.34 ± 0.07	10.7 ± 0.06
	5	2.83 ± 0.08	2.92 ± 0.08	2.64 ± 0.05	3.38 ± 0.03	4.71 ± 0.05	4.73 ± 0.09	10.2 ± 0.3
	10	2.84 ± 0.06	2.94 ± 0.07	2.64 ± 0.04	3.39 ± 0.03	4.86 ± 0.05	4.89 ± 0.06	10.0 ± 0.2
FBTB	3	0.61 ± 0.02	0.69 ± 0.02	0.93 ± 0.02	1.75 ± 0.03	2.47 ± 0.03	3.20 ± 0.04	7.83 ± 0.08
	5	0.62 ± 0.01	0.70 ± 0.02	0.95 ± 0.02	1.76 ± 0.02	2.48 ± 0.02	3.30 ± 0.04	7.75 ± 0.10
	10	0.61 ± 0.01	0.73 ± 0.01	0.97 ± 0.02	1.81 ± 0.04	2.54 ± 0.03	3.31 ± 0.04	7.57 ± 0.10
SBTB	3	0.44 ± 0.04	0.51 ± 0.02	1.20 ± 0.04	2.20 ± 0.03	3.04 ± 0.04	3.61 ± 0.03	8.75 ± 0.20
	5	0.49 ± 0.06	0.52 ± 0.02	1.28 ± 0.02	2.31 ± 0.02	3.14 ± 0.03	3.78 ± 0.03	8.72 ± 0.10
	10	0.51 ± 0.03	0.59 ± 0.02	1.30 ± 0.03	2.38 ± 0.05	3.27 ± 0.03	3.80 ± 0.02	8.72 ± 0.12

Notes: DIW = deionized water, TAW = tap water, ABW = Abonsa water, MEW = Mekie water, ALW = Alemtena water, WOW = Wonji Shoa water, KOW = Koka water.

of tea prepared by infusing for 3, 5, and 10 min using different water sources were in the range of 0.44–20.6, 0.49–20.2, and 0.51–18.1 mg/L, respectively (Table 3).

Table 3 shows that F concentrations of nine different brands of tea varied one brand to the other; this might be due to type of tea plant, fertilizer, soil type, age of leaves, rain fall and quality of water for irrigation, and harvesting time. But also it may be due to types of water used for the preparation of tea infusion.

The highest level of F was found in Black Lion tea powder with mean F concentrations of 20.6 and 20.2 mg/L for 3 and 5 min infusion, respectively, while for 10 min infusion Abyssinia black tea powder was the highest with mean F concentration of 18.1 mg/L in which Koka water was used for tea preparation in both tea brands. Whereas the lowest concentration of F was found in strawberry black tea bag with mean F of 0.44, 0.49, and 0.51 mg/mL for 3, 5, and 10 min infusion in which deionized water was used for tea liquor preparation.

3.2 Amount of F absorbed by tea leaves

In low F water, F is easily released from the tea leaves to the infusion. Depending upon the F content of the water, dried tea leaves are also reported to absorb F from F-rich water [14]. The F absorbing capacity of the tea infusion seems to be independent of the F concentration of the tea leaves. Thus, both the high F tea leaves and low F tea leaves reduced the F concentration of the infusion below the initial water F concentration. This may indicate that factors other than the F content of the tea leaves are of importance for F release and absorption [14].

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Table 4 Percentage (% mean ± SD) of F absorbed from different F-rich water by different brands of tea leaves in which tea liquor is prepared in deionized water as a reference

