SISTER-CHROMATID EXCHANGES IN LYMPHOCYTES OF WORKERS AT A PHOSPHATE FERTILIZER FACTORY

Z Q Meng, H Q Meng and X L Cao
Taiyuan, China

Abstract from Mutation Research - Environmental Mutagenesis and Related Subjects 334 (2) 243-246 1995

The frequencies of sister-chromatid exchange (SCE) in peripheral blood lymphocytes of 40 workers at a phosphate fertilizer factory in North China were studied. HF and SiF$_4$ are main air pollutants in the factory, there is also some dust containing fluoride, phosphate fog, NH$_3$ and SO$_2$. It was shown that the chemicals caused an increase in SCE, and also induced cell mitotic delays. The mean SCEs/cell of the workers and the non-exposed controls were $7.47 \pm 0.31$ and $4.94 \pm 0.14$ ($p < 0.01$) respectively. SCEs/cell in 75% of 40 workers were higher than 6 while 40 controls all had values lower than 6. SCE frequencies of the workers increased with length of the chemical exposure period up to 10 years. Smoking enhanced the SCE frequencies induced by the chemicals.

Key words: Fluoride; Human; Lymphocytes; Phosphate fertiliser factory; SCE; Sister-chromatid exchanges; Smoking.
Reprints: Z Q Meng, Shanxi University, Department of Environmental Science, Taiyuan 030006 China.

SISTER-CHROMATID EXCHANGES AFTER EXPOSURE TO METAL-CONTAINING EMISSIONS

K Sivikova and J Dianovsky
Kosice, Slovakia

Abstract from Mutation Research - Fundamental and Molecular Mechanisms of Mutagenesis 327 (1-2) 17-22 1995

The effect of in vivo feeding of metal-containing emissions from an aluminum refining plant upon the induction of sister-chromatid exchanges (SCEs) and mitotic delay was investigated in cultured sheep lymphocytes. The experimental animals were given a daily dose of industrial emissions (mostly aluminum and fluoride) of either 0.75 g or 1.5 g/animal, for 1 year. The experiments were aimed at induction of chronic fluorosis under clinical conditions. A significant increase in mean serum fluoride, aluminum, arsenic and cadmium levels between both experimental groups and control was observed from 5 and 7 months on, respectively. The occurrence of SCEs in the experimental groups was higher (reaching statistical significance at a dose of 1.5 g/animal) than that seen in the controls. The emission was found to reduce the proliferation index. A significant heterogeneity of the first, second and third metaphases as compared to the controls was also observed.

Key words: Complex metal mixture; Fluorosis; Proliferation index; Sheep lymphocytes; Sister-chromatid exchange.
Reprints: K Sivikova, University of Veterinary Medicine, Department of Veterinary Genetics, Kosice 04181 Slovakia.
CHROMOSOME ABERRATIONS IN CULTURED RAT BONE MARROW CELLS TREATED WITH INORGANIC FLUORIDES

A M Khalil
Irbid, Jordan

Abstract from Mutation Research - Genetic Toxicology 343 (1) 67-74 1995

The genotoxic effects of inorganic fluorides were investigated by treating cultured rat bone marrow cells with varying concentrations (0.1-100 μM) of potassium fluoride (KF) and sodium fluoride (NaF) for different durations (12, 24 and 36 h) and measuring the incidence of cells with aberrations and number of breaks per cell. Both forms of fluoride were found to be weak mutagens relative to the positive control N-methyl-N-nitro-N-nitrosoguanidine (MNNG). A specificity of fluoride ion in inducing chromosome aberrations (CA) was indicated by the observation that both NaF and KF behaved almost equivalently in this study and at significantly higher variations from the results with potassium chloride (KCl) and sodium chloride (NaCl).

Key words: Bone marrow cells; Chromosomal aberrations; Inorganic fluorides.
Reprints: A M Khalil, Yarmouk University, Department of Biological Sciences, Irbid, Jordan.

REVERSIBLE EFFECTS OF SODIUM FLUORIDE INGESTION ON SPERMATOZOA OF THE RAT

M V Narayana and N J Chinoy
Ahmedabad, India


The effects of ingestion of sodium fluoride (NaF), 10 mg/kg body weight for 50 days, on the structure and metabolism of sperm of albino rats (Rattus norvegicus), were investigated. In different groups of rats, the reversible effects upon withdrawal of NaF treatment and by administering some therapeutic agents, viz., ascorbic acid and calcium alone and in combination with NaF (50 and 70 days), on sperm structure and metabolism were also studied. The results revealed that the sperm acrosomal hyaluronidase and acrosin were reduced after 50 days of NaF treatment. Sperm stained with acidic alcoholic silver nitrate revealed acrosomal damage and deflagellation, which might be causative factors for the reduced activity of the enzymes. These alterations also resulted in a decline in sperm motility. The cauda epididymal sperm count was decreased, perhaps because of spermatogenic arrest. Thus, the low sperm motility and count ultimately contributed toward reduction in fertility by NaF treatment. However, withdrawal of NaF treatment for 70 days produced incomplete recovery, while administration of ascorbic acid and calcium, individually and in combination, brought about significant recovery of fluoride-induced effects. Thus, the effects of fluoride on sperm structure and metabolism of rats are transient and reversible.

Key words: Rats; Sodium fluoride; Spermatozoa.
Reprints: M V Narayana, Reproductive Endocrinology and Toxicology Unit, School of Sciences, Gujarat University, Ahmedabad, India.
EFFECTS OF CHRONIC FLUORIDE TOXICITY ON THE MORPHOLOGY OF DUCTUS EPIDIDYMIS AND THE MATURATION OF SPERMATOZOA OF RABBIT

A Kumar and A K Susheela
New Delhi, India

Abstract from International Journal of Experimental Pathology 76 (1) 1-11 1995

This study used light and scanning electron microscopy to observe the effect of chronic fluoride toxicity on the structure of the ductus epididymis, testis and spermatozoa in rabbit. The rabbits were treated with 10 mg NaF/kg body weight/day for 20 and 23 months. Serum fluoride was estimated by the fluoride ion-specific electrode method. Fluoride levels in the sera of both 20 and 23-month treated rabbits were significantly increased (P < 0.001). Loss of stereocilia, significant decrease (P < 0.001) in the height of the pseudostratified columnar epithelium and significant increase (P < 0.001) in the diameter of both the caput and cauda ductus epididymis were observed only in the 23-month fluoride treated rabbits. The decreases in the epithelial cell height (P < 0.01) and the tubular diameter (P < 0.001) of the testis were significant only in 23-month treated animals. Spermatozoa in the lumen of the testis of both treated groups of animals and in the caput and cauda ductus epididymis of 20-month treated animals appeared normal, but spermatozoa in the caput and cauda ductus epididymis of 23-month treated animals were fragmented. In the 23-month fluoride treated rabbits, the weights of the caput and cauda ductus epididymis were significantly reduced (P < 0.025) and there was also a reduction in the number of secretory granules in these organs. The structural changes observed in the caput and cauda ductus epididymis might adversely affect the maturation of spermatozoa.

Key words: Fluoride; Epididymis; Stereocilia; Spermatozoa; Testis; Ultrastructure.
Reprints: A K Susheela. All India Institute of Medical Sciences, Department of Anatomy, Fluroide and Fluorosis Research Laboratory, New Delhi 110029, India

NEUROTOXICITY OF SODIUM FLUORIDE IN RATS

P J Mullenix, P K Denbesten, A Schunior and W J Kernan
Andover, Massachusetts, USA

Abstract from Neurotoxicology and Teratology 17 (2) 169-177 1995

Fluoride (F) is known to affect mineralizing tissues, but effects upon the developing brain have not been previously considered. This study in Sprague-Dawley rats compares behavior, body weight, plasma and brain F levels after sodium fluoride (NaF) exposures; during late gestation, at weaning or in adults. For prenatal exposures, dams received injections (SC) of 0.13 mg/kg NaF or saline on gestational days 14-18 or 17-19. Weanlings received drinking water containing 0, 75, 100, or 125 ppm F for 6 or 20 weeks, and 3 month-old adults received water containing 100 ppm F for 6 weeks. Behavior was tested in a computer pattern recognition system that classified acts in a novel environment and quantified act initiations, total times and time structures. Fluoride exposures caused sex- and dose-specific behavioral deficits with a common pattern. Males were most sensitive to prenatal day 17-19 exposure, whereas females were more sensitive to weanling and adult
exposures. After fluoride ingestion, the severity of the effect on behavior increased directly with plasma F levels and F concentrations in specific brain regions. Such association is important considering that plasma levels in this rat model (0.059 to 0.640 ppm F) are similar to those reported in humans exposed to high levels of fluoride.

Key words: Brain; Fluoride; Neurotoxicity.

Reprints: P J Mullenix, POB 753, Andover, MA 01810 USA.

