

HEALTH/BIOLOGICAL EFFECTS

Regression analysis of cancer incidence rates and water fluoride in the U.S.A. based on IACR/IARC (WHO) data (1978-1992). International Agency for Research on Cancer

Age-specific and age-standardized rates (ASR) of registered cancers for nine communities in the U.S.A. (21.8 million inhabitants, mainly white) were obtained from IARC data (1978-82, 1983-87, 1988-92). The percentage of people supplied with "optimally" fluoridated drinking water (FD) obtained from the Fluoridation Census 1985, U.S.A. were used for regression analysis of incidence rates of cancers at thirty-six sites (ICD-WHO, 1957). About two-thirds of sites of the body (ICD) were associated positively with FD, but negative associations were noted for lip cancer, melanoma of the skin, and cancers of the prostate and thyroid gland. In digestive organs the stomach showed only limited and small intestine no significant link. However, cancers of the oral cavity and pharynx, colon and rectum, hepato-biliary and urinary organs were positively associated with FD. This was also the case for bone cancers in male, in line with results of rat experiments. Brain tumors and T-cell system Hodgkin's disease, Non-Hodgkin lymphoma, multiple myeloma, melanoma of the skin and monocytic leukaemia were also correlated with FD. Of the 36 sites, 23 were positively significant (63.9%), 9 not significant (25.0%) and 4 negatively significant (11.1%). This may indicate a complexity of mechanisms of action of fluoride in the body, especially in view of the coexisting positive and negative correlations with the fluoridation index. The likelihood of fluoride acting as a genetic cause of cancer requires consideration.

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Keywords: Cancer in USA, Epidemiology, Water fluoridation.

Source: J Epidemiol 2001 Jul;11(4):170-9.

Editorial note: For related abstract, see p 199 of this issue.

Vitamin D ameliorates fluoride-induced embryotoxicity in pregnant rats

We have evaluated the ameliorative effect of vitamin D on fluoride-induced embryotoxicity in pregnant rats. Oral administration of sodium fluoride (NaF; 40 mg/kg body weight) from days 6 to 19 of gestation caused, as compared with control, significantly lowered body weight, feed consumption, absolute uterine weight and number of implantations. As compared with the control, higher incidence of skeletal (presence of wavy ribs, 14th rib, dumbbell-shaped 5th sternbrae, incomplete ossification of skull) and

visceral (subcutaneous haemorrhage) abnormalities was recorded in the fetuses of fluoride-treated pregnant rat. Vitamin D (2 ng/0.2 ml olive oil/animal/day po) treatment significantly ameliorated the fluoride-induced reductions in body weight, feed consumption and absolute uterine weight. As compared with fluoride-treated alone, the total percentage of skeletal and visceral abnormalities observed in fetuses was significantly lowered in fluoride plus vitamin D-treated animals. These findings suggest that vitamin D treatment significantly reduced the severity and incidence of fluoride-induced embryotoxicity. The ameliorative effect of vitamin D against skeletal and visceral abnormalities could be due to stimulation of intestinal absorption of calcium and phosphate, thus raising the plasma calcium and phosphate concentrations.

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Keywords: Embryotoxicity, Fluoride toxicity, India, Rat study, Fluoride treatment, Vitamin D.

Source: *Neurotoxicol Teratol* 2001 Mar-Apr;23(2):197-201.

Dental caries among 10- to 14-year-old children in Ugandan rural areas with 0.5 and 2.5 mg fluoride per liter in drinking water

The purpose of this study was to report on dental caries among Ugandan children residing in rural areas with either a low or high fluoride concentration in the drinking water, and to assess factors associated with caries. A random sample of 481 children aged 10-14 years was selected from Mpondwe (n = 81) and Kyabayenze (n = 82) in the Kasese district with 0.5 mg and from Mutolere/Kagera (n = 163) and Kabindi (n = 155) in Kisoro with 2.5 mg fluoride/L in the drinking water. The children were examined for caries using the DMFT index as described by the World Health Organization in 1987. The mean DMFT was 0.34 in the whole material. In one low fluoride area, Kyabayenze, all children were caries-free compared to 75% to 86% in the other areas. In Kyabayenze, tea with sugar was taken significantly less frequently than in the other low-fluoride area. In the high-fluoride district, age and consumption of tea with sugar were positively and significantly correlated with caries. Multivariate analyses showed age to be the only significant risk indicator.

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Keywords: Dental caries, Epidemiology, Fluoride in water, Uganda.

Source: *Clin Oral Investig* 2001 Mar;5(1):45-50.

Multigenerational evaluation of sodium fluoride in rats

Since the mid 1940s, fluoride has been added to tap water in American communities in an effort to reduce the incidence of dental caries in the population. When the levels of fluoride in drinking water were tested and set, water was the only measurable source of fluoride for most communities. Now, adults and children ingest fluoride with foods and beverages prepared with fluoridated water, and they are exposed to fluoride-containing dental products. As a result, exposure to fluoride is greater than had been anticipated. In the early 1990s, the existing reproductive studies were reviewed in several reports and were considered to be inadequate to determine potential reproductive or developmental hazards. The effects of sodium fluoride ingestion at 0, 25, 100, 175, or 250 ppm in drinking water measured in rats throughout three generations are reported here. Feed and fluid consumption, body weights and clinical signs were recorded at regular intervals. Decreased fluid consumption observed at 175 and 250 ppm was attributed to decreased palatability and did not affect reproduction. No cumulative effects were observed in the three generations. Mating, fertility and survival indices were not affected. Organ-to-body-weight ratios and organ-to-brain weight ratios were not affected. Sodium fluoride up to 250 ppm did not affect reproduction in rats.

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Keywords: Fluoride toxicity, Multigenerational rat study.

Source: Food Chem Toxicol 2001 Jun;39(6):601-13.

Editorial note: Contradictory findings indicating fetotoxic effects with 200 ppm of NaF were published in Fluoride 2000;33:79-84.

Urinary fluoride excretion in children drinking fluoridated school milk

Objective: To determine fluoride excretion under various conditions of fluoride intake and to estimate the fractional urinary excretion of fluoride in individual children participating in a school milk fluoridation scheme.

Design: In the first part of the study, individual urine samples were collected from each of eight 4 to 5-year-old children for a continuous period of 55 h. For each child ($n = 8$) and for each day ($n = 3$), the maximum urinary fluoride concentration (p.p.m.F), the maximum fluoride excretion rate (microgram F/h), and the total daily fluoride excretion (mg) were calculated. The second part of the study was carried out to determine the 24 h fractional per-

centage of fluoride excreted following administration of a known dose of fluoride in the absence of other sources.

Results: Under usual conditions of fluoride intake (i.e. milk containing 0.5 mg fluoride, customary diet and toothbrushing with fluoride toothpaste) the children's daily fluoride excretion was 0.33 mg. The fractional urinary fluoride excretion of a 0.5-mg fluoride tablet was 30%.

Conclusions: It is concluded that the children's mean 24 h fluoride excretion was somewhere between that reported in low fluoride conditions and that reported in optimally fluoridated areas. The fractional urinary fluoride excretion was found to be in agreement with the findings of other workers.

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Keywords: Children, Fluoride in milk, Fluoride in urine.

Source: Int J Paediatr Dent 2000 Dec;10(4):260-70.

Prevalence of dental fluorosis in the primary dentition

Objectives: This paper presents data on the prevalence of primary tooth fluorosis among children residing in Iowa, and the relationships between fluorosis prevalence and selected measures of fluoride exposures.

Methods: Children in the study cohort were followed prospectively during the first year of life. This study assessed their home water fluoride concentrations and use of fluoride dentifrice or dietary fluoride supplements. A total of 637 children (320 females and 317 males) were examined for fluorosis using a modification of the TSIF index at age 4 1/2 to 5 years, with 90.4 percent having intact primary dentitions.

Results: Seventy-four children (11.6%) had fluorosis present on one or more of their primary teeth, and Seventy-one children (11.1%) had two or more teeth affected. Nearly all fluorosis was mild, with the primary second molar teeth most commonly affected. Fluorosis was significantly associated with higher water fluoride concentration, but not with the use of dentifrice or fluoride supplements.

Conclusions: The results of this study show that primary tooth fluorosis is relatively uncommon, but is most frequently seen on the posterior teeth, particularly the primary second molars, which form at later stages of development. This finding suggests that primary tooth fluorosis is mostly a post-natal phenomenon, and is associated with higher water fluoride levels.

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Keywords: Dental fluorosis, Primary dentition, USA.

Source: J Public Health Dent 2001 Spring;61(2):87-91.

DIETARY FLUORIDE

Patterns of fluoride intake from birth to 36 months

Objectives: Dental fluorosis prevalence has increased in the United States, Canada, and other nations due to the widespread availability of fluoride in many forms, with fluoride ingestion during the first three years of life appearing most critical in fluorosis etiology. With few contemporary studies of fluoride ingestion in this age group, the purpose of this paper is to describe patterns of estimated fluoride ingestion from birth to 36 months of age from water, dentifrice, and dietary fluoride supplements and combined.

Methods: Repeated responses to separate series of questions about water intake, use of fluoride dentifrice, and use of fluoride supplements were collected by questionnaire as part of the longitudinal Iowa Fluoride Study and used to estimate fluoride intake. Estimated intake is reported by source and combined at different ages. Effects of subject age and other covariates on fluoride intake were assessed using regression methods appropriate for the analysis of correlated data.

