

## FLUORIDE, MAGNESIUM, AND SODIUM IN DENTAL CHEWING STICK PLANTS USED IN BANGLADESH

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**SUMMARY:** Proton induced gamma emission (PIGE) reactions were employed for determining fluorine (F), magnesium (Mg), and sodium (Na) concentrations in samples of some pharmacologically important plants used to make dental chewing sticks in Bangladesh. The amount of F in the samples (above the method detection limit of 12 mg/kg dry sample) ranged from  $50\pm 19$  to  $222\pm 17$  mg/kg, with the highest concentration found in Neem leaves. In plant trunk samples the highest level of F ( $122\pm 14$  mg/kg) was found in the Bely-asra plant, but no F was measured in trunk samples of Nishinda, Neem, or Bohera. Likewise, no F was found in trunk samples of Joytun (the most popular chewing stick source used by devout Muslims), nor in such samples of Kaminee, Akondo, Batul, Olut-kumbal, and Sheora. The biologically important elements Mg and Na were also measured and were present in the range of  $3231\pm 81$  to  $7430\pm 299$  mg/kg and  $10\pm 2$  to  $1521\pm 8$  mg/kg, respectively. Bohera leaves contain the highest amount of Mg, and Joytun trunk samples have the highest level of Na. These findings may be important in considering the value of these chewing sticks on dental and oral health.

**Keywords:** Bangladesh; Chewing sticks; Dental caries and chewing sticks; F in chewing stick plants; Magnesium in chewing stick plants; PIGE; Proton induced gamma emission; Sodium in chewing stick plants.

### INTRODUCTION

Despite widespread use of toothbrushes and toothpastes, a natural tooth cleaning method using chewing sticks prepared from the twigs, stems, or roots of various plant species has been practiced for thousands of years in Asia, Africa, the Middle East, and the Americas.<sup>1</sup> Some clinical studies have shown that chewing sticks, when properly used, can be as effective as toothbrushes in maintaining oral hygiene due to their combined effect of mechanical cleaning and enhancement of salivation.<sup>2</sup> Apart from these mechanical effects, many of these chewing sticks have been shown to have significant antimicrobial activity against a broad spectrum of microorganisms.<sup>3,4</sup> Today, chewing sticks are still used in many developing countries because of religious and/or cultural traditions, and because of their easy availability, low cost, and simplicity.

Bangladesh has many varieties of plants that are used for therapeutic purposes against various diseases and ailments. Most of the rural and even some urban people in Bangladesh use chewing sticks made from these plants for cleaning their teeth instead of toothpaste and a toothbrush. The sticks average 20 cm in length and 1.5 cm in diameter, and the teeth of those who use them are generally strong, clean, fresh looking, and devoid of germs and caries. Decayed, missing, filled, or perforated teeth are rarely found among stick users. The leaves, stems and roots of

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the plants are also made into mixtures such as powders and pastes that are used to treat ailing teeth and as a painkiller.

For over sixty years, there has been widespread acceptance of the belief that F prevents dental caries even though the mechanism for such action appears to be quite complicated and not well understood.<sup>5</sup> F has also been introduced into modern toothpastes for cariostatic and therapeutic purposes. On the other hand, various studies have revealed that excessive F intake causes fluorosis, cancer, arthritis, and other diseases.<sup>6–9</sup> It has also been reported that F ion in excess affects intelligence, especially in children, who are most susceptible to early F toxicity.<sup>10,11</sup> Chronic F intoxication (fluorosis) and sensitivity to fluoride have been found not only in humans,<sup>6–9</sup> but also in domestic animals including cows,<sup>12</sup> horses,<sup>13,14</sup> sheep,<sup>15</sup> and goats.<sup>16</sup>

