CHAPTER - I

INTRODUCTION
1.1 FLUORIDE - ESSENTIAL FOR HEALTH

Fluoride is considered to be essential for human health especially for prevention of dental caries. Although Bunting (1), Mckay (2) and Dean (3) reported low prevalence of caries among populations suffering from mottled enamel, Dean and his coworkers provided for the first time definite proof of the protective action of fluoride against dental decay (4,5,6). A definite relationship between the fluoride content of drinking water and caries experience, expressed as 'decayed, missing, filled teeth' (DMF) per person was shown by Dean; the higher the fluoride content, the lower the caries experience.

The natural occurrence of fluoride in drinking water and the consequent reduction of the incidence of caries have prompted the health authorities to artificially fluoridate the community water supplies. One of the first experimental clinical studies on artificial fluoridation was carried out...
in two cities viz. Kingston and Newburgh in the New York state. The Kingston water supply had 0.15 ppm of $F^-$ and served as the control sample. Newburgh water was fluoridated artificially to 1.0 -1.2 ppm. Eight to nine years after fluoridation, the (DMF) rate among Newburgh children was found to be 60% lower than that among Kingston children (7,8). In a survey carried out by Timmis on decayed missing and filled teeth (DMF) among five year old U.K. children living in naturally fluoridated and non-fluoridated areas, caries experience of children in fluoridated areas (0.7 to 5.0 ppm) was found to be 46% lower than that observed in non-fluoridated areas (0.1 to 0.5 ppm) (9). Beal and James reported their findings after five and half years of fluoridation of Birmingham water supply in U.K. A reduction in DMF rate of children, drinking fluoridated water to an extent of 42 to 50%, gave substantial proof of fluoride action for prevention of dental caries (10).

Newesely (11) and Perdok (12) expressed that presence of a small quantity of fluoride is necessary for the formation of bone whose mineral component has an apatite structure.
Schwarz and Milne reported that addition of fluoride at a level of 2.5 ppm to a highly purified amino acid based diet (0.40 to 0.46 ppm F⁻) significantly enhanced by 30.8%, the growth rate of young rats kept in trace element controlled isolations, compared to that of the unsupplemented controls. The authors interpreted their observation as an evidence for the essentiality of fluoride for optimal growth in the rats (13). Messer, Singer and Armstrong (14) found a marked impairment in fertility of female mice maintained on a low fluoride diet (0.1 to 0.3 ppm). Such impairment in fertility could be prevented and also reversed by mere supplementation with fluoride (50 ppm F⁻ in drinking water). These authors considered this as a very convincing evidence for fluoride as an essential element for mice. Messer et.al. also observed that rats which were raised on the same low fluoride diet that was employed in the above described fertility studies, became more anemic than those on a high fluoride intake when stressed by chronic bleeding (15).
1.2 TOXICITY DUE TO EXCESS FLUORIDE

Fluoride intake beyond a certain limit is considered to be toxic. The clinical picture in fluoride toxicity is the result of accumulated fluoride as fluoride is known to be a cumulative toxin. Fluoride can accumulate in human body either due to repeated ingestion or due to repeated inhalation. The former situation is encountered among residents of areas consisting of high fluoride concentration in drinking water and food grains. Inhalation is the route of entry in occupational settings such as mining and manufacturing industry. Irrespective of the route of entry, fluoride induced changes are similar. Fluoride toxicity depends for its severity on the factors viz. (i) the total dose ingested (ii) duration of exposure (iii) nutritional status and (iv) body's response (16).

Fluorosis, a well defined clinical entity caused by ingesting excessive amounts of fluoride, is characterized by dental mottling, skeletal and other non-skeletal manifestations which are discussed below.
1.2.1 Dental Fluorosis:

Dental fluorosis or mottled enamel was first described by Eager in 1901 among the emigrants from Italy (17). The term 'mottled enamel' was first introduced by Black et al. who described the disease in detail (18). The permanent teeth are particularly affected, though it occasionally affects the primary teeth also. It was in 1931 that a direct relationship between mottled enamel and fluoride content of water was established (19,20). In India, Dental fluorosis was identified in human beings in Madras Presidency by Shortt and his collaborators in 1937 (21,22). The symptoms of dental fluorosis are shown in Figs. 1.1. a to g.