CHANGES IN THE PLASMA ELECTROLYTES AND METABOLITES OF THE RAT FOLLOWING ACUTE EXPOSURE TO SODIUM FLUORIDE AND STRONTIUM CHLORIDE

J Appleton
Liverpool, England

Abstract from Archives of Oral Biology 40 (4) 265-268 1995

Acute exposure of rats to strontium or fluoride by i.p. injection of sodium fluoride or strontium chloride resulted in a systemic response in which changes occurred in the plasma electrolytes and metabolites. Strontium resulted in a rapid but temporary hypercalcaemia while fluoride produced a temporary hypocalcaemia. There was no significant hypophosphataemia after fluoride and only a transient hypophosphataemia with strontium. There was some indication of kidney damage and a general stress response following fluoride injection. These results do not support the hypothesis that interglobular dentine is associated with hypophosphataemia or hypoplastic enamel with hypocalcaemia and are in conflict with the observation that the formation of interglobular dentine following the injection of lead acetate is associated with hyperphosphataemia and hypercalcaemia.

Key words: Electrolytes; Fluoride; Metabolites; Rats; Strontium.

Reprints: J Appleton, University of Liverpool School of Dentistry, Liverpool L69 3BX, Merseyside, England.

COMPARATIVE STUDY OF THE EFFECT OF SODIUM FLUORIDE AND SODIUM MONOFLUOROPHOSPHATE ON GLUCOSE HOMEOSTASIS IN THE RAT

A Rigalli, R Alloatti, I Menoyo and R C Puche
Santa Fe, Rosario, Argentina

Abstract from Arzneimittel - Forschung/Drug Research 45-1 (3) 289-292 1995

The effect of acute and chronic administration of sodium fluoride (NaF) or sodium monofluorophosphate (MFP) on the glucose homeostasis of the rat are compared.

The oral administration of a single dose of 40 µmol/100 g bw of either compound produced similar increases in plasma glucose (up to 1.8 g /L) and diffusible fluoride (up to 130 µmol/L).

In long-term experiments (three months of duration), treatment with NaF (a 5 µmol/L solution as the water supply) produced, in the first month of experiment,
abnormal glucose tolerance tests and increased plasma diffusible fluoride levels (range: 2-12 μmol/L). Treatment with MFP, on the other hand, did not affect glucose homeostasis; plasma diffusible fluoride was always below 2 μmol/L.

The results of these experiments indicate that glucose homeostasis is affected when plasma diffusible fluoride exceeds 5 μmol/L. The basal and glucose-stimulated insulin secretion of isolated Langerhans rat islets (incubated with solutions containing 2, 5, 10 and 20 μmol/L NaF) was significantly inhibited by 5 to 20 μmol/L fluoride. No effect was observed under similar conditions with MFP at concentrations of 2, 5, 10, 20 and 50 μmol/L.

Key words: Antiosteoporotic drugs; Glucose homeostasis; Pharmacology; Sodium fluoride; Sodium monofluorophosphate.
Reprints: R C Puche, Faculty of Medical Science of Rosario, Department of Physiology, Santa Fe 3100, RA-2000 Rosario, Argentina.

INTAKE AND METABOLISM OF FLUORIDE

G M Whitford
Augusta, Georgia, USA

Abstract from Advances in Dental Research 8 (1) 5-14 1994

The purpose of this paper is to discuss the major factors that determine the body burden of inorganic fluoride. Fluoride intake 25 or more years ago was determined mainly by measurement of the concentration of the ion in the drinking water supply. This is not necessarily true today because of ingestion from fluoride-containing dental products, the “halo effect”, the consumption of bottled water, and the use of water purification systems in the home. Therefore, the concentration of fluoride in drinking water may not be a reliable indicator of previous intake. Under most conditions, fluoride is rapidly and extensively absorbed from the gastrointestinal tract. The rate of gastric absorption is inversely related to the pH of the gastric contents. Overall absorption is reduced by calcium and certain other cations and by elevated plasma fluoride levels. Fluoride removal from plasma occurs by calcified tissue uptake and urinary excretion. About 99% of the body burden of fluoride is associated with calcified tissues, and most of it is not exchangeable. In general, the clearance of fluoride from plasma by the skeleton is inversely related to the stage of skeletal development. Skeletal uptake, however, can be positive or negative, depending on the level of fluoride intake, hormonal status, and other factors. Dentin fluoride concentrations tend to increase throughout life and appear to be similar to those in bone. Research to determine whether dentin is a reliable biomarker for the body burden of fluoride is recommended. The renal clearance of fluoride is high compared with other halogens. It is directly related to urinary pH. Factors that acidify the urine increase the retention of fluoride and vice versa. The renal clearance of fluoride decreases and tissue levels increase when the glomerular filtration rate is depressed on a chronic basis.

Key words: Bone; Diet; Fluoride; Fluoridation; Metabolism.
Reprints: G M Whitford, Department of Oral Biology, School of Dentistry, Medical College of Georgia, Augusta, Georgia 30912-1129 USA.
URINARY FLUORIDE EXCRETION IN CHILDREN WITH LOW FLUORIDE INTAKE OR CONSUMING FLUORIDATED SALT

T M Marthaler, M Steiner, G Menghini and P De Crousaz
Zurich, Switzerland

Abstract from Caries Research 29 (1) 26-34 1995

The purpose of this study was to compare urinary fluoride excretion with fluoride ingestion in children who had either a low fluoride intake or received supplemental fluoride through salt or water. The urinary samples were collected in two ways. In procedure A, urine was collected in the morning, at noon and during the afternoon. This covered a continuous period of approximately 7 h from the beginning to the end of the school day. In procedure B, morning, afternoon and night samples were collected. The morning and afternoon samples were obtained under supervision at school. Procedure B was more useful than procedure A for monitoring salt fluoridation projects. Children with a history of low fluoride intake excreted a mean of 10 micrograms F/h during the night and the morning, but 13-16 micrograms F/h after the main meal. Children in a water-fluoridated town or in comprehensive salt fluoridation programs excreted between 19 and 33 micrograms F/h. However, after the intake of meals prepared with fluoridated salt (250 ppm F) the mean excretion of fluoride ranged between 31 and 49 micrograms F/h. The levels of excretion corresponded with the levels of fluoride intake and meal patterns in the various groups.

Key words: Fluoride excretion; Fluoride monitoring; Urinary excretion.
Reprints: T M Marthaler, Department of Preventive Dentistry, Periodontology and Cariology, Dental Institute of University of Zurich, PO Box CH-8028, Zurich, Switzerland.

URINARY FLUORIDE EXCRETION IN JAMAICA IN RELATION TO FLUORIDATED SALT

R A Warpeha and T M Marthaler
Kingston, Jamaica

Abstract from Caries Research 29 (1) 35-41 1995

The purpose of this study was to compare the urinary fluoride levels in subtropical Jamaica just before the beginning of salt fluoridation (250 mg/kg salt) in 1987 and again 20 months later. Four age groups were studied in three locations with low to intermediate fluoride concentrations in the drinking water. The averages of supervised, timed morning fluoride excretions ranged between 12.1 and 27.9 micrograms/h prior to fluoridation as compared with 23.7-67.4 micrograms/h 20 months after the beginning of the sale of fluoridated salt. The fluoride excretions obtained from 24-hour collections ranged from 169 to 485 micrograms/24 h in 1987 and increased to 304-657 micrograms/24 h in 1989. In 1987, the morning fluoride excretions approximated those of French and Swiss children who had a low fluoride intake, but the fluoride levels of 1989 were similar to the data obtained from children who had an intake of 1.2-1.7 mg F/day or consumed fluoridated water.

Key words: Fluoride excretion; Fluoride monitoring; Salt fluoridation; Urinary fluoride.
Reprints: T M Marthaler, Department of Preventive Dentistry, Periodontology and Cariology, Dental Institute of University of Zurich, PO Box CH-8028, Zurich, Switzerland.
THE EFFECT OF FLUORIDATED DRINKING WATER ON AXIAL BONE MINERAL DENSITY - A POPULATION-BASED STUDY

H Kroger, E Alhava, R Honkanen, M Tupperainen and S Saarikoski
Kuopio, Finland

Abstract from Bone and Mineral 27 (1) 33-41 1994

Bone mineral density (BMD) of the spine and femoral neck was measured in a random stratified sample of 3222 perimenopausal women aged 47-59 years. A total of 969 women had used fluoridated drinking water (1.0-1.2 mg/L) for over 10 years. These women were compared with 2253 women with low levels of fluoride in drinking water (< 0.3 mg/L). BMD of the spine was significantly higher in the fluoride group than in the non-fluoride group (1.138 ± 0.165 vs. 1.123 ± 0.156 g/cm², P = 0.026). Femoral neck BMDs did not differ between the groups. When the BMD values were adjusted for confounding factors (age, weight, menopausal status, calcium intake, physical activity level, deliveries, alcohol consumption and estrogen use), the differences between the groups increased (P < 0.001 for the spine and P = 0.004 for the femoral neck, respectively). There was no significant difference between the groups in the prevalence of self-reported fractures sustained during 1980-1989. We propose that the fluoridation of drinking water has a slight increasing effect on axial BMD in women in low fluoride areas.

Key words: Bone; Finland; Fluoridation; Hip fractures; Osteoporosis.
Reprints: H Kroger, Department of Surgery, Kuopio University Hospital, Finland.