The objective of the present study was to estimate the concentrations of F, Mg, and Na in some local chewing stick plants in order to help assess their impact on human health. In this work, proton induced gamma-ray emission (PIGE) was employed for the measurement of these elements in chewing stick plant samples collected from both rural and urban areas of Bangladesh. PIGE analysis is based on the detection of prompt gamma rays emitted from excited nuclei following (p, p'γ) nuclear reactions. The energy of the gamma ray indicates the identity of the isotope, and the intensity of the emission gives a measure of the concentration of the isotope present in the sample. PIGE is a sensitive and reliable method for quantitative analysis of relatively low atomic number elements like F, sodium, and magnesium.<sup>17,18</sup> An important advantage of the PIGE is its ability to determine a number of such elements simultaneously, especially lithium, boron, fluorine, sodium, magnesium, etc. in health-related environmental samples.

## MATERIALS AND METHODS

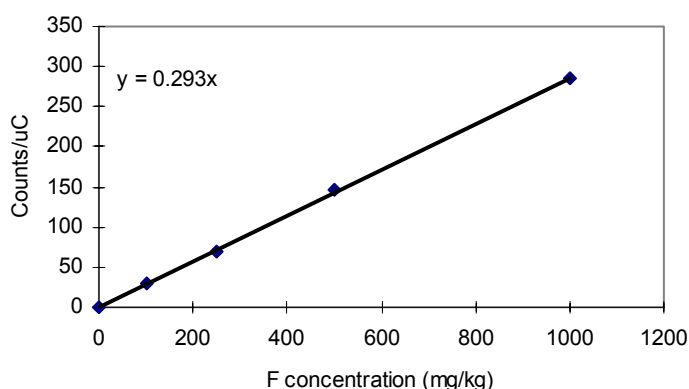
**Sample collection:** Samples from fourteen chewing stick plants were collected for analysis from different localities of the Dhaka and Shirajgong districts of Bangladesh. They are listed in Table 1 with both their local and their international scientific names.

**Table 1.** List of chewing stick plants with their local and scientific names along with the names of the parts of the plants and the places where the samples were collected

Sample No.	Local name	Scientific name	Parts of the plant collected and analyzed	Place of collection
1	Nishinda	<i>Vitex negundo</i> L.	Leaves, Trunk	Savar, Dhaka
2	Neem	<i>Atzadiracha indica</i> A. Juss	Leaves, Trunk	Savar, Dhaka
3	Bely-asra	<i>Achyranthes aspera</i>	Leaves, Trunk	Savar, Dhaka
4	Bhat	<i>Clerodendrum viscosum</i>	Leaves, Trunk	Savar, Dhaka
5	Joytun	<i>Sesbania sesban</i> (L.) Merr.	Trunk	Dhaka city
6	Kaminee	<i>Murraya paniculata</i> (L.) Jacq.	Leaves, Trunk	Savar, Dhaka
7	Akondo	<i>Calotropis procera</i>	Leaves, Trunk	Savar, Dhaka
8	Khejur	<i>Phoenix sylvestris</i>	Trunk	Savar, Dhaka
9	Bohera	<i>Terminalia belerica</i> Roxb	Leaves, Trunk	Shirajgong
10	Moth-bhringraj	<i>Wedelia chinensis</i> (osb.) Merr.	Leaves, Trunk	Shirajgonj
11	Batul	<i>Sapium indicum</i> Willd.	Trunk	Shirajgonj
12	Olut-kumbal	<i>Abroma angusta</i> L.	Root	Savar, Dhaka
13	Sheora	<i>Streblus asper</i> Lour	Trunk	Shirajgonj
14	Motkila	<i>Glycosmis arborea</i> Dc	Leaves	Savar, Dhaka

**Sample preparation:** The samples were sliced into pieces and dried in an oven at a temperature of 130°C for 10 hr followed by grinding and pelletizing for irradiation as described in detail elsewhere.<sup>17,18</sup>

**Concentration calibration:** Standard samples used for calibration were prepared in a cellulose matrix.<sup>17,18</sup> Analar grade NaF at concentrations ranging from 10 to 1000 mg/kg cellulose were homogeneously dispersed in cellulose matrices and dried under an infrared lamp followed by grinding and pressing into 7 mm diameter pellets. The number of gamma rays emitted from the nuclear reaction  $^{19}\text{F}(p, p'\gamma)^{19}\text{F}$  per unit charge was used to construct the calibration plot shown in the following Figure. This plot was used to measure the F concentrations on a dry weight basis (mg/kg) in the irradiated samples.