Type of tea	Infusion time (min)	% of F absorbed from MEW by tea leaves	% of F absorbed from ABW by tea leaves	% of F absorbed from ALW by tea leaves	% of F absorbed from WOW by tea leaves	% of F absorbed from KOW by tea leaves
ABTP	3	31.6 ± 1.3	43.7 ± 0.8	48.7 ± 0.6	53.1 ± 0.7	44.1 ± 0.5
	5	33.8 ± 1.7	46.2 ± 0.6	50.5 ± 2.7	53.5 ± 1.1	44.8 ± 0.4
	10	39.3 ± 1.0	48.3 ± 0.3	57.4 ± 0.6	59.5 ± 0.8	51.2 ± 0.3
BBTP	3	32.6 ± 1.0	35.3 ± 1.5	49.4 ± 0.6	50.1 ± 1.5	41.0 ± 0.4
	5	37.3 ± 1.3	48.8 ± 1.1	48.7 ± 0.3	52.5 ± 0.5	42.8 ± 0.1
	10	50.2 ± 0.1	49.4 ± 0.5	52.9 ± 1.0	56.5 ± 0.9	52.1 ± 0.3
LBTP	3	51.9 ± 2.9	53.0 ± 0.7	52.5 ± 0.6	54.6 ± 0.6	46.0 ± 0.3
	5	56.0 ± 2.9	54.4 ± 2.2	53.7 ± 1.5	54.9 ± 1.2	46.9 ± 0.8
	10	84.4 ± 0.3	57.5 ± 0.3	56.7 ± 0.5	59.0 ± 0.3	50.1 ± 0.5
HGTB	3	70.7 ± 0.8	70.4 ± 0.2	76.5 ± 1.5	71.2 ± 0.3	72.6 ± 0.3
	5	65.4 ± 0.9	70.3 ± 0.2	76.0 ± 0.2	71.7 ± 0.2	73.6 ± 0.3
	10	63.3 ± 1.2	69.2 ± 0.5	74.9 ± 0.1	73.4 ± 0.2	75.0 ± 0.1
QGTB	3	55.2 ± 1.1	62.5 ± 0.1	72.3 ± 0.1	70.0 ± 1.0	71.2 ± 0.2
	5	53.5 ± 1.2	60.5 ± 0.4	69.2 ± 0.2	72.7 ± 0.1	72.2 ± 0.7
	10	50.2 ± 1.0	59.5 ± 0.3	68.9 ± 0.2	74.2 ± 0.1	73.2 ± 0.4
EGTB	3	55.5 ± 1.1	67.3 ± 0.20	70.7 ± 0.5	71.0 ± 0.6	70.8 ± 0.2
	5	53.7 ± 0.9	67.0 ± 0.2	70.7 ± 0.2	71.8 ± 0.2	71.3 ± 0.3
	10	49.3 ± 1.2	66.4 ± 0.3	70.4 ± 0.2	72.2 ± 0.3	71.5 ± 0.2
ABTB	3	46.6 ± 0.8	60.4 ± 0.6	67.0 ± 0.1	71.5 ± 0.7	67.6 ± 0.4
	5	50.1 ± 0.9	61.7 ± 0.2	63.6 ± 0.1	69.1 ± 0.2	69.2 ± 0.3
	10	50.3 ± 0.5	61.6 ± 0.3	62.1 ± 0.3	68.1 ± 0.2	69.7 ± 0.3
FBTB	3	69.7 ± 0.4	73.5 ± 0.3	76.9 ± 0.2	75.6 ± 0.7	74.6 ± 1.8
	5	69.2 ± 0.7	73.4 ± 0.3	76.9 ± 0.4	74.8 ± 0.3	74.9 ± 0.2
	10	68.7 ± 0.6	72.3 ± 0.1	76.3 ± 0.2	74.8 ±0.3	75.4 ± 0.3
SBTB	3	58.7 ± 0.4	65.8 ± 0.2	71.4 ± 0.8	72.1 ± 0.2	71.6 ± 0.4
	5	57.2 ± 1.0	64.5 ± 0.2	70.3 ± 0.2	70.9 ± 0.5	71.6 ± 0.2
	10	56.9 ± 0.7	63.5 ± 0.1	69.2 ± 0.2	70.8 ± 0.3	71.5 ± 1.8

Tables 4 and 5 show the percentage of F absorbed from different naturally F-rich water by different brands of tea in which their F concentration varies. The percentages of F absorbed were calculated by considering deionized water and tap water as references.

3.2.1 When deionized water was used as reference

$$\%\ \text{Fluoride absorbed from water by tea} = \frac{\text{F total} - \text{Ft}}{\text{F total}} \times 100$$

where F total = F in tea liquor prepared in deionized water + F in water used for tea preparation and Ft = F in tea liquor prepared in F-rich water.