Results: For most children, water fluoride intake was the predominant source, especially through age 12 months. Combined daily fluoride intake increased through 9 months, was lower at 12 and 16 months, and increased again thereafter. Mean intake per unit body weight (bw) was about 0.075 mg F/kg bw through 3 months of age, 0.06 mg F/kg bw at 6 and 9 months, 0.035 mg F/kg bw at 12 and 16 months, and 0.043 mg F/kg bw from 20-36 months. Depending on the threshold chosen (e.g., 0.05 or 0.07 mg F/kg bw), variable percentages of the children exceeded the levels, with percentages greatest during the first 9 months. Regression analyses showed fluoride intake (mg F/kg bw) from 1.5-9 months to decrease with increasing child's age, mother's age, and mother's education, with a complex three-way interaction among these factors. From 12-20 months, fluoride intake increased with increasing child age and decreased with increasing mother's age. No statistically significant relationships were found for fluoride intake from 24-36 months.

Conclusions: There is considerable variation in fluoride intake across ages and among individuals. Longitudinal studies may be necessary to fully understand the relationships between fluoride ingestion over time and development of fluorosis.

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Keywords: Children, Dental fluorosis, Fluoride in food, Fluoride in water.
Source: J Public Health Dent 2001 Spring;61(2):70-7.

Fluid consumption related to climate among children in the United States

Objective: Recommended fluoride concentrations in US public water systems are between 0.7-1.2 ppm, depending on the mean daily maximum temperature. This range assumes that water intake is higher in warmer than in cooler climates, based on research from the 1950s. The aim of this analysis is to relate fluid consumption among American children aged 1-10 years to the local climate under modern conditions.

Methods: The quantities of daily total fluid intake per body weight (ml/kg) and plain water intake per body weight (ml/kg) of children were calculated from the 24-hour recall diet survey in the third National Health and Nutrition Examination Survey (NHANES III, 1988-94). The mean daily maximum temperature from 1961 to 1990, averaged for the month during which the NHANES III exam was conducted, was obtained for each survey location from the US Local Climate Historical Database. Multiple regression analysis was conducted using SAS and SUDAAN.

Results: Fluid intake was significantly associated with age, sex, socioeconomic status (SES), and race and ethnicity. No significant association could be found between the amount of either total fluid or plain water intake and mean daily maximum temperature, either before and after controlling for sex, age, SES, and race or ethnicity.

Conclusions: Results indicate that there is no evidence that fluid consumption among children is significantly related to mean temperature in modern conditions. This suggests that the national temperature-related guidelines for fluoride concentration in drinking water may be due for reevaluation.

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Keywords: Children, Fluid intake, Fluoride in water, Temperature, USA.

Source: J Public Health Dent 2001 Spring;61(2):99-106.

High concentrations of fluoride and boron in drinking water wells in the Muenster region – results of a preliminary investigation

In 1998, two cases of severe dental fluorosis in schoolchildren occurred in the Muenster region. These cases took place in one household, where fluoridated toothpaste, fluoridated salt, and fluoride tablets were consumed. Furthermore, the family used drinking water from its private well only. Analyses of the well water ordered by local health officials revealed very high amounts of fluoride, boron, and other electrolytes. This unusual combination

of high amounts of fluoride and boron could also be found in the water of a great number of other private wells that are the only source for drinking water in this rural region of the Muensterland. Anthropogenic sources could be excluded. Because of this, the results of the water samples were collated to the specific geological situation in this area. In the Muenster region there are marl layers of the chalk era covered with quarternary sediments. The quarternary sediments are up to 10 to 20 metres thick and they usually conduct the groundwater. The marl contains high concentrations of fluoride and boron. In some places the groundwater has contact with these layers. To check the amount of fluoride and boron in the groundwater, indicator values were sought, which can give a hint of high contents of these trace elements. In this study the conductivity and acidity were identified as possible indicators of a high amount of fluoride and boron in the drinking water in this specific region. To work economically and efficiently, the drinking water should be checked for fluoride and boron on a regular basis only when these values are extraordinarily high. In the case of high concentrations, especially of fluoride, in the drinking water the persons concerned should be informed about their potential health risk, giving them the opportunity to optimise the total daily intake of fluoride.

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Keywords: Dental fluorosis, Fluoride in water, Germany, Well water.

Source: *Int J Hyg Environ Health* 2001 Mar;203(3):221-4.

Prevention of brick tea fluorosis in rats with low-fluoride brick tea on laboratory observation

To test whether low-fluoride brick tea can prevent the occurrence of fluorosis, rats had access only to a specially prepared low-fluoride brick tea for 1 year. The daily fluoride intake, fluoride metabolism, tissue distribution and development of tooth fluorosis were observed at 4-monthly intervals, at the end of months 4, 8, and 12, respectively. Rats drinking ordinary brick tea (F-503.5 mg/kg) served as control. The daily intake of fluoride in the ordinary brick tea group was 0.3 mg, and this group developed dental fluorosis characterized as brown and white horizontal marks at the end of month 8, and white chalky dental fluorosis developed at the end of month 12. The total incidence was 75%. In contrast, the daily fluoride intake of the low-fluoride brick tea (F-210 mg/kg) group was 0.19 mg, and this group did not develop any signs of dental fluorosis.

Fluoride distribution was mainly retained in the bone tissue, and about half of the absorbed fluoride was excreted via urine and feces. The results

suggest that this low-fluoride brick tea did not induce fluorosis in rats and can be used as an effective control measure for humans.

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Keywords: Brick tea, Dental fluorosis, Rat study.

Source: Food Chem Toxicol 2001 Jun;39(6):615-9.

ENVIRONMENTAL FLUORIDE POLLUTION

Mechanism of degradation of the quality of natural water in the Lakes Region of the Ethiopian rift valley

The natural waters of the Lakes Region in the Main Ethiopian Rift (MER) show serious problems of chemical quality. The high content of fluoride reaches 300 mg/L and affects the health of the population who live in this area. Furthermore, the alkaline and sodic characteristics ($8 < \text{pH} < 10$ and $0.9 \times 10^{-4} < \text{Na} < 0.35 \text{ ml}^{-1}$) of these waters destroy the soil for agricultural use.

This geochemical study is conducted using analytical data issued from literature (UNDP, 1973; Chernet, 1982) and several field sampling and laboratory analyses completed during the last 4 yr. Chemical analyses of 320 samples taken throughout the region have been analysed using the computer software AQUA for geostatistic, statistic, chemical equilibria and simulation of evaporative concentration process. The results show that the waters issuing from volcanic rocks are characterised by a positive alkalinity residual of calcite. When they concentrate due to the effect of climate, the precipitation of calcite causes a decrease in the chemical activity of calcium. This results in an increase in solubility of fluoride, previously controlled by equilibrium with CaF_2 , and the element concentrates without being significantly affected by the precipitation of fluorite. As water concentrates, the low concentration of dissolved calcium emphasises the alkaline characteristics. As a consequence, the pH reaches very high values (9-10) which make the waters unsuitable for agriculture. The adverse elements, namely fluoride, sodium, and alkalinity accumulate in the lower zones of the basins. The acquisition of high fluoride content and the alkaline-sodic characteristics depends primarily on the unbalanced initial stage between the carbonate alkalinity and calcium [$(\text{HCO}_3^-) > 2(\text{Ca} + \text{Mg})$], which results from the weathering and dissolution of the volcanic rocks. The predictive qualities of the model AQUA were tested in the context of this region. A simulation of the concentration of the waters by evaporation has shown chemical variations relatively similar to those obtained with field data which include the minor chemical elements such as the fluoride or the dissolved silica. For field data, enrichment is more variable and may represent localised conditions of mineralization.

This model is then used to study a method of defluoridation by supplying these waters with calcium in the form of gypsum. This method also allows the reduction of the alkalizing and sodifying characteristics of the waters without reaching the fluoride standard concentration accepted for drinking waters. For drinking purpose, waters of the Ethiopian rift should undergo another method of defluoridation.

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Keywords: Defluoridation, Ethiopian rift, Fluoride in water.

Source: Water Res 2001 Aug;35(12):2819-32.

Inhalation exposure in secondary aluminium smelting

Inhalation exposure at seven UK secondary aluminium smelters was investigated to quantify the main exposures and identify their sources. The substances monitored were gases (carbon monoxide, hydrogen sulphide and nitrogen dioxide), total inhalable dust, metals, ammonia, polycyclic aromatic hydrocarbons (PAHs), particulate fluoride salts and acids. The results showed that people were exposed to a range of workplace air pollutants. Personal exposure results for total inhalable dust were between 700 and 5600 $\mu\text{g} \times \text{m}^{-3}$ and the maximum personal exposure result for particulate fluoride salts was 690 $\mu\text{g} \times \text{m}^{-3}$ (as F). The maximum aluminium, total PAH and lead personal exposure results were 900, 19 and 18 $\mu\text{g} \times \text{m}^{-3}$ respectively. The average proportion of aluminium in total inhalable dust samples was 13% and rotary furnace processes generated the most dust. Particulate fluoride salt exposure was more widespread than hydrofluoric acid exposure. The source of the salt exposure was fluoride containing fluxes. The lead exposure source was lead solder contamination in the furnace charge.

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Keywords: Aluminium smelter, Fluoride, Inhalable dust.

Source: Ann Occup Hyg 2001 Apr;45(3):217-25.