**Figure.** Concentration calibration plot for F on a dry weight basis.

**Method of analysis:** Concentration measurements were made with a proton beam energy of 2.5 MeV obtained from the 3 MV Van de Graaff accelerator at the Atomic Energy Centre, Dhaka (AECD) using the external beam PIGE method. The experimental set up is described elsewhere.<sup>17,18</sup> The estimated proton energy on the target after absorption at the exit window and the air between the window and the sample is 2.3 MeV. Each sample was irradiated for a preset charge of 80 C with a beam current of 20 nA. The characteristic  $\gamma$ -rays emitted following the nuclear reactions  $^{19}\text{F}(p, p'\gamma)^{19}\text{F}$ ,  $^{23}\text{Na}(p, p'\gamma)^{23}\text{Na}$ ,  $^{24}\text{Mg}(p, p'\gamma)^{24}\text{Mg}$ , and  $^{25}\text{Mg}(p, p'\gamma)^{25}\text{Mg}$  were detected with a high purity germanium (HPGe) detector having a resolution of 1.75 keV at 1332 keV  $\gamma$ -rays. Standard NIM electronics and a 4095 channel pulse height analyzer were used for data acquisition and analysis. The gamma ray spectrum was analyzed using commercially available  $\gamma$ -ray unfolding software obtained from APTEC Nuclear Inc. USA.

## RESULTS AND DISCUSSION

In this work, the minimum detection limit (MDL), which is a measure of sensitivity, is defined as the minimum amount of an element that gives a count equal to the three times the standard deviation of the background in a spectrum obtained from a target of, ideally, a pure element.<sup>19</sup>

Mathematically,

$$MDL = 3 \sqrt{\frac{N_b}{S}}$$

where,  $N_b$  = No. of counts/ $\mu$ C in the background within an energy interval of two FWHM around the gamma peak, and  $S$  = Sensitivity of the element. In this study the MDL of  $^{19}\text{F}$  was found to be 12 mg/kg and that of  $^{23}\text{Na}$  to be 1 mg/kg, whereas that of  $^{24}\text{Mg}$  was 32 mg/kg and that of  $^{25}\text{Mg}$  was 73 mg/kg.

F concentrations in eight chewing stick plant samples, determined using the calibration shown in the Figure, are presented in Table 2. In most cases, both leaves and trunks were analyzed. F was detected both in the leaves and trunk samples only of Moth-bhringraj. F was detected only in the leaves of the chewing stick plants Nishinda, Neem, and Bohera, whereas F was detected only in the trunk of Bely-asra, Bhat, and Khejur.

**Table 2.** F concentrations in eight different chewing stick plant samples

Sample No.	Local name	Scientific name	Parts of the plant collected and analyzed	Place of collection
1	Nishinda	<i>Vitex negundo</i> L.	Leaves, Trunk	147±13 BDL <sup>a</sup>
2	Neem	<i>Atzadiracha indica</i> A. Juss	Leaves, Trunk	222±17 BDL
3	Bely-asra	<i>Achyranths aspera</i>	Leaves, Trunk	BDL 122±14
4	Bhat	<i>Clerodendrum viscosum</i>	Leaves, Trunk	BDL 78±21
5	Khejur	<i>Phoenix sylvestris</i>	Trunk	57±24
6	Bohera	<i>Terminalia belerica</i> Roxb	Leaves, Trunk	109±11 BDL
7	Moth-bhringraj	<i>Wedelia chinensis</i> (osb.) Merr.	Leaves, Trunk	50±19 78±10
8	Motkila	<i>Glycosmis arborea</i> Dc	Leaves	110±23

<sup>a</sup>Below detection limit of 12 mg F/kg dry sample.