Dental fluorosis depends on the concentration of fluoride in the drinking water during the years, when the teeth are being formed. After eruption of the teeth, the fluoride content of drinking water has no visible effect on the enamel, but the fluoride content of the teeth increases (23). Fluoride is taken up most rapidly into the tooth during the phases of growth and development. In the early stages of amelogenesis and dentinogenesis, a small amount of
FIG. I.1  VARIOUS STAGES OF DENTAL FLUOROSIS
ACCORDING TO DEAN'S CLASSIFICATION

NORMAL

QUESTIONABLE
FIG.I.I DEAN'S CLASSIFICATION CONTD.

c. VERY MILD

d. MILD
FIG. I.1  DEAN'S CLASSIFICATION CONT'D.

e. MODERATE

f. MODERATELY SEVERE
FIG. I.1  DEAN'S CLASSIFICATION CONT'D.

9- SEVERE
calcium interferes with fluoride transport; the overall systematic uptake of fluoride into enamel and dentine is therefore maximal during the period of formation and calcification (24).

Dean classified the degree of clinically observed mottling into seven categories ranging from normal to severe (24,25). The basis for such a classification of mottling was as follows:

1. Normal - the enamel is translucent, smooth and presents a glossy appearance;

2. Questionable - seen in areas of relatively high endemicity and occasional cases are border line and one would hesitate to classify them as apparently normal or very mild;

3. Very mild - small, opaque paper-white areas are seen scattered irregularly over the labial and buccal tooth surfaces.

4. Mild - the white opaque areas involve at least half of the tooth surface and faint brown stains are sometimes apparent;
5. Moderate - generally all tooth surfaces are involved and minute pitting is often present on the labial and buccal surfaces (brown stains are frequently a disfiguring complication);

6. Moderately severe - pitting is marked, more frequent and generally observed on all tooth surfaces and brown stains, if present, are generally of greater intensity and

7. Severe - the severe hypoplasia affects the form of the tooth and stains are widespread and vary in intensity from deep brown to black. This condition may sometimes be referred to as *corrosion* type of mottled enamel.

On the basis of this classification, Dean et al. attempted to determine a mottled enamel index of the community, which indicates the severity of dental fluorosis as a public health problem. They felt that while a percentage of individuals showing dental fluorosis, exhibited the prevailing condition, yet it does not make clear the differences in the degree of severity among the group studied. Hence they mooted the numerical weighted index of clinical severity for dental fluorosis which is

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computed by giving definite weight to each of the seven classifications used in measuring the effects of fluorosis i.e., 0 to 4. The index is determined by multiplying frequency with weight and dividing by the total number of persons surveyed (26).

Dental fluorosis is the earliest sign of fluoride toxicity. Prolonged exposure to excessive fluoride may damage the enzyme involved in process related to enamel mineralization. This damage creates a porotic tooth susceptible to caries as debris and plaque are entrapped in hypoplastic areas (27). The later in childhood the tooth is formed, the greater are the actual prevalence and degree of severity of dental fluorosis, as severity of fluorosis increases with the increase in fluoride intake and accumulation (28).

In the United States dental mottling is seen only among subjects consuming water containing fluoride at levels in excess of 2.0 ppm (29). But in India dental mottling is reported even when the fluoride in water is less than 1 ppm, the prescribed limit by Indian Council of Medical Research and Bureau of Indian Standards (30-33)
I. 2.2 Skeletal fluorosis:

Endemic skeletal fluorosis is a chronic metabolic bone and joint disease caused by ingesting large amounts of fluoride through water and food. This chronic toxic effect of fluoride on the skeletal system has been described from certain geographical regions of the world. Skeletal fluorosis was reported from the villages in the southern part of India as early as 1937 by Shortt et al. (21). Subsequently, cases of endemic skeletal fluorosis have been reported from other parts of the world. Manifestation of skeletal fluorosis is shown in Fig. I.2.

Early symptoms are generally referable to musculoskeletal system. In more advanced cases, severe arthralgia, arthritis associated with stiffness and limitations of movements of the large joints and stiffness of the spine will be recognized. In the next stage, marked limitation to movements of joints will be evident in the knees, ankle and in the pelvis. In due course, restriction of spinal movements leading to incapacitation occurs. While all vertebral joints are affected to varying extent, cervical and lumbar are affected most.
FIG.1.2 SYMPTOMS OF SKELETAL FLOROSIS
Fluoride toxicity afflicts children more severely and after a shorter exposure to fluoride than adults, due to the greater and faster accumulation of fluoride in the metabolically more active, growing bones of the children. Various factors influencing the course and severity of skeletal fluorosis include the following: (1) drinking water fluoride concentration (2) total daily fluoride intake (3) duration of fluoride ingestion (4) fluctuations in fluoride intake (5) solubility of ingested fluoride (6) age at the time of fluoride ingestion (7) nutritional status (8) individual biological response (9) species effect (10) stress factor (11) complexes with other mineral elements and trace elements (13) alkalinity and hardness of drinking water (14) pH of the gastrointestinal tract and (15) renal functions. The most important factors, however are drinking water fluoride concentration, total daily fluoride intake, duration of exposure and dietary intake of calcium and vitamin D.
Entry of fluoride into bone and tooth mineral: Neuman and Neuman (34) proposed a three-stage mechanism to describe the entry of fluoride ions into the crystal lattice of hydroxyapatite which is the main constituent of bone and tooth mineral. The crystallite was considered to be surrounded by a hydration shell and the incorporation of fluoride into the crystal lattice prefaced by its exchange with one of the ions present in this loosely held hydration sheath. The second stage involved the exchange of fluoride in the hydration shell with an ion at the surface of the apatite crystal. Finally, ions present in the crystal surface might migrate slowly into vacant spaces in the crystal interior during recrystallization.