Health effects of fluoride pollution caused by coal burning

Recently a huge amount of fluoride in coal has been released into indoor environments by the combustion of coal and fluoride pollution seems to be increasing in some rural areas in China. Combustion of coal and coal bricks is the primary source of gaseous and aerosol fluoride and these forms of fluoride can easily enter exposed food products and the human respiratory tract. Major human fluoride exposure was caused by consumption of fluo-

ride contaminated food, such as corn, chilies and potatoes. For each diagnostic syndrome of dental fluorosis, a log-normal distribution was observed on the logarithm of urinary fluoride concentration in students in China. Urinary fluoride content was found to be a primary health indicator of the prevalence of dental fluorosis in the community. In the fluorosis areas, osteosclerosis in skeletal fluorosis patients was observed with a high prevalence. A biochemical marker of bone resorption, urinary deoxypyridinoline content was much higher in residents in China than in residents in Japan. It was suggested that bone resorption was stimulated to a greater extent in residents in China and fluoride may stimulate both bone resorption and bone formation. Renal function especially glomerular filtration rate was very sensitive to fluoride exposure. Inorganic phosphate concentrations in urine were significantly lower in the residents in fluorosis areas in China than in non-fluorosis area in China and Japan. Since airborne fluoride from the combustion of coal pollutes extensively both the living environment and food, it is necessary to reduce fluoride pollution caused by coal burning.

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Keywords: Coal burning, China, Dietary fluoride, Fluoride pollution.

Source: *Sci Total Environ* 2001 Apr 23;271(1-3):107-16.

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ENVIRONMENTAL FLUORIDE AND ANALYTICAL ASPECTS

RESEARCH REPORTS

**Fluoride and arsenic concentrations in
groundwater of Aguascalientes, Mexico**

The aim was to quantify simultaneously the fluoride and arsenic content in water, used for human consumption, from wells supplying Aguascalientes City, Mexico. We developed a sampling program that involved 126 wells for fluoride evaluation. From this group of wells, 39 were selected for arsenic evaluation. The minimum and maximum arsenic concentration were 0.0019 mg/L and 0.0326 mg/L respectively. The mean arsenic concentration was 0.0145 ± 0.0077 mg/L. The fluoride concentration ranged from 0.53 mg/L to 11.31 mg/L. The mean fluoride concentration was 1.82 ± 1.42 mg/L. These results and the geographic coordinates of the wells were used to develop the isopleth map to show the fluoride and arsenic distribution in Aguascalientes City. We estimated that a resident of this city could ingest 0.233 mg/kg body weight/day of fluoride and 0.678 μ g/kg body weight/day of arsenic.

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Keywords: Aguascalientes, Mexico, Arsenic in water, Fluoride in water, Isopleth mapping.

Estimation of mean daily fluoride intake in Japan

The aim was to estimate the individual daily fluoride intakes (DFIs) from commercially available food samples. Fluoride was determined by hexamethyldisiloxane (HMDS) microdiffusion method at 60°C for 12 hr. Fluoride concentrations found were: (a) infant formula (ready to feed) prepared with 0.1 ppm F drinking water: 0.14-0.22 μ g/mL, mean 0.17 μ g/mL, n=10; (b) baby foods: cereal mean 0.30 μ g/mL, n=11, meat and fish mean 0.13 μ g/mL, n=20, vegetables mean 0.17 μ g/mL, n=11, juice mean 0.10 μ g/mL, n=6; (c) daily foods: milk mean 0.05 μ g/g, egg mean 0.08 μ g/g, meat and

fish mean 0.023 µg/g, vegetables mean 0.06 µg/g, potatoes and starches mean 0.02 µg/g, fruits mean 0.07 µg/g, cereals mean 0.14 µg/g, sugars mean 0.07 µg/g; (d) beverages: mean 0.46 µg/g; (e) drinking water in a non-fluoridated area in Japan: 0.1 mg/L. The estimated mean DFIs for infants, 3-8 months of age, ranged from 0.166 to 0.266 mg/day and mean DFIs in 1-6 year olds were 0.289-0.321 mg/day. DFIs per body weight ranged from 0.023-0.029 mg/kg in infants aged 3-8 months and from 0.027 to 0.11 mg/kg in 1-6 year olds compared to 0.05-0.07 mg/kg which is the temporary criterion for the occurrence of dental fluorosis (Food and Nutrition Board, Institute of Medicine, National Academy of Sciences, USA).

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Keywords: Fluoride in food, Fluoride intake, Fluoride in water, Japan.

Fluoride mobility in aluminium and calcium loaded soils

The effects of aluminium and calcium on fluoride mobility in soil were studied. Loading a laboratory soil column with calcium caused a linear decrease in fluoride mobility. Aluminium increased fluoride mobility initially, with fixation occurring after 15 days.

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Keywords: Aluminium, Calcium, Fluoride mobility in soil, Soil fluoride.

Preparation of an adsorbent for fluoride removal from drinking water

In this work, we obtained an activated boehmite which maximizes the fluoride removal from water. This new material can be prepared using a process that consists of the solid sphere agglomeration of the product obtained from the partial hydrolytic reaction of aluminium sulfate in an ammonia medium. After a contact time of 30 minutes, the new adsorbent removed 67.8% of fluorides present in 100 mL of a solution with a concentration of 2 mg/L. This activated boehmite provides an adsorption capacity of 5.0 g F/kg Al₂O₃. The boehmite was compared with two commercial adsorbents: bone charcoal and *Keftel* alumina. The removal rate of our adsorbent was about 100% higher than those obtained with the other commercial adsorbents under the same conditions. The performance and behavior of the boehmite was also studied and compared using groundwater samples with a 4 mg/L fluoride concentration. In that part of the study, we used different combinations between adsorbent amount and contact time. It was observed that the pres-

ence of another chemical species in groundwater samples caused a decrease of about 45% in the removal capacity of the activated boehmite.

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Keywords: Boehmite, Water defluoridation.

Effect of fluoride contamination of drinking water on human spermatozoa

The aim was to assess the effect of fluoride contaminated drinking water on the morphology and ultrastructure of spermatozoa of infertile patients. Outpatients at a New Delhi infertility clinic, aged 20-42 years and consuming water with 2.0-19.0 ppm of fluoride, were compared to controls with a drinking water fluoride level of <1.0 ppm. A significant increase in the percentage of abnormal forms of spermatozoa were observed in the study group as compared to the controls. The study group showed structural abnormalities in the sperm head, midpiece and tail, altered axonemal arrangement, hypoplasia of acrosome, swollen mitochondria with obliteration of cristae, and dilated rough endoplasmic reticulum. The extensive structural aberrations in the spermatozoa of the patients consuming the fluoride-contaminated water were seen to be causing the impaired fertility.

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Keywords: Fertility reduction, Fluoride in water, Spermatozoa.

Defluoridation of water using amended clays

The fluoride sorption properties of two different soils, a sand fraction with a particle size of 0.05-2 mm and a silty clay fraction with a particle size of <0.05 mm, were investigated as a function of pH (4-9) and shaking time (60-360 min). The silty clay was found to be a potent fluoride sorbing agent which could be used for the defluoridation of water. The silty clay was further investigated after being amended by the addition of activated alumina, ferric chloride, and calcium carbonate with Al, Fe, and Ca cation concentrations of 50-200 mg/g, pH 4-10, initial fluoride concentration 2.5-25 mg/L, shaking time 5-180 min, and a clay concentration of 5-100 g/L. The amended clay showed improved fluoride sorption properties with alumina being most effective, calcium carbonate least effective, and ferric chloride intermedi-

ately effective. It was most effective at pH 6, when amended by the addition of activated alumina, ferric chloride and calcium carbonate, at a dose for each of 50 mg/g. Shaking for 5 min resulted in a considerable amount of fluoride being removed. Some defluoridation, from stored water with 150 ppm of fluoride, also occurred with earthen pots made from silty clay.

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Keywords: Clay, Defluoridation.

Measurement of bone calcium, phosphorus, and fluoride in growing rats by X-ray fluorescence analysis

The aim was to study bone mineral density in growing rats. Seventy-five 4-week-old SPF rats (Crj.CD (SD) IGS, Charles River, Japan) were fed a normal diet (Oriental Yeast Co. NMF). The mandibles, femurs and lumbar spines were extracted at 4, 6 and 8 weeks. After soaking in 10% formalin solution for one week and drying naturally, they were pulverized for 60 s, using a TI-100N pulverizer and the resulting powder was shaped in a 10 mm or 20 mm ring under pressure. The bones of 5 rats from each group were formed into a sample to allow analysis, for each part and age, of the levels of bone calcium, phosphorus, and fluoride by X-ray fluorescence (XRF) using XRF ZXF 101e (Rigaku Industry).

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Keywords: Bone density, Bone fluoride content, Calcium, Growing rats, Phosphorus.

Statistical neural network analysis of fluoride bioaccumulation in deer bones from Polish industrial areas

The neural networks method of statistical analysis was applied to data on fluoride intoxication of wild animals. The bone fluoride was measured in the lower jaws of 118 deer shot by hunters during 1996-7 in the area of Western Pomerania in Poland where the main sources of fluoride emission occur from mineral fertilizer production (Police Chemical Plant) and a coal fired power station (Dolna Odra Power Plant). To describe the typical random curve resulting from the analysis a neural network was used. The determined variable was the content of fluoride in the bone. As parameters which describe this variable we used: the 12 regions, their distance from the emission source in km, location of the regions relative to the two emission sources with consideration also of the main wind directions (angle one for Police

Chemical Plant, angle two for Dolna Odra Power Plant), age of the animals, and the individual traits of the animals. In the study we used Multilayer Perceptron (MLP), Radical Basis Function (RBF), and Generalized Regression Neural Networks (GRNN). The quality of the networks was assessed by using calculated indices. In order of significance the relevant parameters were: the age of the animals which gives a measure of the duration of exposure; the region; the distance and direction of the emission source; and the animal's features. In this manner the factors which influenced the accumulation of fluoride in the bones were determined, and the process described quantitatively. This method can also indicate possible industries which pollute the environment in a given area. The application of the described computing technique makes it possible to increase the number of parameters put into the computer which are important in the processes of intoxication in various environmental conditions.