As seen in Table 2, F concentrations in chewing stick plant samples range from 50±19 to 222±17 mg/kg. Among the leaf samples, Neem leaves contain the highest amount of F, but no F was detected in the popular chewing stick made from the trunk of Neem. Among the trunk samples, the highest amount of F was observed in Bely-asra, whereas no F was found in its leaves. F was not detected in Joytun, the most popular chewing stick, especially among Muslim devotees, because its use is mentioned in the Holy Quran. No F was also detected in the trunks of the widely used chewing stick plants Nishinda, Neem, and Bohera. F was also not detected in other chewing stick plants such as Kaminee, Akondo, Batul, Olut-kumbal, and Sheora. Overall, only about 40% of the chewing stick plant samples contain F. Moreover, the F content measured in the samples is not significant compared to the amount of F introduced in most commercial toothpastes. The absence of detectable F or a low amount of F in chewing stick plants, especially in the trunks, which are used for tooth care, indicates that F may not play a significant role in the prevention of the tooth decay.

The concentrations of  $^{23}\text{Na}$ ,  $^{24}\text{Mg}$ , and  $^{25}\text{Mg}$  measured in samples from 14 different chewing stick plants are shown in Table 3. These were determined by PIGE after calibration using the certified values of these isotopes obtained from NBS SRM 1515 apple leaves, and NBS SRM 1573 tomato leaves. Concentrations of  $^{26}\text{Mg}$  isotopes were calculated using the natural abundances of magnesium isotopes.

As shown in Table 3, Mg concentrations were found to be relatively high in all the chewing stick plant samples and vary from  $3231\pm 81$  to  $7430\pm 299$  mg/kg. Bohera leaves contain the highest amount of Mg,  $7430\pm 299$  mg/kg. Concentrations of Mg in widely used chewing stick trunk samples such as Nishinda, Neem, Bely-asra, Bhat, Joytun, Kaminee, and Motkia were found to range from  $6903\pm 331$  to  $4028\pm 266$ . Mg is used in dentifrices as an abrasive to provide mechanical aid in the paste to help remove the stains from the tooth surface. Mg is also incorporated in some dentifrices to neutralize acid products on the surface of the tooth, thus inhibiting caries. The results of the present study indicate that most of the chewing sticks analyzed may be effective as an abrasive as well as an acid neutralizer.