Chemical analysis of fluoride-rich hard tissues has shown that the incorporation of fluoride slightly alters the chemical composition of bone and tooth mineral (35). The carbonate content is lowered; the Ca/P ratio however remains unchanged. This observation infers that fluoride replaces the carbonate group situated within or at the surface of the crystallites, but does not replace the phosphate group of hydroxyapatite to any great extent. Another observation
that fluoride uptake by bones was reduced by raising the pH of the incubating medium, suggests an analogous competition between fluoride and hydroxyl ions.

Entry of fluoride into the apatite lattice by chemical reaction with hydroxyapatite, followed by precipitation of calcium fluoride and release of phosphate groups, does not appear to take place, at the level of intake of fluoride encountered in the cases of dental or skeletal fluorosis.

1.2.3 Other Forms Of Non-Skeletal Fluorosis:

In his classical book on fluoride intoxication, Roholm pointed to the wide variety of symptoms encountered in chronic fluorosis (36). The soft tissue organs affected by fluoride are named in the following order: aorta, thyroid gland, lungs, kidney, heart, pancreas, brain, spleen and liver. With advancing age, their fluoride content increases. Non-skeletal changes of fluoride occur early in the disease, before dental and skeletal changes become apparent. For this reason early recognition of these manifestations as indicators of fluoride intoxication is essential (37).
Fluoride is known to cross the placental barrier and get deposited in the tissues especially skeletal tissues of the foetus. It has been suggested that calcification of the aorta, due to excessive incorporation of fluoride could account for still births and neonatal infant mortality in endemic areas (38). A number of recent studies have suggested that fluoride may be genotoxic. Fluoride is known to penetrate easily into many organs, tissues and cells, a process facilitated by the chemical properties of the fluoride ion, namely its small ionic radius as well as its high chemical reactivity (39). Gastro intestinal symptom like non-ulcer dyspepsia may be the first symptom of emerging fluorosis in endemic areas. Fluorosis can affect the neurological system (clinical symptoms: nervousness, depression, polydypsia and polyuria) muscular system (muscle weakness, stiffness and pain), the skin which shows allergic reactions and the urinary tract which results in reduction and discolouration of urine (40). Fluoride is likely to have adverse effects on blood group substances which are lodged on erythrocyte membrane as it accumulates in red blood cell membrane (41).
1.3 CONCEPTS OF IONIC FLUORIDE, NON-IONIC FLUORIDE AND TOTAL FLUORIDE

The overall fluoride balance in the human and animal systems involves a dynamic equilibrium where the fluoride levels in the blood play a vital role. An increase of fluoride level promotes its deposition in the bones and by the same token, a decrease in the fluoride level releases fluoride into the blood from the bones. Any small variation of fluoride level in the blood thus appears to automatically alter the direction of these reactions (42). The fluoride present in the blood exists both in the free ionic state as well as in a complex form. It is believed that the non-exchangeable serum fluoride does not change with the fluoride concentration of drinking water (43). It therefore follows that the adverse effects of high fluoride intake are due to its deposition in bones, which in turn is governed by the increase in the fluoride ion concentration in the blood serum.

The equilibrium between ionic and non-ionic fluoride, the two forms of fluoride that exist in human blood, may be destroyed in skeletal fluorosis. The study of changes in
serum ionic fluoride in monkeys during fluorosis revealed that serum ionic fluoride concentration increases as the degree of poisoning advances, which is in conformity with earlier reports. On the contrary, the non-ionic fluoride (NF) decreases and tends to go towards zero, since total fluoride (TF) = $F^-$ + NF. Regarding the increase in ionic fluoride as well as the decrease in serum non-ionic fluoride in skeletal fluorosis of monkeys, the results show that the two fluoride forms in blood interact. Therefore, two forms of fluoride in blood should be considered simultaneously in diagnosis and therapy (44). Results of many studies suggest that serum ionic fluoride is directly correlated with water fluoride concentration (45-47).