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Keywords: Deer bone fluoride, Environmental fluoride, Industrial fluoride pollution, Poland, Statistical analysis.

British dietary fluoride intake

Fluoride levels were measured in some dietary items with a selective fluoride ion electrode. Fluoride sources included tea, fish, and red meat with chicken meat having a lower content. The fluoride content of Indian tea reflected the source of the tea leaves rather than the water used to make the tea which had a fluoride level of 0.15 ppm in the Thames Metropolitan Area of London.

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Keywords: Fluoride in food, Fluoride in tea.

Water quality and fluoride levels in Lake Biwa of Japan

Lake Biwa is the biggest lake in Japan with a surface area of 675 km² and a water reserve of 237 500 million tons. The fluoride content of the water in Lake Biwa and the rivers flowing into it is 0.01 ppm and the quality of the fresh water is excellent, especially in the northern part of the lake. Three quarters of the population living in the Kyoto, Osaka, and the Kobe-megalopolis, approximately 13 million residents, depend on water from Lake Biwa. Water with a higher fluoride level of up to 3 ppm, in the Ogoto

Spring on the western shore of Lake Biwa and at Arima and Takarazuka in the Hyogo Prefecture, is not used now in the water supply.

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Keywords: Fluoride in water, Japan, Lake Biwa.

RESEARCH REVIEWS

Regression analysis of cancer incidence rates and water fluoridation in the U.S.A.

1. The methodology and results

Our aim was to analyze the association between cancer incidence rates and fluoridated drinking water (FD) in the U.S.A. using WHO data from 1978-92. Age-specific and age-standardized rates of registered cancers for nine communities in the U.S.A. (21.8 million inhabitants, mainly white) were obtained from IARC data (1978-82, 1983-87, 1988-92). About two thirds of the cancer sites of the body were associated positively with FD, but negative associations were noted for melanoma of the skin, and cancers of the lip, prostate and thyroid gland. A limited association was present for the stomach and none for the small intestine. Cancers of the oral cavity and pharynx, colon and rectum, hepato-biliary and urinary organs, and bone cancer in males were positively associated with FD. Of the 36 sites, 23 were positively significant (63.9%), 9 were not significant (25.0%), and 4 were negatively significant (11.1%). Such a broad spectrum of association has never been observed for any particular known carcinogen but may be reasonable for fluoride given its strong electronegative nature.

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Keywords: Cancer incidence, Cancer in the U.S.A., Epidemiology, Fluoride in water.

Editorial note: this material is also reported in another abstract in this issue, Fluoride 2001;34:184.

Regression analysis of cancer incidence rates and water fluoridation in the U.S.A.

2. Cancer of the oral cavity and pharynx

Cancer occurs to a significant extent in the upper digestive tract, including the oral cavity, pharynx and esophagus, and in the lower part of the tract, including the colon and rectum, with the stomach and small intestine being relatively spared. The total incidence rates for the upper digestive tract are

similar to those for the rectum. In the Surveillance, Epidemiology, and End Results (SEER) study (Hoover, 1991) the Observed/Expected (O/E) cancer rates for the upper digestive tract with exposure to fluoridation gave a non-significant confidence interval (95% CI) when five years were analysed individually but was significant when the figures for five years were analysed together (95% CI 1.25-1.83). In the National Toxicology Programme (NTP, 1990) study on rats the rate of cancer in the upper digestive tract for the group receiving the highest concentration of sodium fluoride was not significantly increased compared to the control group. However, our analysis revealed a significant dose-response regression relationship using all the available data and the total daily intake (TDI) (page 69 in the NTP report, $P < 0.03$). In addition the occurrence of neoplasia in the control groups was not seen to be a reasonable ground for disregarding the effect of sodium fluoride in causing cancer as the control groups also received fluoride at a level of 0.2 mg F/kg body weight/day (page 69). The stated absence of focal hyperplasia of the oral mucosa prior to the cancer developing was negated by information found by JA Yiamouyiannis who consulted the original record issued by the Battelle Memorial Institute (1989). A high level of fluoride, 30 ppm has been found in plaque after the use of drinking water containing 1 ppm (Gabler, 1981). Increased plaque fluoride levels having also been found with using fluoride dentifrice (plaque F 30 ppm), fluoride mouth rinse (100 ppm), and fluoride gel (200 ppm) (Zero, 1992). Zero found the plaque fluoride level decreased by 10 ppm after 24 hr. For cultured human diploid cells the minimum clastogenic level for fluoride is 10 ppm (Oguro, 1995) with cell division being inhibited at this level and the cells destroyed at 20 ppm. In conclusion, the data from the various surveys and laboratory investigations support water fluoridation as being causal for cancer in the oral cavity and pharynx.

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Keywords: Cancer incidence, Cancer in the U.S.A., Epidemiology, Fluoride in water, Gastrointestinal tract cancer, Oral cancer.

Regression analysis of cancer incidence rates and water fluoridation in the U.S.A.

3. An approach to the causal genesis

The likelihood of fluoride being a causal factor for cancer was estimated by comparing our epidemiological results with the five criteria proposed in Smoking and Lung Cancer (U.S.A. 1964). The criteria involved the consistency, strength, specificity, temporal relationship, and coherence of the association. The consistency of the association was seen to be confirmed only in

the U.S.A. with data from elsewhere not being available. Although the concentration of fluoride in water was only moderately rather than extremely high, it was seen that the strength was sufficient to cause a wide distribution of cancers with a total excess cancer incidence rate of 20%. The specificity of the association was seen to be consistent with the pathophysiology of fluoride, e.g. the accumulation of fluoride in plaque would lead to higher levels in the oral cavity and pharynx and thus increased cancers in these areas. Data were not available to examine the temporal relationship. The coherence of the association was seen to be supported by the literature. In conclusion fluoride was seen to be a causal factor in human cancer.

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Keywords: Cancer incidence, Cancer in the U.S.A., Epidemiology, Fluoride in water.

Water fluoridation in China

In response to expert dental advice that fluoridation would lower the incidence of caries by 50%, the news media reported in 1999 that China would completely implement fluoridation in the year 2000. In response to this, my colleagues and I published A Report on Fluoridation and Caries Prevention, in August 2000, to provide accurate information. The book covered the history of fluoridation, the points in the debate, the harmfulness of excessive fluoride, the health standards for fluoride in water and total fluoride intake, the etiology and epidemiology of caries, and comprehensive measures for caries prevention. Its influence has played an important role in stopping the blind trend towards universal fluoridation in China. In 1976, a group of Guizhou's fluoride researchers and I found non-water source fluorosis in an area where the concentration of fluoride in drinking water was only 0.18 ppm. This leads to understanding the importance of knowing the total fluoride intake from all sources and the danger of adding fluoride to drinking water to prevent caries according to a "standard". This concept has contributed to the ending, after 18 years, of fluoridation in Guangzhou, China. Our research showed that the maximum fluoride concentration in drinking water should be 0.6 ppm, and that 1.0 ppm was too high. The total fluoride intake standard of China is less than 2.4 mg/day for those under 15 years old, and less than 3.5 mg/day for those aged 15 or more years.

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Keywords: China, Fluoride in food, Total fluoride intake, Water fluoridation.

BIOLOGICAL EFFECTS OF FLUORIDE

RESEARCH REPORTS

Perisinusoidal (Ito) cells in hepatocellular carcinoma treated with fluorouracyl

A primary hepatocellular carcinoma was diagnosed, by open needle liver biopsy in a 49 year old female with a history of consuming a large quantity of carrot juice daily for several years. An unusually marked proliferation of perisinusoidal (Ito) cells was present. This apparently resulted from an excessive vitamin A intake, leading to enhanced myofibroblast formation, interstitial fibrosis, hepatocellular necrobiosis and hepatic cell proliferation. There was an unusually rapid progression of the multifocal, but histologically low-grade hepatocellular carcinoma. Marked clinical improvement with a slower progression of tumour growth followed fluorouracyl chemotherapy combined with local hyperthermic treatment and cessation of excessive intake of carotene and retinol.

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Keywords: Fluorouracyl, Ito cells, Liver cancer, Perisinusoidal cells, Vitamin A.

Skeletal radiographs, hydroxyproline, alkaline phosphatase, and other biochemical parameters in endemic fluorosis

In order to investigate the relationship in endemic fluorosis between skeletal radiographs, urinary hydroxyproline (HOP) and creatinine (Cr), and serum alkaline phosphatase (AKP), 49 patients with dental fluorosis and 15 controls were compared. X-ray examinations were made of the pelvis, and the right forearm and leg. Radiological diagnoses of skeletal fluorosis were made using The Control Work Standard of Endemic Fluorosis (Chinese Endemic Disease Office, 1981). The controls were from a nonendemic disease region in Guiyang. Ten (20.4%) of the 49 patients with dental fluorosis had normal skeletal radiographs and 39 (79.6%) had skeletal fluorosis: osteoporosis type 10 (mild 5, moderate 5); osteosclerosis type 22 (mild 10, moderate 12); mixed type 7 (severe 7). In the mixed type osteoporosis, osteosclerosis, osteomalacia and changes indicating increased bone turnover were present. Urinary HOP was higher in the fluorosis group compared to the controls, increasing with severity. It was lowest in the dental fluorosis group, highest in the mixed type and at a similar level in the osteoporosis and osteosclerosis groups. Serum AKP was increased, compared to the controls, dental fluorosis group, in the osteosclerosis and mixed (osteoporosis and osteosclerosis)

types of fluorosis. Urinary HOP may be used as an early indicator of endemic fluorosis. Elevated levels indicate that increased collagen breakdown is occurring in bone. Serum AKP is increased with the proliferation of osteoblasts in the osteosclerosis and mixed types of skeletal fluorosis. Fluoride increases bone turnover with increased bone destruction and formation.