**Table 3.** Concentrations of Na and Mg measured in chewing stick plant samples

Sample No.	Local name	Parts of the plant	Concentration of isotopes/elements (mg/kg)				
			$^{24}\text{Mg}$	$^{25}\text{Mg}$	$^{26}\text{Mg}$	Mg	Na
1	Nishinda	Leaves	$3135\pm 94$	$298\pm 95$	$328\pm 102$	$3761\pm 291$	$31\pm 3$
		Trunk	$6332\pm 189$	$272\pm 68$	$299\pm 74$	$6903\pm 331$	$208\pm 4$
2	Neem	Leaves	$3454\pm 103$	$368\pm 73$	$405\pm 77$	$4227\pm 253$	$23\pm 3$
		Trunk	$3496\pm 104$	$339\pm 77$	$373\pm 85$	$4028\pm 266$	$17\pm 2$
3	Bely-asra	Leaves	$3499\pm 70$	$483\pm 130$	$531\pm 133$	$4513\pm 333$	$23\pm 4$
		Trunk	$3834\pm 115$	$581\pm 156$	$639\pm 131$	$5054\pm 402$	$298\pm 3$
4	Bhat	Leaves	$4189\pm 83$	$667\pm 153$	$744\pm 124$	$5610\pm 360$	$31\pm 3$
		Trunk	$4593\pm 91$	$588\pm 106$	$647\pm 117$	$5828\pm 314$	$54\pm 3$
5	Joytun	Leaves	-	-	-	-	-
		Trunk	$4558\pm 91$	$81\pm 16$	$88\pm 17$	$4727\pm 124$	$1521\pm 8$
6	Kaminee	Leaves	$3314\pm 66$	$552\pm 125$	$608\pm 137$	$4474\pm 328$	$10\pm 2$
		Trunk	$5285\pm 118$	$405\pm 152$	$446\pm 165$	$6136\pm 435$	$34\pm 4$
7	Akondo	Leaves	$5755\pm 172$	$365\pm 104$	$401\pm 114$	$6521\pm 390$	$856\pm 9$
		Trunk	$4426\pm 132$	$383\pm 122$	$422\pm 134$	$5231\pm 388$	$82\pm 2$
8	Khejur	Leaves	-	-	-	-	-
		Trunk	$3145\pm 62$	$572\pm 137$	$629\pm 151$	$4346\pm 350$	$347\pm 3$
9	Bohera	Leaves	$6165\pm 123$	$602\pm 84$	$663\pm 92$	$7430\pm 299$	$33\pm 3$
		Trunk	$2909\pm 29$	$317\pm 44$	$348\pm 48$	$3574\pm 121$	$34\pm 2$
10	Moth-bhringraj	Leaves	$2638\pm 26$	$844\pm 126$	$928\pm 139$	$4410\pm 291$	$111\pm 2$
		Trunk	$2995\pm 89$	$119\pm 15$	$130\pm 16$	$3244\pm 120$	$124\pm 2$
11	Batul	Leaves	-	-	-	-	-
		Trunk	$3030\pm 60$	$56\pm 10$	$105\pm 11$	$3231\pm 81$	$11\pm 1$
12	Olut-kumbal	Root	$3500\pm 70$	$381\pm 53$	$410\pm 58$	$4301\pm 181$	$291\pm 3$
13	Sheora	Leaves	$2995\pm 59$	$118\pm 20$	$130\pm 22$	$3243\pm 101$	$46\pm 3$
		Trunk	-	-	-	-	-
14	Motkila	Leaves	$3345\pm 100$	$480\pm 67$	$527\pm 73$	$4352\pm 240$	$16\pm 2$
		Trunk	$4905\pm 147$	$513\pm 148$	$563\pm 163$	$5981\pm 458$	$36\pm 3$

Concentrations of Na in the samples range from  $10 \pm 2$  to  $1521 \pm 8$  mg/kg. The trunk of the most popular chewing stick plant, Joytun, contains the highest amount of Na. Concentrations of Na in the trunk of Nishinda, Bely-asra, and Khejur are also relatively high, as also in the leaves of Akondo. The specific functions of Na in regard to improving the qualities of human teeth are not yet well known. Na is a foaming agent in dentifrices; it flushes and cleanses cavities by removing food debris and bacterial growth. Thus, this study indicates that the chewing sticks made from Joytun, Nishinda, Bely-asra, Khejur, and Akondo plants may play significant roles as foaming and cleansing agents that help promote better dental health.

### CONCLUSION

The popular chewing stick plants investigated in this study are used in Bangladesh for cleaning teeth and also for therapeutic purposes in dental health care. This study provides useful data on the concentrations of F, Mg, and Na in these plants. People use these sticks partly for the sake of tradition and partly because they are unable to afford to buy expensive modern toothpastes. Because of the generally excellent condition of the teeth of the users of these chewing sticks, the results of the present study may provide useful information concerning the apparent dental value of the sticks. However, further study is necessary to identify other chemical elements and ingredients present in the chewing stick plants and to take into account dietary and other factors to more fully evaluate their effectiveness in promoting dental health.

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