EVIDENCE OF FLUORIDE-INDUCED EFFECTS ON THE CALCANEUS BY MEASUREMENTS OF BROADBAND ULTRASOUND ATTENUATION (BUA)

A Resch, F Pietschmann, P Bernecker, T Rand, R Willvonseder and H Resch
Vienna, Austria

Abstract from Rofo Fortschritte Auf Dem Gebiete Der Rontgenstrahlen Und Der Neuen Bildgebenden Verfahren 161 (6) 547-550 1994

We measured broadband ultrasound attenuation (BUA) of the calcaneus as well as radial (RBD) and spinal bone density (SBD) in normal and osteoporotic females (n = 188) both before treatment and after long-term fluoride therapy. RBD was measured using single photon absorptiometry of the non-dominant distal forearm (SPA) and spinal bone density (SBD) was assessed using quantitative CT (QCT). In untreated osteoporotic patients (n = 62), BUA as well as the other densitometric methods showed significantly lower values (p < 0.05) when compared to age matched controls (n = 75). Females with osteoporosis and long-term treatment with fluoride (n = 51) had both significantly higher BUA values and higher spinal bone density when compared to women with untreated osteoporosis. BUA and SBD were correlated in the entire group (r = 0.5, r < 0.0001) as well as in the subgroups of untreated (r = 0.48, p < 0.001) and fluoride-treated patients with osteoporosis (r = 0.4, p < 0.05). These results suggest that BUA provides further evidence of an osteogenic response to fluoride at peripheral weight-bearing skeletal sites such as the calcaneus.

Key words: Bone; Fluoride; Osteoporosis.
Reprints: A Resch, Universitatsklinik fur Radiodiagnostik, Wien, Austria.
INTERACTIVE EFFECTS OF FLUORIDE AND ALUMINUM UPTAKE AND ACCUMULATION IN BONES OF RABBITS ADMINISTERED BOTH AGENTS IN THEIR DRINKING WATER

H W Ahn, B Fulton, D Moxon and E H Jeffery
Urbana, Illinois, USA

Abstract from *Journal of Toxicology and Environmental Health* 44 (3) 337-350 1995

Fluoride (F) and aluminum (Al), which are known to form a strong complex, are both present in finished drinking water. The effect of F and Al on one another's tissue accumulation was determined using adult male New Zealand white rabbits. Thirty-six rabbits (three per group) were given Purina Rabbit Chow and drinking water containing no F or Al, F alone (1, 4, or 50 ppm F as NaF), Al alone, (100 or 500 ppm Al as AlCl₃), or a combination of F and Al, *ad libitum* for 10 wk. None of these treatments altered food intake or weight gain in these rabbits. However, rabbits treated with 1 ppm F and 500 ppm Al consumed significantly less water than control rabbits. The F accumulation in plasma, urine, incisors, and tibia was increased as the F addition to the drinking water increased within groups receiving a single concentration of Al. In contrast, F accumulation in plasma, urine, incisors, and tibia decreased as the Al concentration increased within groups receiving a single F concentration, indicative of decreased intestinal absorption. Importantly, Al levels in tibia were significantly increased by the addition of F to the drinking water, even in animals receiving no Al in their drinking water. The effect of F on Al accumulation in bone was confirmed by our evaluating Al levels in sterna harvested from rats treated with 0 or 79 ppm F (as NaF in the drinking water) in a study conducted by the National Toxicology Program (Bucher et al, 1991). Therefore, some of the osteotoxicity seemingly associated with high F levels in bone may be due to the accumulation of Al or an Al-F complex.

Key words: Accumulation; Aluminum; Bones; Fluoride; Rabbit.

Reprints: E H Jeffery, University of Illinois, Institute of Environmental Studies, 1101 W Peabody, Urbana, Illinois 61801 USA.

BONE MINERALIZATION AND HISTOMORPHOMETRY IN BIOPSIES OF OSTEOPOROTIC PATIENTS TREATED WITH FLUORIDE

M D Grynpas, D P Holmyard and K P H Pritzker
Toronto, Canada

Abstract from *Cells and Materials* 4 (3) 287-297 1994

Treatment of osteoporotic patients with sodium fluoride (NaF) has been shown to increase vertebral bone mass but there is a debate as to whether fluoride increases or decreases vertebral fractures. Using bone biopsies of patients investigated for osteoporosis which were either treated with NaF or untreated, we applied the technique of backscattered electron imaging (BSE) to determine the degree of mineralization of the bone biopsies. We found a significant increase in mineralization with fluoride treatment. We also found a poor correlation between fluoride dose and bone fluoride content. Finally all bone formation parameters were elevated in the fluoride treated bone. In conclusion, these studies demonstrate that fluoride treatment increases bone mineralization together with an increase in bone formation parameters.

Key words: Biopsies; Bone; Fluoride; Histomorphometry; Mineralization; Osteoporosis.

Reprints: M D Grynpas, Mt Sinai Hospital, Samuel Lunenfeld Research Institute, 600 University Ave, Toronto M5G 1X5 Canada.
MARKED DECREASE IN TRABECULAR BONE QUALITY AFTER FIVE YEARS OF SODIUM FLUORIDE THERAPY - ASSESSED BY BIOMECHANICAL TESTING OF ILIAC CREST BONE BIOPSIES IN OSTEOPOROTIC PATIENTS

C H Sogaard, L Moselkilde, A Richards and L Moselkilde
Aarhus, Denmark


In order to evaluate the effect of sodium fluoride (NaF) on bone biomechanical competence, iliac crest biopsies were taken before and after one year of treatment in 12 osteoporotic patients, and before and after five years of treatment in 14 patients. Bone fluoride content had increased significantly after both one and five years of treatment, indicating that the administered fluoride had been ingested. After one year of treatment, no difference was observed in iliac crest trabecular bone ash content. A general trend for decreased bone strength and bone quality was observed, but this was insignificant. After five years of fluoride treatment, an insignificant decrease in iliac crest trabecular bone ash content was observed. A significant reduction of 45% was found in trabecular bone strength (p < 0.05), and an even more pronounced reduction of 58% was found in trabecular bone quality (p < 0.01). The results of this study indicate that long-term administration of sodium fluoride may be detrimental to bone quality, at least as measured in non-loaded iliac crest trabecular bone.

Key words: Bone; Bone quality; Bone strength; Fluoride therapy; Iliac crest biopsies; Osteoporosis.


SODIUM FLUORIDE DOES NOT BUILD BONE IN AGED OVARIECTOMIZED RATS

P T Cheng, L Huang and N Low
Toronto, Canada

Abstract from Cells and Materials 4 (3) 299-307 1994

Sodium fluoride builds bone in young intact rats and young ovariectomized (OVX) rats. Here we show that NaF does not build bone in aged OVX rats. Wistar rats were OVX at nine months old and fed with regular rodent chow and de-ionized water ad libitum. After three months of conditioning, they were divided into four groups (five rats per group) and treated for four months as follows: 1) no treatment (control); 2) NaF solution (4 mmol/L) instead of water (NaF); 3) subcutaneous injections of pamidronate (0.25 mg/kg/d; 5 d/w) (APD); and 4) both NaF solution and pamidronate injections (APD + NaF). After four months, the OVX control had significantly less cancellous bone than a sham-operated control, but the NaF treated group has significantly less cancellous bone than the OVX control. Both APD and APD + NaF groups were not significantly different from the OVX control or the sham control. Biochemical analysis of rat urine showed that NaF treated group had significantly higher pyridinoline/creatinine values than all other groups, indicating that the lack of bone gain was due to excessive resorption. We have shown that NaF does not have an anabolic effect on bone in aged OVX rats. On the contrary, it shows a catabolic effect due to excessive resorption which is inhibited by APD.
This adverse finding of NaF on aged rats (12 month old rats ovariectomized for 3 months) does not agree with previous observations made in young rats.

Key words: Bone; Histomorphometry; Mineral content; Osteopenia; Osteoporosis; Paminodronate; Pyridinoline; Rats; Sodium fluoride.

Reprints: P T Cheng, University of Toronto, Mt Sinai Hosp, Department of Pathology, 600 University Ave, Toronto M5G 1X5 Canada.

EFFECT OF AGING ON ANIMAL RESPONSE TO CHRONIC FLUORIDE EXPOSURE
Indianapolis, Indiana, USA

Abstract from Journal of Dental Research 74 (1) 358-368 1995

This study was conducted to test the hypothesis that physiological changes which occur during aging increase the biological impact of fluoride and reduce the threshold of safe fluoride exposure. Four groups of rats were fed a low-fluoride diet (< 1.2 ppm) ad libitum and received 0, 5, 15, or 50 ppm fluoride in their drinking water. Animals were killed after three, six, 12, or 18 months of treatment. Blood and urine were monitored for biochemical markers of tissue function, and plasma, urine, feces, and representative tissues were analyzed for fluoride. In addition, bone marrow cells from animals killed after 18 months of treatment were examined for frequency of sister chromatid exchange (SCE), a marker of genetic damage. Study results indicated that, within treatment groups, fluoride intake, excretion, and retention did not change significantly between three and 18 months. Fluoride concentration in soft tissues did not change with treatment duration in the fluoride-treated animals. Mineralized tissue fluoride concentration and the total fluoride in the carcasses increased continually as the animals aged. In spite of significant, dose-related differences in tissue fluoride levels which occurred in all age groups in this study, there were no indications that increased fluoride in the tissues caused any adverse physiological or genotoxic effects. None of the monitored clinical “wellness” markers of tissue integrity and function was altered by fluoride in a clinically significant manner. Therefore, there was no evidence from this study that aging reduces the threshold of safe chronic fluoride exposure.