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Keywords: Alkaline phosphatase, Collagen destruction, Hydroxyproline, Radiographs, Skeletal fluorosis.

Physicochemical properties of functionally graded fluoridated hydroxyapatite

In human teeth there is a gradient of fluoride concentration from the surface of the tooth enamel to the dentine-enamel junction. We speculated that even each crystal composing the enamel layer may have a fluoride distribution in the crystal structure and designed a continuous gradient fluoride system by developing the previous step-like fluoride supply system. Fluoridated hydroxyapatite was synthesized at $80\pm 1^\circ\text{C}$ and $\text{pH } 7.4\pm 0.2$ using a gradient fluoride supply system. Briefly, 0.5 L of 100 mmol/L $\text{Ca}(\text{CH}_3\text{COO})_2\cdot\text{H}_2\text{O}$ solution and 0.5 L of 60 mmol/L $\text{NH}_4\text{H}_2\text{PO}_4$ solution (tank II), into which 0.5 L of 60 mmol/L $\text{NH}_4\text{H}_2\text{PO}_4$ solution containing a constant concentration of 31.8 mol/L HF (tank I) was continuously supplied, were fed into mechanically stirred 1.3 mol/L acetate buffer solution. X-ray diffraction analysis showed a typically apatitic pattern, although the (300) reflection was broader than that of homogeneous fluoroapatite. Scanning electron micrographic observation indicated that the apatite was composed of rod-like crystals similarly to fluoroapatite. High resolution transmission electron microscopy showed electron damage in the core of the crystal. When the apatite pellet was prepared, electron spectroscopy for chemical analysis showed a negative gradient of fluoride concentration with depth in the crystals. The apparent solubility in 0.5 mol/L acetate buffer solution (37°C and $\text{pH } 4.0$) was 9.16 ± 0.39 mmol/L, much less than that of homogeneous hydroxyapatite 32.3 ± 1.9 mmol/L, and less than that of heterogeneous two-layer fluoridated apatite with an outer fluoride-rich layer 12.5 ± 0.6 mmol/L, which was synthesized previously by supplying fluoride during the latter half of the experimental period. These results suggest that graded fluoridated apatite may be formed by this process and have higher acid resistance than two-layer fluoridated apatite.

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Keywords: Acid resistance, Fluoride in enamel, Graded fluoroapatite.

Effect of fluoride on cytokine expression in murine splenic macrophages during subacute oral exposure

The aim was to study the effect of fluoride on the mRNA expression of cytokines in splenic macrophages from male BALB/c mice exposed to fluoride in their drinking water for one month at the levels of 0, 1, 5, 25, and 125 ppm of fluoride which was supplied as sodium fluoride. After euthanasia, splenocytes from each sample were divided into adherent cells (macrophages) and non-adherent cells (lymphocytes) by incubation in a cell culture plate. Adherent cells were activated by lipopolysaccharide (LPS) and cultured. The mRNA expressions of tumor necrosis factor (TNF) α and interleukin(IL)-1 β in splenic macrophages were examined by RT-PCR. The expression of β -actin of each sample was used as an internal standard. Each amplified product was qualified as a band separated by electrophoresis on agarose gel containing ethidium bromide. The relative expression of TNF α in macrophages was lower among the group treated with 125 ppm fluoride, whereas there were no effects on the relative expression of IL-1 β . The results suggested that the inhibitory effect of fluoride on TNF α production in splenic macrophages is more sensitive compared to IL-1 β *in vivo*. TNF α may therefore play an important role in the mechanism of action of fluoride *in vivo*.

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Keywords: Cytokine, Fluoride, Interleukin-1B, Male mice, Splenic macrophages, Tumor necrosis factors.

The influence of fluoride on the calcification induced in a co-culture of mouse osteoblasts and nacre of mollusk *Hyriopsis schlegeli*

The debate about the role of fluoride on inhibiting or facilitating bone calcification still persists. In recent studies nacre, mother-of-pearl from any shelled mollusc, was reported to be able to induce osteoblasts to form calcified tissue *in vitro*. We used nacre from the mollusc *Hyriopsis schlegeli* to test the influence of fluoride on calcification *in vitro*. In four groups of 35 mm plastic dishes, we co-cultured mouse osteoblasts and undecalcified nacreous chips (300-425 μ m) in a medium with fluoride at concentrations of 0 ppm (n=12), 1 ppm (n=13), 10 ppm (n=11), and 100 ppm (n=5). Only one nacreous chip was placed in each dish. The nutrient medium was an alpha modification of Eagle's medium containing also 1% glutamine and 10% fetal bovine serum. These four groups were cultured for four weeks and then

stained with alizarin red S, which stains calcified tissue red. The area of nacreous chips and red-stained tissue was recorded with a digital camera and calculated with a computer program Scion Image. The results showed that the addition of 100 ppm of fluoride was toxic to the mouse osteoblasts. The red-stained areas produced by the osteoblasts were $25.45 \pm 10.46 \mu\text{m}^2$ (0 ppm), $22.50 \pm 11.68 \mu\text{m}^2$ (1 ppm), and $26.06 \pm 12.15 \mu\text{m}^2$ (10 ppm). The ratios of increased red-stained area to the area of nacreous chips were 0.26 ± 0.11 (0 ppm), 0.21 ± 0.07 (1 ppm), and 0.22 ± 0.12 (10 ppm). We conclude that a concentration of fluoride up to 10 ppm is not cytotoxic and has no obvious influence on the process of bone calcification under these conditions.

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Keywords: Bone calcification, Fluoride, Mollusc nacre, Mouse osteoblasts.

Fluoride-induced apoptosis in human bone marrow and cord blood CD34⁺ cells

The aim was to study the induction, by sodium fluoride (NaF), of apoptosis in human bone marrow (BM) and cord blood (CB) CD34⁺ cells. Two different assays were used to evaluate the executive phase of apoptosis. BM and CB CD34⁺ cells were exposed to different doses of NaF (0, 1, 10, and 50 mg/L) and stored at 37°C for 24 hours. The following day the percentage of apoptotic cells was assessed. Apoptotic and necrotic cells were detected after Annexin-V and propidium iodide staining respectively and evaluated by a Flow Activated Cell Sorter (FACS). The cell culture supernatants were evaluated using enzyme-linked immunosorbent assay for quantitative detection of human copper zinc superoxide dismutase (Cu/ZnSOD ELISA). Apoptosis was detectable both in CB and BM hematopoietic progenitor cells exposed to different doses of NaF. High doses of NaF caused an increase in the executive phase of the apoptotic process, especially at the concentration of 50 mg/L measured by FACS. For CB the differences were more significant. Both BM and CB CD34⁺ cells exposed to the high dose of NaF (50 mg/L) secreted significantly more Cu/ZnSOD to the medium when compared to equivalent numbers of cells exposed to lower doses of NaF or unexposed cells. We conclude that exposure to sodium fluoride could potentially damage human cells involved in hematopoiesis.

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Keywords: Apoptosis, Fluoride toxicity, Hematopoiesis, Human bone marrow.

Antidotes for fluoride and arsenic-induced kidney toxicity in mice

The aim was to study the effects on the renal effects in mice (*Mus musculus*) of the administration, for 30 days, of sodium fluoride (NaF), 5 mg/kg body weight, and arsenic trioxide (As_2O_3), 0.5 mg/kg body weight, and the potentially beneficial effects of calcium phosphate, vitamins C and E. The histology and protein, creatinine, alkaline and acid phosphatases (ALP, ACP) were studied using standard protocols. The doses of NaF and As_2O_3 were based on LD_{50} values while those of the antidotes were based on previous work. The administration of NaF and As_2O_3 , singly and in combination, resulted in decreased renal total protein, ACP and ALP. This was seen to affect enzymes, structural proteins, membrane permeability and cellular functions. The alterations correlated with the severe histological changes in the kidney including vacuolization, distortion of tubules and haemorrhagic regions. The tubules had necrotic epithelium, highly pyknotic nuclei and obliteration of the lumen. The glomeruli were shrunken with pyknosis, the presence of vacuoles and nearby leucocyte infiltration.

After the withdrawal of the NaF and As_2O_3 , a degree of recovery occurred in the parameters apart from the histology. The administration of vitamin C, calcium phosphate and vitamin E, alone or in combination, during the withdrawal period, resulted in almost complete recovery, which was more pronounced with the combination of antidotes. Vitamin C and vitamin E are potent antidotes and function as therapeutic agents in several disease states especially those involved in oxidative related events. Calcium is important for cellular metabolism. In conclusion combined treatment with vitamins C and E, and calcium phosphate was effective in treating the fluoride and arsenic induced nephrotoxicity.

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Keywords: Arsenic poisoning, Ascorbic acid, Calcium, Fluoride toxicity, Mice, Nephrotoxicity, Toxicity antidotes, Vitamin E.