The percentage of F absorbed from naturally F-rich water from Meki (2.46 mg/L), Abomsa (6.0 mg/L), Alemtena (10.1 mg/L), Wonji Shoa (12.5 mg/L), and Koka (30.2 mg/L) by ABTP, BBTP, and LBTP was increased with increasing brewing time to 3, 5, and 10 min. But also from Koka water (30.2 mg/L) by HGTB, EGTB, QGTB, ABTB, SBTB, and FBTB teas and from Wonji Shoa (12.5 mg/L) by HGTB, EGTB, and QGTB were shown similar increment. Whereas the percentage of fluoride absorbed from naturally fluoride-rich water (Mekie, Abonsa, and Alemtena) by HGTB. EGTB, QGTB ABTB, SBTB, and FBB decreased when brewing time was increased to 3, 5, and 10 min. This may be due to the presence of higher concentrations of F complexing metals (Fe, Al, Ca, etc.) in some brands of tea leaves and infused to the tea liquor in high amount which resulted the percentage of F absorbed from F-rich water by tea leaves decreased as brewing time increased to 3, 5, and 10 min by forming complex to free F in naturally F-rich water. But when the F concentration is so high as in Koka water F absorbed by tea leaves increased as brewing time increased to 3, 5, and 10 min, even though F complexing metals infused from tea leaves to water reduce free F in Koka water there is

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Table 5 Percentage (% mean ± SD) of F absorbed from different naturally F-rich water by different brands of tea leaves in which tea liquor is prepared in tap water as a reference

Type of tea	Infusion time (min)	% of F absorbed from MEW by tea leaves	% of F absorbed from ABW by tea leaves	% of F absorbed ALW by tea leaves	% of F absorbed WOW by tea leaves	% of F absorbed KOW by tea leaves
ABTP	3	35.1 ± 0.8	46.1 ± 0.1	50.4 ± 0.3	54.4 ± 0.4	44.9 ± 0.3
	5	41.2 ± 1.2	50.5 ± 1.6	53.7 ± 2.5	56.1 ± 0.7	46.2 ± 0.3
	10	46.1 ± 3.4	52.4 ± 1.8	59.8 ± 0.8	61.7 ± 1.2	52.5 ± 0.5
BBTP	3	33.4 ± 1.2	36.9 ± 0.7	46.6 ± 1.0	50.3 ± 1.6	42.2 ± 1.6
	5	41.2 ± 2.2	51.0 ± 1.3	50.4 ± 0.8	53.8 ± 0.9	42.7 ± 1.6
	10	44.2 ± 1.0	51.7 ± 0.1	54.6 ± 0.3	57.9 ± 0.7	50.8 ± 0.5
LBTP	3	53.8 ± 08	53.9 ± 0.7	53.1 ± 0.8	55.0 ± 0.4	46.2 ± 0.4
	5	55.1 ± 0.9	55.0 ± 0.3	54.0 ± 0.5	55.2 ± 0.3	47.0 ± 0.4
	10	62.8 ± 0.9	58.7 ± 0.3	57.5 ± 0.6	59.6 ± 0.4	50.7 ± 0.4
HGTB	3	71.2 ± 0.8	70.6 ± 0.2	76.6 ± 0.2	71.3 ± 0.3	71.7 ± 0.2
	5	66.1 ± 0.2	70.6 ± 0.1	76.1 ± 0.1	71.9 ± 0.2	72.8 ± 0.3
	10	64.1 ± 1.4	69.5 ± 0.6	75.1 ± 0.1	73.5 ± 0.2	74.3 ± 0.1
QGTB	3	55.6 ± 0.8	62.7 ± 0.1	72.4 ± 0.2	70.1 ± 1.1	71.2 ± 0.2
	5	55.1 ± 0.4	61.2 ± 0.3	69.5 ± 0.2	72.9 ± 0.1	72.3 ± 0.7
	10	51.2 ± 0.9	59.9 ± 0.2	69.1 ± 0.2	75.5 ± 0.1	73.3 ± 0.4
EGTB	3	56.4 ± 0.9	67.5 ± 0.2	70.8 ± 0.2	71.1 ± 0.6	70.8 ± 0.2
	5	54.5 ± 0.2	67.4 ± 0.4	64.5 ± 0.2	71.9 ± 0.1	71.4 ± 0.3
	10	50.3 ± 1.3	66.6 ± 0.4	70.5 ± 0.1	75.6 ± 0.3	71.5 ± 0.2
ABTB	3	48.1 ± 0.9	61.4 ± 0.1	67.3 ± 0.6	71.8 ± 0.7	67.7 ± 0.4
	5	50.9 ± 0.2	62.1 ± 0.2	63.9 ± 0.2	69.3 ± 0.2	69.3 ± 0.3
	10	51.1 ± 0.7	62.1 ± 0.1	62.7 ± 0.2	68.3 ± 0.2	69.8 ± 0.1
FBTB	3	70.5 ± 0.5	73.8 ± 0.2	77.1 ± 0.2	75.5 ± 0.8	74.7 ± 0.2
	5	69.9 ± 0.1	73.7 ± 0.3	77.0 ± 0.2	75.0 ± 0.3	74.9 ± 0.2
	10	69.6 ± 0.5	73.1 ± 0.1	76.5 ± 0.2	75.0 ± 0.3	75.5 ± 0.3
SBTB	3	59.6 ± 0.6	62.2 ± 0.4	71.3 ± 0.1	72.3 ± 0.2	71.5 ± 0.4
	5	57.0 ± 0.3	64.6 ± 0.3	70.4 ± 0.2	71.0 ± 0.5	71.6 ± 0.3
	10	57.4 ± 0.9	63.9 ± 0.2	69.4 ± 0.1	71.0 ± 0.3	71.7 ± 0.3