Any substance able to complex or combine with fluoride and renders it insoluble, hinders fluoride absorption from the gastrointestinal tract. The presence of calcium, and magnesium in diet in large amounts reduces the absorption of fluoride owing to the formation of less soluble fluorides. The mechanism of fluoride absorption seems to be one of simple diffusion through the entire gastrointestinal tract (48). Reduction of fluoride absorption results in reduced toxicity of fluoride. Fluoride ions reach the blood stream
from the stomach and upper intestinal tract as quickly as 10 minutes after ingestions, the maximum concentration is reached after 60 minutes (23).

1.4 CONTRIBUTION OF WATER TO FLUORIDE INTAKE

1.4.1 Fluoride Bearing Minerals and Water Fluoride:

The occurrence of fluoride rich ground water has been known in peninsular India for over four decades (49). The chief sources of fluoride in ground water are the fluoride bearing minerals in the rocks and sediments that form the reservoir. Fluoride is a component of over 80 minerals. The natural fluoride content of water in different areas varies according to the source of water, the geological formation of the area, the amount of rainfall and the quantity of water lost by evaporation. The various factors that govern the release of fluoride into natural water by the fluoride bearing minerals in the soil and rocks are: (i) the basic chemical composition of the water (ii) the presence and the accessibility of the fluoride and (iii) time of contact between the source minerals and the water (50).
Some of the important fluoride bearing minerals are listed below:


Among the above minerals, fluorite, apatite, and topaz have very wide distribution in India.

Fluoride leaching from fluoride bearing minerals: Igneous and sedimentary rocks contain fluoride bearing minerals. They undergo leaching process and the fluoride is finally leached out into the ground water and surface water.

Sedimentary rocks mainly contribute the rock phosphates. Leaching of sedimentary rocks is particularly active in the oxidising environment. As the leaching proceeds, water dissolves the fluorapatite and franceolite present in the rock phosphates. Finally the fluoride present in these minerals goes into ground water.
Igneous rocks mainly contribute the fluorite, mica, amphibole, silicates (topaz) and a little amount of fluorapatite. Igneous rocks are not directly subjected to the leaching process. They are first subjected to the complicated and prolonged chemical weathering process, which is caused by the action of rain, surface and ground water consisting of the dissolved solids and gases. The presence of water is the basic requirement of chemical weathering. The ground water charged with CO$_2$ is a powerful weathering agent, able to break up nearly all minerals and to form new compounds consisting of bicarbonates, carbonates and sulphates of calcium, magnesium and soluble alkali silicates. On the other hand aluminium, iron and silica remain for the most part in the insoluble residue. Calcium fluoride is the most common form of fluoride occurring in igneous, metamorphic and sedimentary rocks. Solubility studies have indicated that transport of fluoride in aqueous solution is dependent on the solubility of CaF$_2$. In experiments connected with possible conditions related to hydrothermal solutions, the ratio of the concentration of calcium to that of fluoride ions in solution appears to be important.
1.4.2 Fluoride in River Water:

Generally the river water contains less fluoride than the ground water. Only when the river passes through the areas with fluoride bearing minerals, fluoride content in the river water is significant. For example, Kongal river of Nalgonda district, Andhra Pradesh flowing over Archaean granites records 12 ppm of fluoride. The fluoride levels of various river waters in India are shown in the table I.i.
Table I.1 Fluoride levels of various river waters in India (51):