Key words: Age; Animals; Fluoride.

Reprints: A J Dunipace, Oral Health Research Institute, 415 Lansing St, Indianapolis, Indiana 46202 USA.

MANDIBULAR BONE FLUORIDE ACCUMULATION IN WILD RED DEER (CERVUS ELAPHUS L) OF KNOWN AGE
U Kierdorf, H Kierdorf, M Erdelen and Z Machoy
Aarhus, Denmark

Abstract from Comparative Biochemistry and Physiology A - Physiology 110 (4) 299-302 1995

Mandibular bone fluoride concentration and its relation to age were studied in a sample of 39 red deer of known age (between 2 and 18 years) from an area not exposed to increased fluoride deposition (Hart mountains, Germany). Bone fluoride level ranged from 208 to 1026 ppm dry weight and was positively correlated with age (r = 0.808, P < 0.001), the rate of skeletal fluoride accumulation being higher in
younger individuals. This indicates that during the period of skeletal growth, fluoride uptake is more rapid than in later life when accumulation at a lower rate occurs during normal bone turnover. Bone fluoride levels found in the present sample are regarded as resulting from "normal" fluoride exposure in a recent central European red deer population.

Key words: Age dependence; Bone; Cervus elaphus; Environmental fluoride; Fluoride accumulation; Fluoride exposure; Mandibles; Red deer.

Reprints: U Kierdorf, Aarhus University, Royal Dental College, Vennelyst Blvd, DK-8000 Aarhus, Denmark.

PHENYTOIN AND FLUORIDE ACT IN CONCERT TO STIMULATE BONE FORMATION AND TO INCREASE BONE VOLUME IN ADULT MALE RATS

T Ohta, J E Wergedal, T Matsuyama, D J Baylink and K H W Lau
Loma Linda, California, USA

Abstract from Calcified Tissue International 56 (5) 390-397 1995

We have recently demonstrated that phenytoin is an osteogenic agent at low doses. The present paper describes observations that a mitogenic dose (ie, 20 μM in BGJ(b) medium) of fluoride significantly augments the phenytoin-dependent stimulation of normal human bone cell proliferation and alkaline phosphatase (ALP) activity in cell culture. Additionally, the present study was designed to investigate whether fluoride and phenytoin would interact to increase bone formation in rats in vivo. Four groups of weight-matched adult male rats received daily I.P injection of 1) vehicle (10% DMSO), 2) 5 mg/kg/day phenytoin, 3) 5 mg/kg/day phenytoin and 50 ppm NaF, and 4) 50 ppm NaF and vehicle, respectively, for 36 days. Sodium fluoride (NaF) was delivered in drinking water. Blood samples were drawn weekly and analyzed for serum osteocalcin, ALP, calcium, phosphorus, and 25(OH)D-3. Rats were labeled with tetracycline at day 21 and 30 and histomorphometric analysis was carried out on the tibia at the end of the experiment. Neither agent by itself or together affected the serum calcium, phosphorus, or 25(OH)D, levels. All measures of bone formation, ie, serum osteocalcin level and ALP activity, bone ALP specific activity, mineral apposition rate, bone formation rate, and % bone formation surface, were increased by each agent. Fluoride and phenytoin together produced bigger increases in each parameter than did each agent alone. Trabecular bone volume was increased in the tibial metaphysis by fluoride or phenytoin alone; and when administered together, the two agents produced a greater increase. The combined effect of fluoride and phenytoin on each serum and bone formation parameter appeared to be less than additive. Phenytoin or fluoride alone did not significantly reduce the metaphyseal % resorptive surface. However, treatments with both agents together caused a highly significant reduction in the metaphyseal % resorptive surface. Phenytoin and fluoride together also significantly reduced (by 36%) the mineralization lag time, indicating that these agents did not promote osteomalacia. In summary, fluoride and phenytoin act in concert to stimulate bone formation and increase trabecular bone volume without causing mineralization defects in vivo and thus, may be a potential combination therapy for low bone mass in osteoporosis.

Key words: Bone formation; Fluoride; Osteoporosis; Phenytoin.

Reprints: K H W Lau, Jerry L Pettis Memorial Veterinary Medical Center, Mineral Metabolism Unit 151, 11201 Benton St, Loma Linda, California 92357 USA.
COMPARISON OF ALENDRONATE AND SODIUM FLUORIDE EFFECTS ON CANCELLOUS AND CORTICAL BONE IN MINIPIGS A ONE-YEAR STUDY
M H Lafage, R Balena, M A Battle, M Shea, J G Seedor, H Klein, W C Hayes and G A Rodan
West Point, Pennsylvania, USA

Abstract from *Journal of Clinical Investigation* 95 (5) 2127-2133 1995

Fluoride stimulates trabecular bone formation, whereas bisphosphonates reduce bone resorption and turnover. Fracture prevention has not been convincingly demonstrated for either treatment so far. We compared the effects of 1-year treatment of 9-month-old minipigs with sodium fluoride (NaF, 2 mg/kg/d p.o.) or alendronate (ALN, 4 amino-1-hydroxybutylidene bisphosphonate monosodium, 1 mg/kg/d p.o.) on the biomechanical and histomorphometric properties of pig bones. As expected, NaF increased and ALN decreased bone turnover, but in these normal animals neither changed mean bone volume. NaF reduced the strength of cancellous bone from the L4 vertebra, relative to control animals, and the stiffness (resistance to deformation) of the femora, relative to the ALN group. In the ALN-treated animals, there was a strong positive correlation between bone strength and L5 cancellous bone volume, but no such correlation was observed in the NaF group. Furthermore, the modulus (resistance to deformation of the tissue) was inversely related to NaF content and there was a relative decrease in bone strength above 0.25 mg NaF/g bone. Moreover, within the range of changes measured in this study, there was an inverse correlation between bone turnover, estimated as the percentage of osteoid surface, and modulus. These findings have relevant implications regarding the use of these agents for osteoporosis therapy.

Key words: Alendronate; Bone; Minipigs; Sodium Fluoride; Strength.

Reprints: G A Rodan, Merck Sharp and Dohme Ltd Research Laboratories, Department of Bone Biology and Osteoporosis, WP26-331, W Point, Pennsylvania 19486 USA.

IN VIVO EXPOSURE TO SODIUM FLUORIDE DOES NOT MODIFY THE YIELD OF VIRAL TUMOUR-INDUCED PERIOSTEAL BONE NOR OF HETEROTOPIC BONE INDUCED BY HUMAN TUMOUR KB CELLS IN MICE

P K Wlodarski, K H Wlodarski, K Galus, A C Skarpetowski, M Kowalski, M Luczak and M M Marciniak
Warsaw, Poland

Abstract from *Folia Biologica* 41 (2) 88-96 1995

In mice local or systemic administration of fluoride (5-25 mg NaF/kg body weight) during the proliferative phase of bone formation (up to 20 days) has no effect on the yield of bone formed either by local stimulation of periosteal membrane by Moloney sarcoma virus-induced tumour or on bone induced heterotopically by human KB cells. The lack of stimulatory activity of fluoride oil rapidly induced osteogenesis in mice is in agreement with recent reports which show that fluoride is not a potent mitogen for human osteoblasts grown in vitro.

Key words: Bone; Fluoride; Heterotopic; Periosteum; Tumours.

Reprints: K H Wlodarski. Medical Academy of Warsaw, Department of Histology and Embryology, Chalubinskiego 5, PL-02004 Warsaw, Poland.
THE DEMOGRAPHIC AND SOCIAL VARIATION IN THE PREVALENCE OF DENTAL ENAMEL OPACITIES IN NORTH WALES

R P Ellwood and D M O'Mullane
Wilton, Cork, Ireland

Abstract from Community Dental Health 11(4) 192-196 1994

The variation in the prevalence of developmental defects of enamel in areas of North Wales with low levels of fluoride in the drinking water was studied. Significant differences in the prevalence of diffuse opacities were found for the ten county districts in the study area. In addition, there was an association between area deprivation, as measured using the Townsend index, and diffuse opacity prevalence. The prevalence of subjects with one or more diffuse opacities was lower in more deprived electoral wards than those less deprived. For demarcated opacities and hypoplastic defects in the ten county districts, the prevalences were not significantly different and there was no association with area deprivation.

Key words: Enamel; Epidemiology; Fluoridation; Social class; Wales.
Reprints: R P Ellwood, Dental Health Unit, Unit 3a, Skelton House, Manchester Science Park, Lloyd Street North, Manchester M15 6SH England.