enough free F absorbed by tea leaves during each brewing time. From Tables 4 and 5 the percentages of F absorbed from naturally F-rich water by different brand tea leaves were different; this may be due to the fact that each brand of tea has its own maximum F binding capacity depending upon the presence of F complexing metal in tea or in naturally F-rich water.

3.2.2 When tap water is used as reference

$$\%\ \text{Fluoride absorbed from water by tea} = \frac{\text{F total} - \text{Ft}}{\text{F total}} \times 100$$

where F total = F in tea liquor prepared in tap water + F in water used for tea preparation and Ft = F in tea liquor prepared in F-rich water.

Generally, the amount of F absorbed from naturally F-rich water by different brands of tea leaves follow similar trend in the above case in which tap water was used as reference.

3.3 F intake by human and safety evaluation of the teas

F is used worldwide to control dental caries. However, ingestion of high levels of F during tooth formation and mineralization is responsible for dental fluorosis. Prolonged ingestion of high levels of F leads to corroded, pitted rusty brown teeth [15].

Surplus of F in organism can cause teeth and skeleton fluorosis. F inhibits many enzymes. Affected enzyme contains metal ion which unites with F and creates metal–F complex. F in organism has its optimal, security-tolerant and toxic dose, which depends on person's age, weight, and health. In the first year of life,

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the optimal content of F is 0.045 mg/kg of body mass, tolerant 0.073 mg/kg of body mass, chronically toxic 0.150 mg/kg of body mass. Optimal dosage of F for adults is 0.020–0.025 mg/kg of body mass [15]. As intake of F, according to World Health Organization (WHO) recommendation is limited in the range from 2 to 4 mg/day, it is necessary to give the content of F in all products that are used in human consumption [15].

The Food and Nutrition Board of the Institute of Medicine (FNBIM, USA) has developed adequate intakes (AIs) for F. The AI is the "estimated F intake that has been shown to reduce the occurrence of dental caries maximally in a population without causing unwanted side effects including moderate dental fluorosis." The AIs for each age group are presented in Table 6 (Relevance to Public Health [16], http://www.atsdr.cdc.gov/ToxProfiles/tp11-c2.pdf).

In this study, the F level of different brands of tea in different F-rich water ranges from 0.51 to 20.57 mg/mL in 3 min brewing time which is mostly practiced in Ethiopia. Considering that on the average a person consumes two cups (200 mL) of tea each day, the daily F intake from tea alone ranges from 0.10 to 4.11 mg. Thus according to the FNBIM (USA) recommended daily intake of F, that is, consumption of two cups of tea from all brands (except ABTP, LBTP, ABTB, and BBTP which are prepared in KOW) is safe for all population (>9 years old) in which the tea is prepared in all different naturally F-rich water. But all brands of tea prepared using all types of water are not safe for infants (0–6 months old). In this study, it is also observed that consuming two cups of tea of ABTP (in all types of water), all brands of tea in KOW, BBTP (in ABW, ALW, and WOW), and LBTP (in ALW and WOW) will exceed the recommended daily intake 1 mg by FNBIM for all population below 8 years. Except FBTB in ALW, BBTP and ABTB (in all types of water), all brands of tea (in WOW), and EGTB and ABTB (in ALW) are also exceeding the recommended daily intake 0.7 mg by FNBIM for all population below 3 years. The data are given in Table 7.