<table>
<thead>
<tr>
<th>River</th>
<th>Location</th>
<th>Fluoride (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ganga</td>
<td>Rishikesh, UP</td>
<td>0.12</td>
</tr>
<tr>
<td></td>
<td>Rajmahal, Bihar</td>
<td>0.35</td>
</tr>
<tr>
<td>Hooghly</td>
<td>Dakshineswar, WB</td>
<td>0.35</td>
</tr>
<tr>
<td>Narmada</td>
<td>Jamtara, Jabalpur, MP</td>
<td>0.10</td>
</tr>
<tr>
<td>Netravati</td>
<td>Mangalore, Karnataka</td>
<td>0.10</td>
</tr>
<tr>
<td>Periyar</td>
<td>Alwaye, Kerala</td>
<td>0.10</td>
</tr>
<tr>
<td>Chalakudiyar</td>
<td>Chalakudiyar, Kerala</td>
<td>0.10</td>
</tr>
<tr>
<td>Mahanadi</td>
<td>Rajim, MP</td>
<td>0.10</td>
</tr>
<tr>
<td>Indravati</td>
<td>Chitrakut, M.P</td>
<td>0.40</td>
</tr>
<tr>
<td>Vekkilu</td>
<td>Chagla marri, A.P</td>
<td>1.50</td>
</tr>
<tr>
<td>Krishna</td>
<td>Vijayawada, A.P</td>
<td>1.00</td>
</tr>
<tr>
<td>Paleru</td>
<td>Rajapet, A.P</td>
<td>1.50</td>
</tr>
<tr>
<td>Sabri</td>
<td>Chitoor, A.P</td>
<td>1.00</td>
</tr>
<tr>
<td>Indravati</td>
<td>Bhopal Patna</td>
<td>1.00</td>
</tr>
<tr>
<td>Muneru</td>
<td>Khammam, A.P</td>
<td>2.00</td>
</tr>
<tr>
<td>Bhima</td>
<td>Yadgir, Karnataka</td>
<td>1.50</td>
</tr>
<tr>
<td>Chitravati</td>
<td>Dharmavarm, A.P</td>
<td>3.50</td>
</tr>
<tr>
<td>Pennar</td>
<td>Panidi, A.P</td>
<td>1.50</td>
</tr>
<tr>
<td>Tambraparni</td>
<td>Tirunelveli, Tamil Nadu</td>
<td>3.50</td>
</tr>
<tr>
<td>Vaigai</td>
<td>Madurai, Tamil Nadu</td>
<td>1.00</td>
</tr>
<tr>
<td>Amaravati</td>
<td>Darapuram, Tamil Nadu</td>
<td>1.00</td>
</tr>
</tbody>
</table>
1.4.3 Fluoride in Rain Water:

In rain water presence of significant amount of fluoride is possible only in industrialised areas with fluoride emitting industries like phosphate fertilizer industry, aluminium manufacturing industry, poly tetra fluoro ethylene industry etc. Otherwise rain water consists of very low amount of fluoride.

1.4.4 Ionic and Complex Fluorides in Water:

Iron and aluminium ions react with fluoride forming complexes like $\text{ALF}_6^{3-}$ and $\text{FeF}_6^{3-}$. Hence if iron and aluminium are present in a particular water sample, part of the fluoride present will exist in the form of complex-fluoride and part of the fluoride will be in ionic form. Presence of iron and aluminium reduces the concentration of ionic fluoride in water.

1.5 CONTRIBUTION OF FOOD TO FLUORIDE INTAKE

There have been many incidences of fluorosis where fluoride in food was identified as the main source of fluorides. One extreme example is the prevalence of dental
fluorosis among the native people of South Atlantic Island consuming water with 0.2 ppm fluoride only and fish with 7mg/kg of fluoride (52).

Wei Zan - dao et.al. discovered skeletal fluorosis in the Bijie country in the western part of Guizhou province, China where fluoride in the water averaged to 0.1Sppm (53)- The average fluoride consumed through food to an extent of 7.1, mg/day, was established as the cause of the disease. The authors pointed out that dental fluorosis has been shown to prevail in Vietnam and Thailand also where water fluoride is low but food stuffs consumed by people contain high levels of fluoride.

Dietary ingestion of fluoride has become a matter of great importance in recent years due to its possible adverse effects on health.

1.6 OTHER FACTORS INFLUENCING FLUORIDE TOXICITY

Studies by Irving suggested that population living on restricted and inadequate diets and exposed to endemic fluorosis may respond with more severe signs of fluorosis
than well nourished healthier population (54). Singh suggested that the decreased calcium content and hardness and an increased sodium content and alkalinity of water in the endemic areas may be contributing to the increased fluoride toxicity (55).

The effect of climatic variations on water and fluoride intake, especially in warmer areas has been considered by Galagan (56), who pointed out that intrinsically the amount of water required is influenced by the body size and weight, by the kind of food eaten, by habit patterns and by physical activity. Of the climatic factors affecting water intake, mean annual temperature, excessive day time temperatures, radiant heat gain, relative humidity and wind movement are of the greatest importance. Factors like temperature which promote increased ingestion of water obviously increase the total fluoride ingested. In tropics, this disease manifests at an early age, a fact which may be attributable to greater water intake.
Fluoride is known to be a bone seeking element. For a person with a small stature with a consequent small skeleton, the amount of total fluoride that would be toxic may be expected to be less than that for a person with a big stature, with a relatively big skeleton. The average weight of a well-nourished western adult is around 70kg, while the mean body weight of an agricultural labourer in India who is exposed to the risk of skeletal fluorosis rarely exceeds 55kg.