FLUOROSIS: FOCUS ON CHEMICAL AND BIOCHEMICAL ASPECTS

T Aoba, Tokyo, Japan

Abstract from Advances in Dental Research 8 (1) 66-74 1994

In order to assess fluoride accumulation and effects in developing dental tissues, one must determine the concentration profile of fluoride in the tissue and to assess separately the labile (ie, free ions in fluid and ions associated with organic matter) and stable (ie, incorporated into apatite lattice) pools of fluoride. Free fluoride ions in the mineralizing milieu markedly affect the driving force for precipitation and, as a result, the nature of precipitating crystals. The fluoride incorporated into the crystalline lattice increases the stability of the formed mineral. Improvement in the understanding of the mechanism of dental fluorosis requires more comprehensive information about the effects of fluoride on the ionic composition of the fluid phase, the nature of the initially precipitating mineral(s), the interactions between crystals and matrix proteins, and the enzymatic degradation of the proteins. Recent observations relevant to the role of fluoride in enamel formation include: 1) that there are threshold concentrations of fluoride below which the precipitation and hydrolysis of thin-platy octacalcium phosphate is facilitated but beyond which de novo apatite precipitation prevails; 2) that the presence of fluoride in the mineralizing milieu most likely affects the steady-state concentrations of mineral lattice ions; 3) that incorporation of fluoride into the stable pool is retarded by the presence of matrix proteins, particularly amelogenins, which inhibit the growth of apatite crystals; 4) that increasing the degree of fluoridation of apatite crystals enhances the adsorption of amelogenins onto the crystal surface, and 5) that amelogenins pre-adsorbed onto apatite crystals are more resistant to enzymatic cleavages by trypsin (used as a prototype of amelogeninases).

Key words: Enamel; Fluoride; Fluorosis; Metabolism.
Reprints: T Aoba, The Nippon Dental University, Department of Pathology, 1-9-2-Fujimi, Chiyoda-ku, Tokyo, Japan.
DENTAL FLUOROSIS: ITS USE AS A BIOMARKER

P K Den Besten
Rochester, New York, USA

Abstract from Advances in Dental Research 8 (1) 105-110 1994

Several epidemiological studies, beginning with those of Dean and co-workers in the 1940s, clearly demonstrate the relationship between dental fluorosis in humans and the level of fluoride in water supplies. These studies and others have shown that, in a population, there is a direct relationship among the degree of enamel fluorosis, plasma and bone fluoride levels, and the concentration of fluoride in drinking water. However, dental fluorosis is a reflection of fluoride exposure only during the time of enamel formation, somewhat limiting its use as a biomarker. In addition, the degree of fluorosis is dependent not only on the total fluoride dose, but also on the timing and duration of fluoride exposure. At the level of an individual response to fluoride exposure, factors such as body weight, activity level, nutritional factors, and the rate of skeletal growth and remodeling are also important. These variables, along with an individual variability in response to similar doses of fluoride, indicate that enamel fluorosis cannot be used as a biological marker of the level of fluoride exposure for an individual.

Key words: Biomarker; Enamel; Fluorosis.
Reprints: P K Den Besten, Department of Pediatric Dentistry, Eastman Dental Center, 625 Elmwood Ave, Rochester, New York 14620 USA.

FLUORIDE-INDUCED EARLY TEETH WEARING IN ARGENTINIAN CATTLE

T A Lopez, M R Busetti, M C Fort and D O Bedotti
Buenos Aires, Argentina

Abstract from Biomedical and Environmental Sciences 7 (3) 205-215 1994

The observed relationship between early teeth wearing in cattle and the existence of large areas with excess fluoride in underground water prevails in Argentina, but has never been studied. Average values of bone fluoride exceeding 5000 ppm and well water containing up to 10.5 ppm of this element were found in a farm (ETWF) where early teeth wearing is observed, while in a control farm (NETWF), those values were 1480 ppm and 3.0 ppm respectively. Urine samples from animals from the ETWF contained almost 4 times more fluoride than those from the latter. Forage in both farms contained about the same levels of fluoride (15 to 25 ppm). None of the farms was exposed to industrial, volcanic, or geothermal activity. Lactating and adult animals were transferred from one farm to the other. After 30 months, adult animals from the NETWF showed no damage in teeth, while adults from the ETWF continued a rapid teeth wearing. Young animals from the NETWF developed severe-ly damaged permanent teeth, while young animals from the ETWF developed normal teeth. We concluded that the problem exists in the ETWF and being a standard farm, the condition could be extrapolated to other areas.

Key words: Argentina; Cattle; Fluoride; Teeth wearing; Tooth demineralization.
Reprints: T A Lopez, Laboratorio de Toxicologia Veterinaria-Estacion Experimental Agropecuaria Balcarce (INTA), Balcarce, Buenos Aires, Argentina.
INFLUENCE OF EXPOSURE TO VARIOUS FLUORIDE TECHNOLOGIES ON THE PREVALENCE OF DENTAL FLUOROSIS

D C Clark, H J Hann, M F Williamson and J Berkowitz
Vancouver, British Columbia, Canada

Abstract from Community Dentistry and Oral Epidemiology 22 (6) 461-464 1994

An increase in the prevalence of dental fluorosis among children in North America is well documented. Published reports of the relationship between the occurrence of dental fluorosis and early exposure to various fluorides and the use of different types of infant feeding practices have begun to provide insights into possible causes for this increase. This study was designed to investigate this issue for children living in a non-fluoridated and a fluoridated community in British Columbia, Canada. Parents or guardians completed a questionnaire which detailed exposure to different types of fluorides and infant feeding practices during the first 6 yr of life. Completed questionnaires were returned and examinations were performed on 1131 children. 60% of children had dental fluorosis, and only 8% presented with scores of 2 or greater. Logistic regression analyses showed that the use of infant formula and parental educational attainment were significantly associated with the occurrence of dental fluorosis in the range of scores from 2 to 6. Despite these statistically significant findings, these variables actually had little additional predictive value beyond a chance occurrence in determining which children would have dental fluorosis.

Key words: Dental fluorosis; Fluoride ingestion; Public Health.
Reprints: D C Clark, Department of Clinical Dental Sciences, Faculty of Dentistry, University of British Columbia, 2199 Wesbrook Mall, Vancouver BC V6T 1Z3 Canada.

STRATEGIES FOR IMPROVING THE ASSESSMENT OF DENTAL FLUOROSIS: FOCUS ON OPTICAL TECHNIQUES

B Angmar-Månsson, E de Josselin-de Jong, F Sundström and J J ten Bosch
Huddinge, Sweden

Abstract from Advances in Dental Research 8 (1) 75-79 1994

In its milder forms, enamel fluorosis is characterized clinically by diffuse opacities. The appearance is due to optical properties of a subsurface or surface porous layer with lower mineral content. These areas usually have texture and color similar to those of initial caries lesions but generally another shape and location. Therefore, several optical methods, previously used to diagnose initial caries lesions, were applied to fluoride-induced opacities on extracted premolars and on incisors of four subjects in vivo. These methods included light-scattering measurements, white light illumination, violet light illumination, ultraviolet illumination, and laser fluorescence. Video images were captured with a charge-coupled-device (CCD) camera, digitized, and computer-processed. It is concluded that the light-scattering monitor can be used for the determination of the local porosity of fluorotic enamel and that the laser fluorescence method might be developed into a method applicable for the assessment of the severity of enamel fluorosis.

Key words: Enamel; Fluorosis; Fluorescence; Lasers.
Reprints: B Angmar-Månsson, Department of Cariology, School of Dentistry, Karolinska Institute, Box 4064, S-14104 Huddinge, Sweden.
ALKALI-SOLUBLE AND INSOLUBLE FLUORIDE IN Erupted AND UNERUPTED HUMAN ENAMEL FROM A HIGH FLUORIDE AREA WITH A LOW FLUOROSIS SCORE
S R Grobler, J F van Zyl, I Stander and T J V W Kotze
Tygerberg, South Africa
Abstract from Archives of Oral Biology 39 (8) 679-684 1994

The amounts of fluoroapatite and ‘CaF₂-like’ fluoride (F) were determined in enamel of unerupted and erupted teeth that had been exposed in vivo to 1.8-2.6 parts/10⁶ F in the drinking water and to brushing with F dentifrice at least once a day, and occasionally to a F mouth-rinse (0.022% F). Enamel was sampled by acid-etching and the F levels were measured with an adapted F ion-selective electrode. More F was built into the deeper enamel in the high-F area than in a similar low-F area. Unerupted enamel did not etch significantly (p > 0.05) deeper than erupted enamel. No significant differences (p > 0.05) were found in the F concentrations amongst the following: alkali-washed erupted, unwashed erupted, alkali-washed unerupted and unwashed unerupted at the outer most enamel (approx. 6 microns). However the erupted enamel (alkali-washed or not) showed higher F levels than unerupted enamel (alkali-washed or not) between approx. 6 microns and greater than 100 microns. The increase of F for this high-F area was about 100% in the deeper enamel while for a low-F area it was approx. 78% in the most outer enamel with no increase after a depth of about 20 microns. In contrast to a similar low-F area (water F < 0.10 parts/10⁶), no significant observed ‘CaF₂-like’ F could be detected in erupted or unerupted enamel for the high-F area.

