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ACKNOWLEDGEMENTS. I acknowledge the critical comments and suggestions of Michel Danino. I thank Archaeological Survey of India for cooperation during studies on the Delhi Iron Pillar.

Received 12 December 2007; revised accepted 1 August 2008

Determination of calcium dose for minimizing fluoride bioavailability in rabbits

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Fluorosis results from excess fluoride ingestion and is characterized by marked abnormalities of bones and teeth. Most of the absorbed fluoride from the diet and drinking water can readily enter into the bones and teeth thus the bones become hypertropic and coarse but are fragile. Earlier studies indicated that calcium binds with fluoride forming an insoluble calcium fluoride in the gastrointestinal tract; thus the adverse effect of fluorosis may decrease. Quantification of calcium needed to reduce excess fluoride absorption is necessary for the treatment of fluorosis. In this study different doses of calcium in the form of calcium carbonate were administered to albino rabbits and it was observed that significant decrease in fluoride bioavailability was observed in the group of animals administered with 11.4, 14.3, 17.2, 21.4 and 50 mg calcium per kg body wt. This calcium dose level is equivalent to 1200, 1500 and 3500 mg of calcium required for a 70 kg body wt human beings.

Keywords: Absorption, bioavailability, calcium carbonate, fluoride, fluorosis.

The primary adverse effects associated with chronic, excess fluoride intake are dental and skeletal fluorosis. There is much evidence in the literature indicating a decreased fluoride concentration and a greater faecal excretion of fluoride when sodium fluoride was administered with high calcium diet. Prevention of gastrointestinal absorption of fluoride by calcium is due to the formation of insoluble calcium fluoride accounted for a decreased serum fluoride level in calcium carbonate-treated animals. Calcium supplementation does not promote urinary excretion of fluoride because in earlier studies no significant changes were observed in the concentration of urine when animals received sodium fluoride and calcium-supplemented diet. Amount of calcium required to reduce excess fluoride was studied in the present study using albino rabbits. Rabbits were chosen because they are small yet large enough to permit blood samples to be withdrawn over the course of several hours and also rabbits are mammalian type of species, so the absorption trend may be similar to human beings. Albino rabbits were also used in earlier studies to elucidate the fluoride absorption. Some of the important pharmacokinetic parameters like percentage of fluoride absorption, elimination half

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life ($T_{1/2}$), elimination rate constant ($K_E$), peak plasma concentration ($C_{max}$), time for peak plasma concentration ($T_{max}$) and relative bioavailability of fluoride were studied to elucidate the time course of the absorption, distribution, metabolism and excretion of the fluoride. The term bioavailability was defined as the rate and extent to which fluoride is absorbed from a product and reaches the systemic circulation. Effect of calcium dose on fluoride bioavailability was also assessed by in vitro analysis.

Eighty male healthy young albino rabbits, having average weight around 3 kg with similar demographic characteristics were chosen and divided into six groups (A to F; Table 1). The animals received a control diet and deionized water ad libitum and received less than 1 mg fluoride from the control diet and deionized water per day. To ensure consistent gastrointestinal conditions, the rabbits were kept fasting for 12 h prior to administration of the fluoride and calcium doses. The fluoride dose was given in solution form prepared from pure sodium fluoride AR grade sample and the calcium dose in the form of calcium carbonate in double distilled water was given as suspension using gastroagvage. Blood samples were drawn through marginal ear vein puncture, from each animal by using sterile syringes at pre dose and at 15, 30, 60, 120, 180, 240 and 300 min time intervals of post dose and collected in heparinized tubes (Table 2). The blood samples were centrifuged and plasma was harvested. The plasma fluoride level was measured by Hall method using fluoride ion selective electrode Orion 9609. The slope value obtained from the calibration curve was -58.8 mV, which is the best slope value for the estimation of fluoride (Figure 1).  

The relative bioavailability of fluoride was calculated using the following equation

$$K_E = \frac{0.693}{T_{1/2}}.$$  

Analysis of variance within the group was performed and $p > 0.001$.

Plasma fluoride level after oral administration of fluoride reached maximum ($T_{max}$) at 30 min in all the groups after which it started to decline. The oral dose of 2 mg fluoride/kg body wt was chosen, since earlier studies indicated that this dose produces mild degree of enamel hypomineralization similar to human dental fluorosis. Sodium fluoride is generally used as a standard for assessing relative fluoride bioavailability from foods and other products and its bioavailability is arbitrarily set at 100%. Calcium carbonate was chosen because it has higher elemental calcium than most other potential food grade calcium sources. Calcium carbonate is a preferred calcium source for fortifying foods as it is absorbed readily from the gastrointestinal tract. Intestinal fluoride absorption seems to depend on gastric acidity because fluoride is absorbed readily in the gastrointestinal tract after the formation of soluble hydrogen fluoride under normal acidic pH in the stomach. Antacids have been shown to inhibit absorption of fluoride. Though aluminium, calcium, magnesium and sodium-based antacids can reduce stomach pH, sodium forms highly soluble NaF, aluminium imparts toxicity and magnesium fluoride is more soluble than calcium fluoride.  