About 80% of the fluoride normally occurring in the human diet is absorbed. According to Hodge and Smith (57) fluoride absorption is reduced to 50% if calcium compounds are added. Calcium renders fluoride less soluble causing fecal excretion to increase. Some workers suggested that the low intake of calcium in the diet leads to greater retention of fluoride in the bones.

The type of staple consumed might also modify fluoride toxicity. Results of some metabolic studies have shown that diets based on great millet resulted in significant increase in retention of fluoride, mean increase in retention being
12.2% as compared to diets based on rice at identical intakes of fluoride. The increased retention was not due to changes in the intestinal absorption of fluoride, but in the urinary excretion of fluoride. It may partly be due to differences in the trace element profile of the two staples (58). It has been reported that molybdenum promotes fluoride retention (59) and the molybdenum content of great millet has been found to be somewhat higher than that in rice.

Serum ionic fluoride decreased in monkeys administered with calcium and vitamin D, which showed that vitamin D and calcium are useful in alleviating fluorosis (44). Results of diet surveys have indicated that low levels of calcium and vitamin C in the diet may be related to increased toxicity of fluoride. Controlled studies carried out at the National Institute of Nutrition, Hyderabad, India using monkeys, have in fact, provided the experimental proof of low amounts of calcium and vitamin C in the diets predispose to the development of experimental skeletal fluorosis. The precise mechanism is however not known (30). Yu and Hwang have reported that high intake of vitamin C mitigated the effects of fluoride in mice (60). The therapeutic effects
of calcium against fluoride have also been long known. Narasinga Rao et al. (61) have studied calcium turnover in endemic fluorosis and reported its mitigating influence after calcium ingestion in fluoride poisoned rats. Chi i n >y et al. (62) reported recovery of all activities in rabbits affected by sodium fluoride, by the combined treatment, of ascorbic acid and calcium. It is therefore suggested that the effects of administration of combination of ascorbic acid and calcium might be highly beneficial in the recovery of fluoride treated animals. This has a direct bearing on reducing the suffering of fluorosis-afflicted human, also.

1.7 FLUORIDE TOXICITY-WORLD PICTURE

Incidence of fluorosis has been reported both in developed and developing countries like USA, Italy, Holland, Spain, France, Germany, China, Japan, and several African and South American countries.

In USA, mottling is reported to be associated with at least 3 to 4 ppm of fluoride level at which many workers in India have recorded cases of skeletal fluorosis. Lower
Mascheroni and Reussi (65) reported abnormal skeletal changes attributed to the ingestion of fluoride in drinking water in Argentina. Steyn reported chronic fluoride poisoning in South Africa (66). Rosenzweig reviewed the prevalence of endemic fluorosis in Israel (67). Sea fish and tea are the important items in the Israeli diet which may serve as additional sources of fluoride.

In Kenya endemic fluorosis has been chronic for over 50 years. Over 30% of Kenya's population suffers from dental fluorosis and in isolated regions where the people depend on ground water for domestic use, nearly 100% of the population manifests varying degrees of dental fluorosis. Drinking water is Kenya's major source of fluoride ion, especially in those regions of the country associated with volcanic rocks and hot springs (68,69). Other important sources include food and drinks (70,71).

In 1976, Wei zan-dao et al. discovered skeletal fluorosis in the Bijie country in the western part of Guizhou province where the fluoride level in the water averaged 0.15 ppm. The high fluoride content of food was
temperature, ingestion of smaller quantities of water and better nutritional condition are undoubtedly the factors responsible for the absence of crippling disabilities in countries such as Great Britain and the USA. This is well borne out by the studies of Leone (63) and Stevenson and Watson (64). In a review of approximately 170,000 X-ray examinations of the spine and pelvis of patients, mostly residents of Texas and Oklahoma, osteosclerotic changes were noted in only 23 by Stevenson and Watson. Each of these patients lived his/her entire life in an area in which the water contained fluoride in concentrations ranging from 4 to 8 ppm. No osteosclerotic changes were evident in this study in persons whose drinking water contained less than 4 ppm fluoride.

In Japan people consume large amount of marine products, such as fish having high fluoride content, as their main protein source. This may cause dental fluorosis at a comparatively lower water fluoride concentration. In Kyoto district, dental fluorosis was found in places where the water contained about 0.8 ppm. Many cases of rather severe dental fluorosis can be seen in districts where the fluoride content of water is above 1.1 ppm.
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temperature, ingestion of smaller quantities of water and better nutritional condition are undoubtedly the factors responsible for the absence of crippling disabilities in countries such as Great Britain and the USA. This is well borne out by the studies of Leone (63) and Stevenson and Watson (64). In a review of approximately 170,000 X-ray examinations of the spine and pelvis of patients, mostly residents of Texas and Oklahoma, osteosclerotic changes were noted in only 23 by Stevenson and Watson. Each of these patients lived his/her entire life in an area in which the water contained fluoride in concentrations ranging from 4 to 8 ppm. No osteosclerotic changes were evident in this study in persons whose drinking water contained less than 4 ppm fluoride.