Key words: Dentifrice; Enamel; Fluoride levels; Fluorosis; Water.
Reprints: S R Grobler, Oral and Dental Research Institute, Faculty of Dentistry, University of Stellenbosch, Private Bag X1, Tygerberg 7505, South Africa.

FLUORIDE PROFILES IN DIFFERENT SURFACES OF HUMAN PERMANENT MOLAR ENAMELS FROM A NATURALLY FLUORIDATED AND A NON-FLUORIDATED AREA
J Li, H Nakagaki, S Tsuboi, S Kato, S Huang, M Mukai, C Robinson and M Strong
Aichi-Gakuin, Japan
Abstract from Archives of Oral Biology 39 (8) 727-731 1994

The surfaces, sites and depths of fluoride profiles of enamel from a naturally fluoridated area (West Hartlepool, WHP, 1.9 ppm F in drinking water) and a non-fluoridated area (Leeds, LDS, < 0.1 ppm F in drinking water) were examined in detail. Using a regression curve, y = ax-b, comparisons of fluoride concentrations were made at depths of 1, 3, 5, 10, 20 and 30 microns at occlusal, middle and cervical sites of buccal, lingual, mesial and distal molar enamel surfaces. In the outermost enamels (<10 μ in depth), fluoride concentrations tended to be lower in the occlusal sites than in the cervical sites. In interior enamels (>20 microns in depth), the fluoride concentrations of molars from WHP were statistically higher than those from LDS, while few significant differences were found in the outermost layers. In the mesio- and disto-occlusal sites of approximal surfaces, there were some significant differences in the exponential regression coefficients of fluoride profiles between the two groups of molars.

Key words: Enamel; Fluoridation; Fluoride; Tooth surface.
Reprints: J Li, Preventive Dentistry and Dental Public Health, School of Dentistry, Aichi-Gakuin University, 1-100 Kusumoto-cho, Chikusa-ku, Nagoya 464 Japan.
TIMING OF ERUPTION OF PERMANENT TEETH:
STANDARD FINNISH PATIENT DOCUMENTS
J I Virtanen, R S Bloigu and M A Larmas
Oulu, Finland

Abstract from Community Dentistry and Oral Epidemiology 22 (5 Pt 1) 286-288 1994

Current standards for the eruption of teeth are constructed mostly on the basis of cross-sectional data. The aim here was to analyze the suitability of the standard patient documents created for health center dental care purposes for the collection of longitudinal data on tooth emergence. Copies of the oral health records of the 911 children born in 1970 and 1971 and in 1980 and 1981 living in three rural communities in Finland were re-examined and analyzed using a specially compiled computer program. The means and standard deviations are in line with previous results. The sex difference in emergence timing ranged from 0.1 to 1.0 years for the various teeth. The emergence of the teeth of the second phase of the mixed dentition was later in the children living in an endemic fluoride area, this difference being statistically greater for the boys than for the girls (95% CI for differences between means was used to evaluate statistical significance). A secular trend in the eruption of permanent teeth was found between 1970 and 1980. Patient documents are shown to be suitable for the collection of longitudinal data on dental emergence.

Key words: Endemic fluoride; Eruption; Permanent teeth.
Reprints: Jorma Virtanen, Department of Preventive Dentistry and Cariology, University of Oulu, Finland.

FLUORIDE DISTRIBUTION OF RAT MOLAR CEMENTUM IN RELATION TO AGE AND FLUORIDE LEVELS IN THE DRINKING WATER
K Kondo, H Nakagaki, K Kato, N Narita, T Ito, T Kanayama and C Robinson
Nagoya, Japan

Abstract from Caries Research 29 (3) 218-222 1995

The present study was undertaken to determine the fluoride distribution profile in rat molar cementum with age in relation to fluoride in drinking water. Fifty-four female Wistar rats were used for the experiment. Before the experiment 6 rats were killed under chloroform anesthesia at 4 weeks of age as controls. The remaining 48 rats were divided into two groups: a control group given distilled water and the other group given water containing 100 ppm fluoride ad libitum. Six rats from each group were killed at the ages of 6, 12, 24, and 48 weeks. The fluoride distribution in the molar cementum was analyzed from the surface to the cementodentinal junction by abrasive microsampling. The fluoride concentrations in molar cementum from control rats on distilled water remained relatively constant until 24 weeks. A small increase then occurred between 24 and 48 weeks. The fluoride concentration in cementum from rats drinking water containing 100 ppm fluoride increased markedly with age, both in outer and inner regions of the molar cementum for all rats. On the other hand, the fluoride concentration in the cementum of older rats drinking water with 100 ppm fluoride was significantly higher in the outer than in the inner region of the cementum. It was concluded that the fluoride distribution in rat molar cementum may increase throughout life in relation to the fluoride level in the drinking water.

Key words: Age, Cementum; Drinking water fluoride; Fluoride distribution.
Reprints: K Kondo, Aichi Gakuin University School of Dentistry, Chikusa Ku, 1-100 Kusumoto CHO, Nagoya, Aichi 464, Japan.
BIOMINERALIZATION DURING EARLY STAGES OF THE DEVELOPING TOOTH IN VITRO WITH SPECIAL REFERENCE TO SECRETORY STAGE OF AMELOGENESIS

J H M Woltgens, D M Lyaruu, A L J J Bronckers, T J M Bervoets and M Vanduin
Amsterdam, The Netherlands

Abstract from International Journal of Developmental Biology 39 (1) 203-212 1995

In this survey we summarize data on mineralization of enamel mostly obtained in organ culture experiments in our laboratory. Historically, the enzyme alkaline phosphatase has been proposed to stimulate mineralization by supplying phosphate or by splitting away inorganic pyrophosphate PPI, a potent inhibitor of mineralization. Localization of alkaline phosphatase in developing teeth by enzyme histochemistry shows that cells of the stratum intermedium contain extremely high levels of alkaline phosphatase but secretory ameloblasts that are engaged in deposition of the matrix and in transport of mineral ions lack alkaline phosphatase. The function therefore must be an indirect one, since no activity was seen at the site of enamel mineralization. We propose that the main function of alkaline phosphatase in the stratum intermedium is to transport phosphate or nutrients from blood vessels near the stratum intermedium into the enamel organ. Another function of the enzyme in stages of cell differentiation was deduced from inhibition experiments with the specific alkaline phosphatase inhibitor I-pBTM, showing that in tooth organ culture the enzyme may be involved in the generation of phosphorylated macromolecules from P ions originating from pyrophosphate. Calcium plays an indispensable role in enamel mineralization in vitro. Low calcium concentration in the culture medium prevented initial dentin mineralization and enamel formation. Moreover, differentiating ameloblasts did not become secretory, in contrast to odontoblasts that secreted a layer of predentin matrix. Variations in phosphate concentration in the culture medium do not seem to affect tooth organ cultures adversely during mineralization in vitro. Exposure to F, however, has adverse effects on enamel mineralization depending on concentration and exposure time and produces a variety of disturbances. Many of the fluoride-induced changes in the enamel organ are reversible: young ameloblasts recover and resume secretion and mineralization of the fluorotic matrix when fluoride is removed from the medium. This recovery is enhanced when medium calcium levels are increased. Only the changes in the hypermineralized enamel remain irreversible. Thus, we hypothesize that fluoride induces a local hypocalcemia in the enamel fluid surrounding the enamel crystals by stimulating a hypermineralization of the pre-existing enamel crystals.

Key words: Alkaline phosphatase; Amelogenesis; Biomineralization; Calcium; Developing teeth; Fluoride.

Reprints: J H M Woltgens, Free University of Amsterdam, Department of Oral Cell Biology, Van der Boechorststr 7, 1081 BT Amsterdam, The Netherlands.
FLUOROSIS IN A WILD COTTON RAT (*SIGMODON HISPIDUS*) POPULATION INHABITING A PETROCHEMICAL WASTE SITE

M G Paranjpe, A M S Chandra, C W Qualls, S T Mcmurry, M D Rohrer, M M Whaley, R L Lochmiller and K Mcbee
Stillwater, Oklahoma, USA

Abstract from *Toxicologic Pathology* 22 6 569-578 1994

We have developed an *in situ* mammalian model for evaluating environmental contamination using wild cotton rats. In a series of experiments, 200 male cotton rats were captured during 4 collection periods (spring 1991 = 35; fall 1991 = 60; spring 1992 = 53; fall 1992 = 52). A total of 103 of these cotton rats were captured from control sites, and the remaining 97 were captured from an abandoned oil refinery. All sites were located in the vicinity of Cyril, Oklahoma. There were alterations in the incisors of cotton rats captured from the refinery site. Normal color of cotton rat incisors is deep yellow-orange, which is imparted by a pigment normally produced by ameloblasts. Grossly, the upper incisors of 37 of 97 rats and lower incisors of 54 of 97 rats were affected. The affected incisors were white, chalky, and thin with striations and erosions of the enamel. Microscopic examination revealed that there were dysplastic and necrotic changes in the ameloblasts. The bone fluoride levels were significantly higher in rats captured from the refinery as compared to the rats captured from the control sites.