Antidotes for fluoride and arsenic-induced testicular and ovarian toxicity in mice

This work was designed to study the effects of vitamins E and C, and calcium on the recovery in testicular and ovarian steroidogenesis in mice after the administration, for 30 days, of sodium fluoride (NaF), 5 mg/kg body weight, and arsenic trioxide (As_2O_3), 0.5 mg/kg body weight. A significant decrease occurred in the protein levels and in the activities of 3β HSD and

17 β HSD in both gonads of the mice given NaF and As₂O₃ compared to the control mice. These data and the associated histological changes suggest that the NaF and As₂O₃ affected steroidogenesis in the testis and ovary of mice. Withdrawal of NaF and As₂O₃ produced incomplete recovery. Supplementation with vitamins C or E, or calcium during the withdrawal period of the treated mice increased recovery from the induced effects. Vitamin C treatment was more effective than vitamin E or calcium. The recovery might be due to the powerful reducing action of ascorbic acid which also activates several enzymes and together with calcium inhibits phosphodiesterase (PDE), whereby the level of C-AMP is increased leading to enhanced enzyme activity and cellular metabolism. Vitamin E is a potent antioxidant which exerts its effect through destruction of free radical oxygen species. The radical scavenging activity of α -tocopherol is enhanced in the presence of ascorbic acid. The almost complete recovery caused by combined treatment with the antidotes might be due to their synergistic action. The effects of fluoride and arsenic may be reversed by their withdrawal and giving antidotes. The findings are relevant to humans exposed to high fluoride and arsenic levels.

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Keywords: Arsenic poisoning, Ascorbic acid, Calcium, Fluoride toxicity, Ovarian steroidogenesis, Testicular steroidogenesis, Toxicity antidotes, Vitamin E.

Genotoxicity of sodium fluoride and aluminium chloride on human lymphocytes

We conducted a micronuclei assay and aneuploidy scoring after the addition of sodium fluoride (NaF) and aluminium chloride (AlCl₃) to peripheral blood cell cultures of normal healthy volunteers. Doses of 20 μ g NaF and AlCl₃ respectively were added to 7 ml of the culture medium. A significant increase was observed in the frequencies of binucleates with micronuclei (MN) and the total number of MN in the fluoride and aluminium treated cultures. In some individuals a single binucleate had more than one micronucleus in the treated groups, suggesting multiple chromosome or chromatid elimination, which could be attributed to multiple chromosome or chromatid lagging at anaphase due to non-disjunction. A higher frequency ($P < 0.01$) of aneuploidy was observed after fluoride and aluminium treatment which might have resulted from microtubular malfunctioning. Increased MN in the present study might be the result of increased non-disjunction and lagging at anaphase. This may cause aneuploidy and increase the incidence of abnormality in the population being exposed to these chemicals. The data reveal

that populations in endemic areas exposed to fluoride and/or aluminium might be at risk of developing genotoxic effects.

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Keywords: Aluminium toxicity, Fluoride toxicity, Genotoxicity, Human lymphocytes.

HEALTH EFFECTS ON HUMANS AND ANIMALS

RESEARCH REPORTS

Expression of skin $\alpha 1(I)$ collagen gene in rats exposed to sodium fluoride in drinking water

The aim was to investigate the expression of the skin $\alpha 1(I)$ collagen gene in rats exposed to NaF in drinking water. One hundred 6-week-old Wistar rats (males and females) were divided into 4 groups. Animals in group 1 (controls) were given ordinary drinking water while those in groups 2, 3, and 4 were given 60 ppm NaF in drinking water for 1, 3, and 6 months respectively. At the end of the experiment the animals were sacrificed and fragments of the abdominal skin were taken for analysis. The total RNA in the skin samples was isolated according to the modified method of Chomczynski with guanidine thiocyanate. Semi-quantitative analysis of $\alpha 1(I)$ collagen gene expression was performed using RT-PCR assay by comparing amounts of $\alpha 1(I)$ collagen gene mRNA with the house-keeping gene mRNA encoding L19 ribosomal protein. No statistically significant differences in the expression of skin $\alpha 1(I)$ collagen gene between the examined and control groups were found. The expression of the skin $\alpha 1(I)$ collagen gene was not affected by sodium fluoride at the doses used.

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Keywords: Fluoride in drinking water, Rat study, Skin $\alpha 1(I)$ collagen gene.

Effect of low formula weight antioxidants on fluoride toxicity and fluoride excretion

The aim was to study the effects of small molecule antioxidants on fluoride toxicity and the basis of the theories and techniques of quenching free radicals in order to develop antioxidants for fluorosis treatment. Fluoride toxicity was produced in Wistar rats by adding it to the drinking water. Glutathione (GSH) was given by gastric instillation, and Se, vitamin C, vi-

tamin E, β -carotene, and superoxide dismutase (SOD) by drinking. The determinations consisted of the contents of LPO (MDA), $\text{NO}_2^-/\text{NO}_3^-$, GSH and $-\text{SH}$ as well as the activities of SOD and GSH-Px. The urinary and faecal fluoride were also analysed. In the simple fluoride-treated group the contents of MDA in the serum and the various tissues of the rats increased. $\text{NO}_2^-/\text{NO}_3^-$ decreased in the kidney and the brain but increased in the heart and liver. The $-\text{SH}$ and GSH levels in the blood and the various organs decreased significantly while the activities of GSH-Px and SOD decreased dramatically. After taking the antioxidants orally the MDA the rise in MDA was considerably inhibited, the abnormal synthesis of NO was antagonized, and GSH, $-\text{SH}$, SOD, and GSH-Px recovered in different degrees. Moreover, antioxidants can accelerate the excretion of urinary F and reduce the blood fluoride. When these antioxidants were given in combination the changes in the indexes were more obvious. Urinary and faecal fluoride levels increased with the various combinations of the antioxidants.

In summary we find that active oxygen and free radical damage play an important role in the occurrence of fluoride toxicity. Both laboratory and epidemiological investigations suggest that fluoride can lead to oxidative damage. To some extent the small molecule antioxidants mentioned above can inhibit these fluoride-induced oxidative damages. Moreover, they can accelerate the excretion of fluoride. The antioxidants work in cooperation if utilized in various combinations, especially with vitamin C, vitamin E, Se and GSH.

The research was supported by the National Fund of the Health Department of China.

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Keywords: Antioxidants, β -carotene, Fluoride excretion, Fluoride toxicity, Glutathione, Vitamin C, Vitamin E, Selenium, Superoxide dismutase.

Increased water fluoride and physical development in children

The aim was to examine the bone, teeth, and physical development of children, aged 10-15 years (n=103, 48 boys, 55 girls), living in Moldavia (Korneshti, Kalarash, Faleshti) in areas with a high level of fluoride in the drinking water. The fluoride levels in Kalarash and Faleshti were increased 2-5 fold with the maximum level being 3.5 ppm in Faleshti. The daily food ration of teenagers from all the towns was characterized by a lower calorie content, insufficient protein consumption, a low level of micronutrients, and an imbalance in the consumption of carbohydrates and fats. The excess fluoride in the water produced dental fluorosis, the frequency and degree of which depended on the fluoride level in the drinking water. The physical

development of the children was delayed in the boys and unbalanced in the girls. The raised water fluoride content adversely affected the peak bone mass, physical development, and the teeth. Measures to prevent osteoporosis and fluorosis resulting from the high drinking water fluoride are required.

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Keywords: Bone mass peaking, Dental fluorosis, Fluoride in water, Physical development, Protein deficiency, Ukraine.

Interaction of fluoride ions with fatty acids present in atheromatous plaques from carotid arteries

The purpose of this study was to examine possible interactions between fluoride ions, ionic strength, and fatty acids in human atheromatous plaque. For studying the effects of fluoride ions plaques were incubated with 0, 0.025, 0.25 and 2.5 mM NaF and for studying the effect of ionic strength with 0 and increasing concentrations of NaCl. Fatty acids were extracted according to Folch, methylated with 14% BF₃ in CH₃OH for 20 minutes at 70°C and separated on a gas chromatographic column. Fatty acids in the atheromatous plaque were degraded in the presence of fluoride ions to shorter ones. The accumulation of fluoride in plaque appears to alter its physicochemical properties, e.g. resistance to blood pressure and shear forces, facilitating the formation of emboli.

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Keywords: Atheromatous plaque, Carotid artery, Fatty acid composition, Fluoride in plaque.

Arsenic and fluoride-induced toxicity in gastrocnemius muscle of mice and its reversal by therapeutic agents

The effects on muscle physiology of sodium fluoride (NaF) and arsenic trioxide (As₂O₃) were investigated by administering it, either individually or in combination, to adult mice (*Mus musculus*) of the Swiss strain. The reversal of toxicity was also investigated by the withdrawal of the treatment and the use of the antidotes ascorbic acid, calcium, and vitamin E. All the biochemical parameters were assayed in the gastrocnemius muscle using standard procedures in both the control and the treated mice. The doses of NaF and As₂O₃ (5 and 0.5 mg/kg body weight) were based on the LD₅₀ value

while those of the antidotes were drawn from earlier work. The data revealed alterations in muscle metabolism after treatment, with a decline in the total protein levels which would affect the contractile pattern. The significantly enhanced levels of glycogen with a concomitant decrease in phosphorylase activity indicates impaired carbohydrate metabolism and thus in the activity of the quick contracting white fibres. The decrease in ATPase and succinate dehydrogenase activities suggested that the oxidative and energy metabolisms were disturbed, probably due to structural and functional changes in muscle. The depletion in cholinesterase activity was seen to lead to improper synaptic transmission and thus to affect muscle contraction. The withdrawal of NaF and As₂O₃ treatment produced incomplete recovery. However, the administration of ascorbic acid, calcium, and vitamin E, alone or in combination, during the withdrawal period resulted in the recovery of muscle. The combined treatment was more effective than treatment with the individual antidotes. Ascorbic acid may act through its strong reducing and antioxidant properties which activate hydroxylating enzymes and influence the redox potential of tissues. Ascorbic acid and calcium both inhibit phosphodiesterase thus elevating cyclic AMP levels which would influence muscle metabolism. Vitamin E (α -tocopherol) scavenges free radicals and this activity is enhanced in the presence of ascorbic acid.