Therefore, calcium carbonate is a suitable antacid and it reduces considerably the formation of hydrogen fluoride and decreased absorption of fluoride in calcium carbonate-treated animals. Calcium was co-administered with fluoride because earlier studies showed that serum fluoride level decreased markedly when calcium was simultaneously administered

<table>
<thead>
<tr>
<th>Group</th>
<th>Fluoride (mg/kg body wt)</th>
<th>Calcium (mg/kg body wt)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>2</td>
<td>-</td>
</tr>
<tr>
<td>B</td>
<td>2</td>
<td>11.4</td>
</tr>
<tr>
<td>C</td>
<td>2</td>
<td>14.3</td>
</tr>
<tr>
<td>D</td>
<td>2</td>
<td>17.2</td>
</tr>
<tr>
<td>E</td>
<td>2</td>
<td>21.4</td>
</tr>
<tr>
<td>F</td>
<td>2</td>
<td>50.0</td>
</tr>
</tbody>
</table>

Figure 1. Mean plasma fluoride after administration of fluoride alone and upon co-administration with calcium in rabbits.
with sodium fluoride and no significant reduction was observed on administration of calcium carbonate and sodium fluoride separately.\(^{2,3,19}\)

The results show that the relative bioavailability of fluoride is reduced to 50% only in the group F which received 50 mg calcium/kg body wt in the form of calcium carbonate. This dose of calcium is equivalent to 3500 mg of calcium in 70 kg body wt human beings which is much higher than the normal recommended dietary allowance of 1000 mg calcium\(^{20}\). Rabbits in the group E received only 21.4 mg calcium as calcium carbonate which is less than half of what the group F animals received, but the percentage of fluoride reduction is 35. This suggests that, desired reduction in bioavailable fluoride is effected even at lower calcium dose. The elimination half-life period \(T_{1/2}\) is more when the bioavailable fluoride is low, which may be due to low concentration gradient. If the elimination half-life period is more, then the rate of elimination is less which is evident from the \(K_{e}\) values.

The present study reveals that a dose of calcium as low as 17.2 mg/kg body wt, which is equivalent to 1200 mg calcium for a 70 kg body wt human beings, is capable of reducing the bioavailable fluoride to about 31%. Excess intake as high as 50 mg/kg body wt reduced the bioavailable fluoride to 50%, and the difference in percentage of reduction is not appreciable when compared to the huge dose. Moreover, such high doses may lead to other health complaints. Thus 31% reduction in bioavailable fluoride was achieved in rabbits administered with calcium little higher than what is recommended for human beings.


\(^*\) SD, standard deviation.
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Municipal solid waste disposal in Pune city – An analysis of air and groundwater pollution

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The urban centres of India produce 120,000 t of solid waste per day. The unscientific disposal of solid waste creates many environmental and pollution problems. The 1000–1200 t of solid waste in Pune municipal area is disposed at Urali-Devachi village. This communication reveals the problems of air and groundwater pollution caused due to unscientific disposal of solid waste observed at Urali-Devachi village. The average annual emission of SPM found at the disposal site is 1708.3 μg/m². The average annual emission of SO₂ in landfill site is 285.33 μg/m² and NOₓ is 234.07 μg/m². The SPM, SO₂ and NOₓ concentrations are more than those stipulated by the Indian standard limit. The leachate samples are acidic and corrosive in nature. The COD and BOD of tested well-water samples III and IV are 834, 703 mg/l and 716, 412.16 mg/l respectively. The concentration of sodium in well-water samples III and IV is 2437 and 2612 mg/l respectively. The COD, BOD and sodium concentration in well water are higher than the limits of IS: 2291. Rag-pickers, workers, vehicle drivers, and those residing in the nearby areas are continuously exposed to air pollution. It has been found that leachates originate from solid waste landfill-contaminated groundwater. Well water found in Urali-Devachi village is not safe for drinking, outdoor bathing, propagation of aquatic life, industrial cooling and for irrigation. If this municipal solid waste landfill continues, it may create a toxic bomb in future. Conversion technology entailing conversion of solid waste to energy is proving helpful for decomposing solid waste without affecting the environment. Also, the community-based solid waste decomposition is an ideal and a safe disposal method, biological decomposition of segregated organic waste is more beneficial for solid waste management, as it easily converts waste to valuable fertilizer.

Keywords: Air and groundwater pollution, municipal solid waste, land fill, leachate.

DURING the flourishing of civilizations from 300 to 1000 bc, solid waste was placed in large pits with a layer of soil cover. The municipal refuse is referred as any waste that is generated by the domestic and industrial sectors in municipality. The municipal solid waste (MSW) is heterogeneous in nature and contains paper, plastic, rag, metal, glass piece, ash and compostible matter. In addition, other substances like scrap materials, waste papers, dead animals, discarded chemicals, paints, hazardous hospital waste and agricultural residue are also categorized under MSW. Till date, the biomedical waste generated from clinics, hospitals, nursing homes, pathological laboratories, blood banks and veterinary centres has also been disposed along with MSW at the dumpsite.

In Indian cities solid waste generation rate is on the increase. The average per capita solid waste generation in India has increased from 0.32 kg/day in 1971–73 to 0.48 kg/day in 1994 (Table 1). Daily per capita generation of MSW in India ranges from about 100 g in small towns to 500 g in large towns. In 1995, EPRTI, Hyderabad showed that 23 big Indian cities generated 11 million tonnes (mt) of solid waste every year. But now urban centres of India produce 1,20,000 t of solid waste each day; this is expected to reach 300 mt per annum by the end of 2047. In 1996, NEERI, Nagpur carried out a study

ACKNOWLEDGEMENT. We thank UGC, New Delhi for financial assistance.

Received 13 June 2007; revised accepted 14 August 2008

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CURRENT SCIENCE, VOL. 95, NO. 6, 25 SEPTEMBER 2008 773