In Japan people consume large amount of marine products, such as fish having high fluoride content, as their main protein source. This may cause dental fluorosis at a comparatively lower water fluoride concentration. In Kyoto district, dental fluorosis was found in places where the water contained about 0.8 ppm. Many cases of rather severe dental fluorosis can be seen in districts where the fluoride content of water is above 1.1ppm.
Mascheroni and Reussi (65) reported abnormal skeletal changes attributed to the ingestion of fluoride in drinking water in Argentina. Steyn reported chronic fluoride poisoning in South Africa (66). Rosenzweig reviewed the prevalence of endemic fluorosis in Israel (67). Sea food and tea are the important items in the Israeli diet which may serve as additional sources of fluoride.

In Kenya endemic fluorosis has been chronic for over 30 years. Over 30% of Kenya's population suffers from dental fluorosis and in isolated regions where the people depend on ground water for domestic use, nearly 100% of the population manifests varying degrees of dental fluorosis. Drinking water is Kenya's major source of fluoride ion, especially in those regions of the country associated with volcanic rocks and hot springs (68,69). Other important sources include food and drinks (70,71).

In 1976, Wei zan-dao et.al. discovered skeletal fluorosis in the Bijie country in the western part of Guizhou province where the fluoride level in the water averaged 0.18ppm. The high fluoride content of food was
established as the cause of the disease (53). These workers pointed out that dental fluorosis has been shown to prevail in Vietnam and Thailand where water is low in fluoride, but food contains high level of fluoride.

In a survey of 662 British school children between 5 and 15 years of age, 92.5% were found to drink tea regularly. The total daily fluid intake per child is 1924 ml. Thirty varieties of tea consumed in Great Britain were found to contain fluoride in the range from 121 to 260 mg F~/kg with an average of 186.1 mg F~/kg or 0.52 mg per cup. The daily fluoride intake through tea in children up to the age of 15 was calculated to be 1.26 mg as compared to 2.55 mg in adults (72). Webb-peploe and Bradley reported a case of crippling fluorosis with neurological complications in a Hampshire-man whose only known exposure to F~ intake was through consumption of tea (73). Thus fluoride in tea contributes materially to the daily fluoride intake in the British population and might well constitute a hazard to health.
In Spain, wine contaminated by F\textsuperscript{−} produced a bone disease in chronic alcoholics characterized by osteomalacia which is vastly different from the other forms of fluorosis usually encountered (74). In Sicily, where much sea food is eaten, 45% of the patients with skeletal fluorosis had gastro-intestinal disorders (75). In Jordan, the severity of dental fluorosis in children may be related to excessive drinking of tea daily (76).

1.8 FLUORIDE TOXICITY–INDIAN SCENE

The study of human fluorosis is more than fifty years old in India. Since 1937 when fluorosis was first reported by Shortt et.al., there have been a number of reports from different parts of the country (49,77–84). Endemic fluorosis has been detected in 15 states of India by the National Drinking Water Mission. They are: Andhra Pradesh, Bihar, Delhi, Gujarat, Haryana, Karnataka, Maharashtra, Orissa, Punjab, Madhya Pradesh, Rajasthan, Tamil Nadu, Uttar Pradesh, Jammu and Kashmir and Kerala. Currently at least 25 million people are estimated to be afflicted with fluorosis and an equal number are said to be at risk (40).
The prevalence of crippling endemic skeletal fluorosis in some of the states of India is a serious public health problem. The tremendous increase in the annual use of fluorides compared with that thirty years ago, together with the increased diversity of their use, has correspondingly increased the hazards associated with these materials. In India skeletal fluorosis is due to excess ingestion of fluoride through drinking water^ the fluoride level of which is high naturally (85). The problem of skeletal fluorosis attracted attention in 1969 (86) when it was discovered in children and adults who resided since their birth in the endemic fluorosis areas in the district Rai Bareli of the state of Uttar Pradesh. The drinking water in these areas contain 24 to 26 ppm of fluoride.