Key words: Ameloblasts; Enamel; Environmental contamination; Rodents.
Reprints: C W Qualls, Oklahoma State University, College of Veterinary Medicine, Department of Veterinary Pathology, Stillwater, Oklahoma 74078 USA.

FORMATION AND STRUCTURE OF DENTINE IN THE RAT INCISOR AFTER CHRONIC EXPOSURE TO SODIUM FLUORIDE

J Appleton
Liverpool, England

Abstract from *Scanning Microscopy* 8 (3) 711-719 1994

Weanling rats were chronically exposed to sodium fluoride by feeding them for eight weeks with a standard powdered diet incorporating sodium fluoride at 0.025%, 0.05% and 0.1% by weight. After eight weeks blood was removed by cardiac puncture and the levels of plasma calcium and phosphate determined. The incisor teeth were removed and their structure examined in the scanning electron microscope (SEM) using backscattered electron imaging to determine the relative concentration and distribution of the mineral phase in the dentine. There was no significant alteration to the normal serum calcium and phosphate levels. Small scattered inter-globular spaces were seen in the incisor dentine of rats consuming the 0.025% sodium fluoride diet. The principle feature with the 0.05% diet were marked striations in the circum-pulpal dentine but with the 0.1% diet there was severe disruption of dentine structure with continuous bands of interglobular spaces. These spaces were larger labially than lingually reflecting differences in the size and shape of calciospherites. The distribution of interglobular dentine would suggest that its formation takes time to establish.

Key words: Dentine; Dentine formation; Mineralisation; Sodium Fluoride; Rat incisor.
Reprints: J Appleton, University of Liverpool School of Dentistry, Department of Clinical Dental Science, Liverpool L69 3BX, Merseyside, England.
THE INFLUENCE OF FLUORIDE ON THE ADSORPTION OF PROTEOGLYCANS AND GLYCOSAMINOGLYCANS TO HYDROXYAPATITE

R Hall, G Embry, R Waddington and A Gilmour
Cardiff, Wales

Abstract from *Calcified Tissue International* 56 (3) 236-239 1995

Proteoglycans and their spatial arms, the glycosaminoglycans, are known to interact with hydroxyapatite, and are considered to have a role in the regulation of mineralization. This study investigates the interactive mechanisms, with particular attention directed at the influence of fluoride on the adsorption process. Proteoglycans and glycosaminoglycans were adsorbed to hydroxyapatite in the presence of fluoride (1-20 ppm range). The adsorbates included a chondroitin 4-sulfate-rich proteoglycan extracted from rat incisor dentine, hyaluronan, chondroitin 4-sulfate, and dermatan sulfate. The order of glycosaminoglycan in decreasing affinity for hydroxyapatite was chondroitin 4-sulfate, dermatan sulfate, and hyaluronan, and the individual glycosaminoglycans showed different responses to the presence of fluoride. Graded increases in fluoride (1-4 ppm) led to 540% reduction of glycosaminoglycan adsorption to hydroxyapatite. The proteoglycan showed less affinity for hydroxyapatite, and demonstrated a reduction in adsorption of up to 22% with 20 ppm fluoride. The inhibitory effect of fluoride indicated an electrostatic mechanism, presumably via the calcium sites in the hydroxyapatite lattice.

Key words: Biomineralization; Dentine; Fluoride; Glycosaminoglycans; Proteoglycan.

Reprints: R Hall, University Wales College of Medicine School of Dentistry, Department of Basic Dental Science, Heath Park, Cardiff CF4 4XY, Wales.

THE AVERAGE DAILY DOSE OF FLUORIDE:
A MODEL BASED ON FLUID CONSUMPTION.

J D Shulman, J A Lalumandier and J D Grabenstein
Dallas, Texas, USA

Abstract from *Pediatric Dentistry* 17 13-18 1995

The quantity of fluoride needed to prevent caries but avoid dental fluorosis is unknown. To estimate the desired daily dose of fluoride, we analyzed fluid consumption data from a stratified random sample of 7,345 children studied during the 1977-78 US Department of Agriculture Nationwide Food Consumption Survey and applied it to Dean’s observations of optimally fluoridated communities in the 1940s. The average daily fluoride intake per kilogram body weight from optimally fluoridated tap water was highest (0.080 mg/kg/d) from 7 to 9 months of age, and declined linearly to 0.034 mg/kg/d at 12.5 to 13 years of age. The mean was 0.068 ± 0.008 mg/kg/d from birth to age 7 years, and 0.042 ± 0.006 mg/kg/d from age 7 to 13 years. The American Academy of Pediatrics supplementation schedule delivers fluoride dosage rates that are below our findings of the average daily dose of fluoride after the third month of life, although the two curves are within 0.006-0.013 mg/kg/d from 3 months to 16 years of age. While supplementation alone does not exceed the average daily dose of fluoride, the cumulative effects of fluoride from tap water, processed foods, ingested toothpaste, and dental treatments after the third birthday should be evaluated for their role in the increased prevalence of fluorosis.

Key words: Beverages; Child; Dental caries; Diet; Dose; Fluoride.

Reprints: J D Shulman, Department of Public Health Sciences, Baylor College of Dentistry, Dallas, Texas, USA.
DENTAL CARIES EXPERIENCE AND DEFECTS OF DENTAL ENAMEL AMONG 12-YEAR-OLD CHILDREN IN NORTH LONDON, EDINBURGH, GLASGOW AND DUBLIN
M C Downer, A S Blinkhorn, R D Holt, C Wight and D Attwood
London, England

Abstract from Community Dentistry and Oral Epidemiology 22 (5 Pt 1) 283-285 1994

A multi-center study of caries experience and defects of dental enamel was conducted among 12-yr-old children in north London, Edinburgh, Glasgow and Dublin. None of the cities had water fluoridation except Dublin, which was included in the national program introduced in the Republic of Ireland in 1964. A random sample of children was drawn from state schools in each location and identical methods of clinical examination were used throughout under the same standardized conditions. All examiners were trained and calibrated with a reference examiner and achieved high levels of inter- and intra-examiner consistency. Mean DMFT values for the 4 cities were 1.27 (London), 1.39 (Edinburgh), 2.70 (Glasgow) and 1.48 (Dublin) (P < 0.001). Proportions of subjects free from caries in the same order were, 50, 47, 24 and 43% (P < 0.001), and child prevalence of diffuse opacities, 28, 29, 7 and 17% (P < 0.001), respectively. The relatively low caries levels recorded in London and Edinburgh (lower than Dublin) were considered to be related most probably to fluoride effects other than water fluoridation.

Key words: Defects of dental enamel; Dental caries; Epidemiology; Fluoride.
Reprints: M C Downer, Department of Dental Health Policy, Institute of Dental Surgery, Eastman Dental Hospital, 256 Gray’s Inn Road, London WC1X 8LD England.

DIETARY INGESTION OF FLUORIDE AND CARIES PREVALENCE IN PRESCHOOL AND SCHOOL CHILDREN IN CITIES WITH DIFFERENT FLUORIDE CONTENT IN THE DRINKING WATER AND DIET
P Cisternas, S Guerrero, A Morales and R Uauy
Santiago, Chile

Abstract from Revista Medica De Chile 122 (4) 459-464 1994

The consumption of dietary fluoride and its relationship with dental caries prevalence was determined in 780 preschool children and 802 school children from Quique, Arica, Santiago and San Antonio, where drinking water has different fluoride concentration. Dietary fluoride was determined from dietary records and fluoride content of foods. The fluoride consumption from drinking water was calculated by Galagan and Vermillion formula. The total fluoride consumption was obtained. The dental caries prevalence was established by the evaluation of dental caries index in deciduous (dmft) and permanent (DMFT) dentition. Significantly lower DMFT values were founded in cities with lower fluoride levels in drinking water. The exception was San Antonio, where high dietary fluoride explained the lower DMFT values relative to Santiago. Our results suggest that not only fluoride in water but also the consumption of fluoride rich foods has a significant impact on the prevalence of caries.