Table 6 AI levels for F (Relevance to Public Health [16], http://www.atsdr.cdc.gov/ToxProfiles/tp11-c2.pdf)

Age range	AI level (mg/day)	AI level (mg/kg/day)
0–6 month	0.01	0.0014
7–12 month	0.5	0.056
1–3 years	0.7	0.054
4–8 years	1	0.045
9–13 years (females or males)	2	0.05
14–18 years (males)	3	0.046
14–18 years (females)	3	0.053
>18 years (males)	4	0.052
>18 years (females)	3	0.049

The data in Table 8 showed that F ingestion in consuming two cups of tea ranges from 4.4% (in deionized water) to 206% (in KOW) of recommended daily intake for a person with a weight of 40 kg. A person who consumes two cups (ABTP, BBTP, LBTP, and ABTB) of tea prepared using KOW was ingested even above the recommended daily intake from all sources.

$$\%\ \mathrm{RDIF} = \frac{\mathrm{DIF}}{\mathrm{RDIF}} \times 100$$

where %RDIF = percent of recommended intake of F/kg/day. DIF = daily intake of F from any source. RDIF = recommended intake of F/kg/day (0.05 mg/kg/day).

According to WHO, RDIF is 0.05 mg/kg/day [7], so for an adult with an average weight of 40 kg, the amount of recommended fluoride intake is 2 mg/day which can be expressed as 2 mg/40 kg/day.

Table 7 Expected daily F intake from different brands of teas prepared by different types of water if one consuming two cups (200 mL) of tea in mg

Type of tea	DIW	TAW	MEW	ABW	ALW	WOW	KOW
ABTP	1.26	1.37	1.21	1.38	1.68	1.76	4.08
BBTP	0.94	0.95	0.96	1.36	1.60	1.71	4.11
LBTP	0.22	0.24	0.34	0.67	1.06	1.23	3.38
HGTB	0.19	0.20	0.20	0.41	0.52	1.23	1.71
EGTB	0.16	0.17	0.29	0.44	0.64	0.77	1.81
QGTB	0.12	0.13	0.28	0.50	0.59	0.79	1.77
ABTB	0.55	0.58	0.55	0.68	0.85	0.87	2.13
FBTB	0.12	0.14	0.19	0.35	0.49	0.64	1.57
SBTB	0.088	0.10	0.24	0.44	0.61	0.72	1.75

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Table 8 Percent of recommended F intake (0.05 mg/kg/day) for different brands of tea prepared in different types of water for a person (40 kg weight) consuming two cups of tea

Type of tea	DIW	TAW	ABW	MEW	ALW	WOW	KOW
ABTP	62.9	68.3	60.3	69.2	84.0	88.1	204
BBTP	46.9	47.6	48.1	67.9	79.9	85.7	206
LBTP	10.8	12.2	17.0	33.3	53.1	61.7	169
HGTB	9.50	10.1	10.0	20.6	26.0	38.8	85.5
EGTB	7.80	8.40	14.4	22.2	31.9	38.5	90.5
QGTB	6.20	6.50	13.8	24.8	29.7	39.3	88.7
ABTB	27.4	28.8	27.7	34.2	42.4	43.4	106.7
FBTB	6.10	6.90	9.30	17.5	24.7	32.0	78.3
SBTB	4.40	5.10	12.0	22.0	30.4	36.1	87.5

4 Conclusions

The F concentration of tea infused in different water at three different brewing times and the F concentration of different water in Ethiopian Rift Valley were assessed. The main findings of this study are the following: (i) a person who consumes high F level water can reduce the daily F intake by addition of tea leaves to F-rich water, especially tea leaves with low F level have a high potential to reduce the F level of water, (ii) depending upon the brands of tea or F-rich water used for tea preparation, increasing brewing time can increase or decrease the amount of F infused from F-rich water to tea leaves, which may be due to the F binding capacity of tea, F complexing metals present in either tea or water used for tea preparation, and (iii) the Ethiopian black tea powder and black tea bag have high levels of F than imported ones. From imported ones, HGTB has high level F than EGTB which is Ethiopian green tea. This variation may be due to the high soil F level, soil pH, low soil calcium level which is used to precipitate F in the soil and roots of the plant, or old tea leaf which causes high F level.

This study was limited to the determination of F concentration in the water and tea infusions. The determination of metal ion concentration in the water and tea infusions is necessary to understand the mechanism of F absorption capacity of tea leaves during tea infusion.

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