The disease appears to be largely of hydro-geochemical origin. Epidemiological observations on the nutritional status of the Indian population affected with fluorosis indicate that dietary deficiencies and nutritional imbalances, particularly the low intakes of protein, calcium and vitamin D, enhance the severity of fluoride toxicity and its effects on bone metabolism(87).
Hydro-Geochemical basis of endemic fluorosis in India (87)

Rocks
(Igneous)

Sedimentary, Metamorphic
country rocks, Earth's crust

Diagenesis:Lithification:Remobilization:
Hydrothermal Reaction: Weathering: Leaching:

Fluid phases rich in fluoride bearing minerals
Fluorite: Cryolite: Apatite: Phosphate: Topaz

Dissolution: Dispersion: Distribution

Fluoride release: High fluoride in
Hydro-geochemical environment

Ground water  Surface water  Soils
Drinking water  Prolonged Intake

Dietary deficiencies; Nutritional imbalances: Protein
Calcium, Vitamins; Soft and alkaline water; Increased water
consumption; Body size; Habits; Sweating; Climate;
Interaction of other elements (Mo, Cu, Ca)
affect fluoride toxicity.
The world's fluoride reserves under the ground are estimated to be 85 million tons of which nearly 12 million tons are located in India. Fluoride bearing minerals are widely distributed in India. There is a close connection between the distribution of fluoride bearing minerals and prevalence of endemic fluorosis (87). Pandit (49) opined that the wide variation in the nutritional status in the endemic fluorosis areas of India may augment or exaggerate the harmful effects of excessive fluoride storage.

1.9 NEED AND OBJECTIVES OF THE PRESENT STUDY

It is now well recognized that besides fluoride concentration in drinking water, several other factors determine the magnitude and severity of fluoride toxicity. For example, higher intakes of calcium, C and D vitamins through diet are found to reduce the prevalence and degree of fluorosis to a considerable extent.

Nevertheless, no systematic studies have so far been carried out to correlate the ill-effects of excess fluoride in drinking water with various other chemical parameters and
constituents of the same drinking water sources. Such studies may lead to certain generalizations as to which chemical parameters and constituents actually influence fluoride toxicity at a given fluoride concentration of drinking water. Based on such generalizations, one can guide people to select the relatively less harmful drinking water sources with identical fluoride concentrations.

Objective 1: One of the objectives taken up in the present study is therefore, to analyse the drinking water sources of two control areas and three fluorotic areas, for various chemical parameters like hardness, alkalinity and pH and chemical constituents viz. fluoride, chloride, sulphate, phosphate, calcium, magnesium, strontium, iron, aluminium, sodium, potassium, copper, zinc, molybdenum and manganese, and analyse the problem of fluoride toxicity in the light of the results obtained.

Objective 2: There have been many reports in the literature that contribution of fluoride
through food grains grown in fluorotic areas is significant. It was therefore felt necessary to analyse the fluoride contents of the chief staple food grains grown in the control and fluorotic areas, viz. great millet, finger millet, pearl millet, little millet and paddy and study the contribution of fluoride through these food grains. This is the second objective of the present work.

Though certain studies have been reported in the literature about the total fluoride intake and intake of fluoride through diet, no quantitative attempts have been made to develop the methodology to evaluate exactly the contribution of water and food to the total daily intake of fluoride. For example, while mentioning the dietary intake, some authors have included food alone, others food and beverages at meal time, and still others, food, beverages and water at and between meals. These differences are crucial in assessing the total dietary F~ intake because of the use of water (containing even the modest amount of fluoride) in the preparation of food and beverages. Moreover, the water used for preparing different varieties
of foods and beverages was not taken into consideration while calculating the relative contribution of fluoride through water.

Objective 3: It was a felt need to develop a suitable methodology for the evaluation of total fluoride intake, fluoride intake through water and through food alone per day by human subjects. Development of such a new methodology is the third objective of the present study.

Objective 4: Based on this methodology it was planned to study the relative contributions of water and food to the total daily fluoride intake by human subjects living in control and fluorotic areas. This is the fourth objective of the present work. Such a comparative study is expected to resolve to a greater extent, the question whether we can treat the effect of water fluoride and food fluoride on equal grounds.
Objective 5: The fifth objective of the present study is to develop a suitable methodoloy for the analysis of the total daily intake of the various elements like calcium, magnesium, strontium, iron, aluminium, sodium, potassium, copper, zinc, molybdenum, and manganese by the subjects living in control and fluorotic areas and the daily intake of these elements by them through water and through food alone.

Objective 6: Studies correlating fluoride toxicity with the total daily intake of several other elements by human subjects living in control and fluorotic areas are scanty in literature. No such study has been reported so far under Indian conditions. So it is set to be the sixth objective of the present work to correlate the fluoride toxicity with the total daily intake of the elements listed above, under Indian conditions.