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Keywords: Arsenic poisoning, Arsenic-fluoride poisoning, Ascorbic acid, Fluoride toxicity, Gastrocnemius muscle, Mice, Muscle effects, Vitamin E.

Serum fluoride and bone turnover markers in humans

The concentrations of fluoride in bone tissues increase with age, reaching several hundred $\mu\text{g/g}$ after age 60 years. Bone turnover changes with age, especially after the menopause in women. Serum fluoride concentrations may also increase with increased bone turnover. The aim was to investigate the relationship between markers of bone turnover and serum fluoride ion concentrations. The subjects were 143 healthy women, aged 20 to 77 years, chosen from women undergoing health examinations in the agricultural areas adjacent to Morioka city. They lived in areas with fluoride concentrations of $< 0.05 \text{ mg/L}$ in the drinking water and $< 0.01 \text{ mg/m}^3$ in the air. They were seen 5 or more hours after their last meal. Serum samples were stored at -80°C until analysis. The serum fluoride ion concentrations were measured using the flow-injection apparatus with a fluoride ion selective electrode. The serum was diluted 5 times using the diluting solution, 0.02 mol/L CH₃COONa (pH 3.0), and the pH value was adjusted to 5.4-5.6 using less

than 10 μL of 0.5 N HCl or 0.5 N NaOH. Bone alkaline phosphatase (B-ALP) and cross-linked carboxyterminal telopeptide of type I collagen (I CTP) in the serum were measured as markers of bone remodelling and bone resorption respectively. The mean serum fluoride ion concentration of 143 subjects was 7.21 ± 2.79 $\mu\text{g/L}$. The serum fluoride ion concentrations increased linearly with age ($p < 0.001$, $r = 0.583$). The mean concentrations of B-ALP and I CTP in the serum were 28.4 ± 11.4 U/L and 3.15 ± 0.70 $\mu\text{g/L}$ respectively. Significant relationships were present between serum fluoride and B-ALP ($p < 0.001$, $r = 0.453$) and serum fluoride and I CTP ($p = 0.012$, $r = 0.211$). Serum fluoride ion concentrations may be a useful marker of bone metabolism.

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Keywords: Alkaline phosphatase, Bone turnover, Fluoride accumulation, Fluoride in serum, Type I collagen.

Renal damage from continuous intravenous administration of sodium fluoride to rats

The aim was to study the renal effects of the continuous intravenous administration of sodium fluoride solution at the rate of 3 ml/hr, through the jugular vein using an infusion pump in 3 groups of male Wistar rats for 6 hr. The doses of F were 0, 18 and 36 mg/kg respectively. Urine samples, collected from the bladder every 2 hr, were analysed for urine volume, the excretion of F, creatinine (Cr), N-acetyl-beta-glucosaminidase (NAG), and μ -glutathion-S-transferase (GST). GST was found in high concentrations in the distal straight and convoluted regions of the rat renal tubules and may indicate distal tubule damage. F excretion increased with the increase in dose. No significant decrease of Cr excretion was observed. NAG and GST excretion were increased in the 36 mg/kg group. The doses of fluoride given would have been lethal if given as a single injection. The accumulation of F caused time dependent renal damage with GST being a more sensitive nephrotoxic indicator of acute renal tubular nephrotoxicity than NAG.

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Keywords: Intravenous NaF, Rat study, Renal damage, Urinary GST, Urinary NAG, Urinary parameters.

High fluoride concentrations in the serum and bone of patients with chronic renal failure

The aim was to study the effect of ingested fluoride in patients with chronic renal failure (CRF). Serum fluoride concentrations were measured in 104 subjects, who formed three groups: nondialyzed CRF, dialyzed CRF, and a control group. The iliac bone fluoride was measured in 20 subjects. Serum, urine and water fluoride concentrations were determined by the method of Fuchs. The bone calcium and fluoride levels were measured by non-destructive neutron activation analysis. Serum concentrations of fluoride in the nondialyzed and dialyzed CRF groups and the control group were 46.4 ± 12.6 (n=14), 28.2 ± 12.3 (n=40), and 7.92 ± 2.64 (n=50) $\mu\text{g/L}$ respectively. Serum fluoride levels in both patient groups were higher than in the control group ($p < 0.05$). The serum fluoride level of the nondialyzed patients correlated with their creatinine clearance ($r = 0.379$), indicating that fluoride excretion from the body was dependent on renal function. In dialyzed patients, the serum fluoride level decreased from 28.2 ± 12.3 to 14.7 ± 4.57 $\mu\text{g/L}$ (n=40) 4 hours after a single hemodialysis. The fluoride concentration of the tap water was 53.1 ± 15.9 $\mu\text{g/L}$ (n=8). The fluoride content of iliac bone obtained from dialyzed patients was 4640 ± 106 $\mu\text{g/g}$ dry weight and were approximately 10 times higher than the control 440 ± 35 $\mu\text{g/g}$ dry weight ($p < 0.001$) suggesting that skeletal fluorosis occurs in patients with CRF using fluoridated water. The bone calcium of the dialyzed patients and the control group were 119 ± 13 mg/g and 125 ± 29 mg/g ($p = \text{ns}$) respectively. Fluoride excretion was dependent on renal function with the serum fluoride concentration increasing in both the nondialyzed and dialyzed patients. Bone fluoride levels increased over 10 times those of the control group. This strongly suggests that the high fluoride contents in the bone is one of the factors inducing renal osteodystrophy. These results therefore strongly indicate that fluoridation of drinking water will be toxic for patients having decreased renal function.

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Keywords: Chronic renal failure, Fluoride in bone, Fluoride in serum, Hemodialysis, Renal function, Renal osteodystrophy.

Cortical and trabecular bone fluoride in female patients with proximal femur fractures

In this study the relationship between bone fluoride concentration, age, bone mineral density (BMD) and fracture location was examined in 51 female patients with an average age of 73 years (range, 54-92 years) who had

undergone hip arthroplasty because of proximal femur fracture. The hip fractures were divided using the AO/ASIF classification of fractures. There were 42 articular fractures and 9 supratrochanteric fractures. The bone fluoride content was measured in samples of cortical bone taken from the resected femoral head. The fluoride concentration was measured with an Orion fluoride ion-selective electrode after dissolving the defatted bone pieces in perchloric acid. Bone mineral density measurements were performed in the contralateral femoral neck and Ward triangle by dual energy x-ray absorptiometry at 7 days after operative treatment. Linear regression analysis was done to determine whether a correlation existed between the fluoride concentration, BMD, and the patients' age. Student's unpaired t-test was used to compare bone fluoride concentrations in the articular and supratrochanteric fractures. There was a very strong positive correlation between age and fluoride concentration in the cortical bone ($r=0.71$, $P<0.0001$), and age and fluoride concentration in the trabecular bone ($r=0.78$, $P<0.0001$). A strong negative correlation was found between BMD in the Ward triangle and the fluoride concentration in the trabecular bone ($r=-0.53$, $P<0.0001$). A strong negative correlation was also observed between BMD in the femoral neck and the fluoride concentration in the cortical bone ($r=-0.56$, $P<0.0001$). Patients with supratrochanteric fractures had higher fluoride concentrations in the cortical bone ($P<0.001$) compared to patients with articular fractures.

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Keywords: Bone fluoride, Bone mineral density, Cortical bone, Femoral neck, Hip fractures, Trabecular bone.

Positive correlation between fluoride in drinking water and markers of bone resorption

The aim was to study the effects of a high level of exposure to fluoride (F) on bone metabolism in women, aged 33 to 45 years, living in the grassland area of Inner Mongolia. We measured the concentrations of F in urine and drinking water from the household of each subject. Serum bone specific alkaline phosphatase (B-ALP) and osteocalcin (OC) were used as markers of bone formation, and urine CrossLaps (CL), free deoxypyridinoline (f-Dpyr), total deoxypyridinoline (t-Dpyr) and hydroxyproline (Hyp) as markers of bone resorption. Other calcium metabolism indices were also measured. The average F concentration in the drinking water in the grassland area, 3.57 ppm, was higher than that in the low F urban area in Inner Mongolia, 0.40 ppm. The urinary fluoride concentration, 8.15 mg F/g creatinine was also higher than that in the low F area, 1.20 mg F/g creatinine.

The subjects were divided into a younger group of 23 subjects, 33-40 years, and an older group of 15 subjects, 41-45 years. The subjects were also divided into 4 groups according to the F levels in the drinking water: group I 0-2 ppm; group II 2.1-4 ppm; group III 4.1-6 ppm; group IV 6.1-8 ppm and the markers of bone metabolism were compared in the different age and F level groups. In the younger group, the F/g creatinine levels in groups III and IV were significantly higher than those in groups I and II, and the drinking water F was positively correlated with the F/g creatinine. Urine F/g creatinine is thus suggested as an index of F exposure in this younger group.

The markers of bone turnover in group I showed no significant difference from those in the low F area. However the urine CL/g creatinine and f-Dpyr/g creatinine in groups III and IV were higher than those in groups I and II. The T-Dpyr/g creatinine was higher in group IV and showed a trend to be higher in group III. The serum OC in groups III and IV tended to be higher than in group II. However the B-ALP showed no increase. The PTH was lower in groups III and IV than in group I. The F level in the drinking water was positively correlated with the level of the markers for bone resorption. A significant acceleration of bone resorption was seen when the water F was more than 4 ppm. In the older group, the water F showed no correlation with the markers of bone resorption and the urine F/g creatinine.

The CL/g creatinine and Hyp/g creatinine in group I were higher than those in group I of the younger group. A trend to a higher level of $1,25(\text{OH})_2\text{VD}_3$ was also seen. In this older group the urine F/g creatinine was not seen to be a suitable index of F exposure. In conclusion it is suggested that F accelerates bone resorption and that for women experiencing long term exposure to higher levels of F, the urine F/g creatinine is not a suitable index of F exposure.

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Keywords: Alkaline phosphatase, Bone, Fluoride in water, Inner Mongolia, Osteocalcin, Urine crosslaps, Urine deoxypyridinoline, Urine hydroxyproline.

Compressive strength of cancellous bone in young fluoride-treated rats

The aim of the study was to investigate the effect of fluoride exposure on cancellous bone strength in rats. Twenty-eight 6-weeks-old female rats were randomized into four groups. One group served as a control group and received distilled water to drink while the other three groups received water containing sodium fluoride (NaF) at levels of 8, 30, and 60 mg F/L. The rats

were observed for 6 weeks. The left femurs from each rat were extracted at the time of necropsy. The bones were frozen in saline-soaked tissues and stored at -20°C . Prior to mechanical testing, the femurs were slowly thawed overnight at 7°C and held at room temperature on the day of the testing. The mechanical tests were performed by recording the forces necessary to penetrate the cancellous bone of the distal femoral rat bone with a 1.0 mm needle, rounded at the end. The penetration forces at 2.0, 3.0, and 4.0 mm were calculated. Strength values were reported as maximal penetration force and mean penetration stress at 2.0, 3.0, and 4.0 mm or as penetration work from the initial phase of needle penetration to 6.0 mm depth. The highest values of maximal penetration force, mean penetration strength and penetration work were found in the group exposed to NaF at 8 mg F/L. The maximal penetration force for the group treated with 8 mg F/L was significantly higher than in the control group ($P<0.05$) and in the 60 mg F/L ($P<0.01$). The mean penetration strength was significantly higher in the 8 mg/L group than in the control group ($P<0.01$) and in the 60 mg F/L ($P<0.05$). The penetration work for the group exposed to 8 mg F/L was significantly higher than in the control group ($P<0.001$). The results of this study show that fluoride, at a level of 8 mg F/L in drinking water, increases the compressive strength of cancellous bone in growing rats.

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Keywords: Bone fluoride, Bone penetration, Cancellous bone, Compressive strength, Fluoride in water, Young rats.

RESEARCH REVIEWS

Monitoring endemic fluorosis in China: 1991-1999

The defluoridation of drinking water and the provision of cooking stoves with an outside flue are seen to be the most effective measures for controlling endemic fluorosis in China. The rate of improvement in 1999, compared to 1991, was 12.7% for water defluoridation and 21.0% for cooking stoves. In the endemic fluorosis areas, drinking water fluorosis decreased from 60.08% in 1991 to 43.45% in 1999, and for coal-burning fluorosis the decrease was from 70.41% in 1991 to 53.76% in 1999. Improving the rate of construction of the defluoridation projects and strengthening their management is necessary to increase their benefits.

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Keywords: China, Coal burning, Defluoridation, Endemic fluorosis.

Fluoride deposition in atheromatous plaques

Consistent with the thrombogenic theory of Rokitansky, 1842, that the contents of atheromatous plaques are derived from the circulating blood, the possibility that blood fluoride is deposited in such plaques, "fluorification", is raised for consideration.

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Keywords: Fluoride content in atheromatous plaque.

FLUORIDE IN DENTISTRY

RESEARCH REPORTS

Fluoride and magnesium content in superficial enamel layers of permanent human teeth in relation to age

The aim of the study was to determine the relationship between age, and the fluoride and magnesium content in the superficial layers of the enamel of human teeth. Ten upper premolar teeth extracted from children, aged 14 years, and 10 from adults, aged 45-50 years, receiving dental care in Białystok were examined. The mineral composition in the three superficial layers of enamel of the buccal surface was measured using the acidic biopsy method. The calcium and magnesium content was measured with atomic absorption spectrometry while fluoride was measured with gas chromatography. The fluoride content of the adult teeth was significantly higher than that in the children's teeth. This was attributed to the accumulation of calcium fluoride. The children's teeth contained more magnesium. The total and structural fluoride and magnesium decreased in parallel to layer depth.

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Keywords: Age, Białystok, Poland, Calcium in enamel, Dental enamel, Fluoride accumulation, Magnesium in enamel, Premolar teeth.

Fluoride levels in the enamel of permanent teeth of children exposed to fluoride pollution

The aim was to determine the enamel fluoride content of the permanent teeth in 14-year-old children residing in two cities, Szczecin and Białystok, which differed in the level of environmental fluoride pollution. The mineral composition in the three superficial layers of the buccal surface enamel, of

10 teeth from each city, was determined using the acidic biopsy method as modified by Łagocka. Calcium was measured with atomic absorption spectrometry and fluoride with gas chromatography. A higher environmental fluoride level was associated with an increase of structural fluoride in the superficial layers of the enamel. The total and structural fluoride content decreased in parallel to the layer depth.

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Keywords: Dental enamel, Fluoride in enamel, Fluoride pollution.

Foam preparations of sodium fluoromonophosphate and sodium fluoride are ineffective for the management of dentin hypersensitivity

The aim was to assess the effectiveness of two fluoride compounds in the form of foam for the management of dentin hypersensitivity. Twenty-one patients with dentin hypersensitivity, divided into three equal groups, received one spoonful of a preparation every seven days on five occasions: group A 1.23% sodium fluoromonophosphate pH 3.5; group B 0.9% neutral sodium fluoride; group C 0.9% sodium chloride (placebo). The preparation remained in the oral cavity for one minute. The results were evaluated after 6 weeks from the beginning of the treatment. Enamel biopsy was done in each patient before and after therapy and the fluoride content was measured with a fluoride electrode. Calcium levels were determined with atomic absorption spectroscopy. The fluoride content in the superficial layer of enamel 4.2 µm thick increased by 3087.6 ppm in group A and by 721.9 ppm in group B. No change was noted in group C. With the exception of two patients in group A (28.6%) reporting partial improvement, no other patients benefited from this therapy. In spite of elevating the content of fluoride in the superficial layer of enamel, fluoride preparations in the form of foam were ineffective for the treatment of dentine hypersensitivity.

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Keywords: Dental caries, Dentin hypersensitivity, Fluoride dental foam, Fluoride professional applications, Fluoride in saliva, Sodium fluoride, Sodium monofluorophosphate.

Release of fluoride into enamel and saliva from paracervical dental filling materials

The aim was to compare the quantity of fluoride released into artificial saliva and enamel from three materials used for filling paracervical defects. Cylindrical defects 1 mm deep and 2 mm in diameter were produced on the buccal surface near the neck and filled according to the manufacturer's instructions with: group A: polyacid modified composite resin, Compoglass F (Vivadent); group B: resin modified glass ionomer cement, Fuji II LC Improved (GC); group C: fluoride-releasing composite Esthet X (Dentspy). Fluoride release was monitored for 20 days by placing each tooth in a plastic test tube filled with 3 ml of artificial saliva (pH 7) at 37°C. The tooth was transferred to a new tube every 24 hr and the fluoride concentration in the saliva was measured with an ion-selective electrode. Enamel biopsy was done before and after 20 days of contact with the filling material. The calcium content was determined with atomic absorption spectrometry. The concentration of fluoride in the saliva after 24 hr from application of the filling material was: group A 40 µg/mL, group B 120 µg/mL, group C 40 µg/mL. The peak fluoride concentration was observed on day 10 in group A and on day 1 in group B. Concentrations remained constant in group C. After 15 days the release of fluoride stabilized at 30 µg/mL in all groups. The highest total quantity of fluoride released into the saliva was in group B, followed by groups A and C. The increases in the enamel content of fluoride were: group A 96.6 ppm; group B 30.8 ppm; group C 45.4 ppm. The fluoride in group A originated from ytterbium trifluoride and barium-aluminium-fluorosilicate glass.

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Keywords: Dental caries, Fluoride professional applications, Fluoride in saliva.

RESEARCH REVIEWS

30 years of accidental dental fluorosis in Takarazuda and Nishinomiya in Japan

In 1971 many cases of dental fluorosis were detected in Takarazuka city (180 000 inhabitants) and Nishinomiya city (410 000 inhabitants) near Osaka city due to inadequate control of the fluoride in the water supply. Many persons were afflicted by lower concentrations of fluoride than 0.8 ppm, thus showing that the National Standard Level of 0.8 ppm is not necessarily safe. Compensation for damages to individuals is still being awarded by the administration of both cities. Data were available from the examina-

tional and judging committees of both cities. The cost of treatment, 100 000 JPY per tooth, in Nishinomiya city for the period 1977-2000 has been 53 236 380 JPY (approved for treatment: males 154, females 166; treated: males 26, females 40) and in Takarazuka city for the period 1976-1087 has been 152 808 000 JPY (approved for treatment: males 630, females 742; treated: males 73, females 127). In conclusion it was impossible to set the "optimum" level of fluoride as the level of 0.8 ppm was not safe in several areas. The total daily intake of fluoride needs to be considered. We are concerned that the Administration of Health and Welfare has again permitted, from November 2000, water fluoridation up to 0.8 ppm.

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Keywords: Dental fluorosis, Fluoride in water, Nishinomiya, Japan, Takarazuka, Japan.

Fluoridation and dental health

Although fluoride has been added to drinking water in some countries for over 50 years, with the intention of improving dental health, the measure has remained controversial and much additional research is needed.

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Keywords: Fluoridation.

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