Key words: Child; Chile; Dental caries; Diet; Fluoride.
Reprints: P Cisternas, Instituto de Nutricion y Tecnologia de los Alimentos (INTA), Universidad de Chile, Santiago, Chile.
STUDIES ON THE CARIOSTATIC MECHANISMS OF FLUORIDE
R P Shellis and R M Duckworth
Bristol, England

Abstract from International Dental Journal 44 (3 Suppl 1) 263-273 1994

This paper reviews mechanisms underlying the caries-preventive effects of fluoride, with special reference to factors which govern the efficacy of topical treatments. Fluoride reduces demineralisation in several ways: by reducing bacterial acid production and acidurance; by reducing the equilibrium solubility of apatite; and by the fluoridation of apatite crystal surfaces, reducing the dissolution rate, whether or not there is reduced solubility of the bulk mineral. On available evidence, the last seems to be the most important effect. The effect depends upon the presence of sufficiently high, dissolved fluoride concentrations to maintain the surface fluoridation. The provision of dissolved fluoride is the key to successful therapy, especially topical treatments. Fluoride also promotes remineralisation even at low concentrations, thus slowing or preventing overall mineral loss. The formation of intra-oral fluoride reservoirs capable of supplying ions for a prolonged period is crucial to the success of topical treatments. Such reservoirs include calcium fluoride, formed mainly at tooth surfaces, and fluoride associated with organic components of plaque and oral soft tissues. The patterns of fluoride clearance from intraoral reservoirs are discussed. Fluoride may be present in dentifrices as the fluoride ion ($F^-$) or as the monofluorophosphate ion ($FPO_3^{2-}$). It is concluded that the efficacy of $FPO_3^{2-}$ probably depends on enzymic hydrolysis to $F^-$. Monofluorophosphate appears to be retained less well in intra-oral fluoride reservoirs and reasons for this are discussed.

Key words: Cariostatic mechanisms; Dental caries; Fluoride.
Reprints: R P Shellis, MRC Dental Group, Dental Hospital and School, Lower Maudlin Street, Bristol BS1 2LY England.

THE RELATIVE CONTRIBUTION OF DENTAL SERVICES TO THE CHANGES AND GEOGRAPHICAL VARIATIONS IN CARIES STATUS OF 5- AND 12-YEAR-OLD CHILDREN IN ENGLAND AND WALES IN THE 1980S
P Nadanovsky and A Sheiham
London, England

Abstract from Community Dental Health 11 (4) 215-223

The aim of this study was to assess the relative contribution of dental services to the changes and geographical variations in caries status of 5- and 12-year-old children in England and Wales in the 1980s. A secondary aim was to assess the association between caries experience and social factors. An ecological study analysing data at a district health authority level in England and Wales was conducted. Twenty-eight, 43 and 34 per cent of the variations in 5-year-old dmft in 1985, 1987 and 1989 respectively were explained by dental service activity indicators and 53, 62 and 57 per cent by social factors. Twenty-nine per cent of the variation in 12-year-old DMFT in 1988 was explained by dental service activity indicators and 46 per cent by social factors.

Key words: Dental caries; Dental services; Social factors; Trends.
Reprints: P Nadanovsky, Department of Epidemiology and Public Health, University College London and London Hospital Medical College, Gower Street, London, England.
THE DIFFUSION AND ENZYMIC HYDROLYSIS OF MONOFLUOROPHOSPHATE IN DENTAL PLAQUE

E I F Pearce and G H Dibdin
Wellington, New Zealand

Abstract from Journal of Dental Research 74 (2) 691-697 1995

Although the ability of dental plaque to hydrolyze sodium monofluorophosphate (MFP) has been known for some time, its effect on the F\(^{-}\) concentration at the plaque-enamel interface is undefined. We have determined enzyme kinetic values for MFP hydrolysis and diffusion coefficients so that the penetration and degradation of MFP in plaque can be modeled by computer. The K-M and V-max Values for natural human plaque were 1.77 mmol/L and 41.4 nmol/min/mg protein, respectively, at pH 8.0. At pH 6.0, the V-max was lower, 15.6 nmol/min/mg, but K-M was not significantly different. Competitive inhibition by orthophosphate gave a K-i of 4.55 mmol/L. The diffusion coefficient for MFP in artificial plaque was \(1.91 \times 10^{-6}\) cm\(^2\)/sec. When these data were used for mathematical modeling of the effects of rinsing with MFP and F\(^{-}\) solutions, compared with an equivalent NaF application, the concentration of F\(^{-}\) from MFP was lower at the inner surface of plaque, acid the peak occurred later. Both pH and plaque thickness had a marked effect on the amount of MFP that could penetrate: At pH 8.0, almost none reached the inner surface of a 1-mm-thick plaque intact. At pH 6.0, however, more MFP was able to penetrate, due to lower MFPase activity. While MFP diffusion is inherently slower than that of F\(^{-}\), enzymic degradation increases the gradient for inward diffusion. If the conventional view that MFP in toothpaste acts as a source of F is true, then MFP toothpaste should be formulated to optimize MFPase activity in dental plaque.

Key words: Dental plaque; Diffusion; Hydrolysis; Monofluorophosphate.

STOICHIOMETRY OF FLUORIDE RELEASE FROM FLUORHYDOXYAPATITE DURING ACID DISSOLUTION

E I F Pearce, N Guhachowdhury, Y Iwarni and T W Cutress
Wellington, New Zealand

Abstract from Caries Research 29 2 130-136 1995

Release off from fluorhydroxyapatite (FHAp) during acid dissolution was studied to validate the use of this mineral as a plaque reservoir of F. FHAp minerals having a wide range of F concentrations were synthesised by aqueous precipitation, and samples repeatedly exposed to 50 mM lactic acid solution, pH 4.5, or similar lactic/acetic/formic acid mixtures, until dissolution was complete. While the Ca/P ratio in solution remained relatively constant and close to the ratio in the solid, the solution F/Ca ratio invariably changed during dissolution. During initial stages the F/Ca solution ratio was lower than in the solid but rose to reach a plateau higher than in the solid as dissolution progressed, an effect that was more pronounced with low-F FHAp. With these minerals the plateau F/Ca level never reached 0.2, suggesting that a F-enriched FHAp rather than pure fluorapatite precipitates during dissolution. It is concluded that a high-F FHAp mineral would best serve as an apatitic plaque reservoir.

Key words: Dissolution; Fluorhydroxyapatite; Fluoride.

Reprints (both above): E I F Pearce, Health Research Council of New Zealand, Dental Research Unit, POB 27007, Wellington, New Zealand.
ANTICARIES EFFECT OF DIFFERENT AMINE FLUORIDE CONCENTRATIONS IN SCHOOLCHILDREN

K Rosingrget and I Lincir
Zagreb, Croatia

Abstract from Caries Research 29 (3) 168-171 1995

A double-blind 3-year clinical trial was conducted in a nonfluoridated water community in which 354 schoolchildren aged 9-10 years were assigned randomly to three groups (A, B and C) and subjected to a treatment with 1 of 3 different concentrations of amine fluoride solution every 2 months. Clinical examinations were conducted at baseline and after 1, 2, and 3 years. For group A (positive control) receiving topical applications of amine fluoride solution (1%), the 3-year DMFT and DMFS increments were 4.0 ± (SD) 2.7 and 8.5 ± 5.6, respectively. For group B (0.5% F), the DMFT and DMFS increments were 4.4 ± 3.0 and 8.4 ± 5.7, respectively; these differences being statistically not significant. The mean caries increments in group C (0.25% F) were 5.1 ± 2.9 for DMFT and 10.0 ± 5.5 for DMFS. The differences in incremental caries scores between group C and groups A or B were statistically significant (p < 0.05). Thus, the amine fluoride solution, in which the fluoride content was reduced by half, provided similar caries reductions in schoolchildren as did a standard amine fluoride solution with 1.0% F.

Key words: Amine fluoride solution, Dental caries; Topical application.

Reprints: K Rosingrget, University of Zagreb Faculty of Dentistry, Salata 11, Zagreb 41000, Croatia.

UPTAKE AND RELEASE OF FLUORIDE FROM BIRCH AND LIME TOOTHPICKS

H Kashani, D Birkhed and L G Petersson
Gothenburg, Sweden

Abstract from European Journal of Oral Sciences 103 (2 Pt 1) 112-115 1995

Birch and lime toothpicks were impregnated in a 4% sodium fluoride (NaF) solution for different periods of time, viz. 30 s, 30 min and 3 d. After impregnation for 30 min, an average of 4.1 mg NaF per birch toothpick was taken up. The corresponding value for lime toothpicks was 5.1 mg. About 60% of the fluoride (F) was released within 1 h in vitro when the toothpicks were kept in water. In vivo, the concentration of F was determined in whole saliva from five adults during 30 min after using birch toothpicks immersed in a 4% NaF solution. After 2 min, the mean salivary F concentration was 2.7 mM. Impregnation of birch toothpicks in 1, 2 or 3% NaF resulted in lower F concentrations both in vitro and in vivo, with a clear dose-response effect. When comparing the use of a 4% NaF impregnated birch toothpick, a mouthrinse with 10 mi of 0.025% NaF, sucking on a F tablet containing 0.55 mg NaF, and toothbrushing with 1 g of an 0.068% F (as NaF) dentifrice - all procedures carried out in the mouth during 2 min - the highest concentration of F in saliva was obtained after using the fluoridated toothpick. Thus, NaF impregnated birch and lime toothpicks show a quick release of F in vitro as well as in vivo and may be suitable as home care products for prevention of dental caries.

Key words: Fluoride; Saliva; Toothpicks.

Reprints: D Birkhed, Gothenburg University Faculty of Odontology, Department of Cariology, Medicinaregatan 12, S-41390 Gothenburg, Sweden.
THE PREVALENCE AND RISK FACTORS OF FLUOROSIS AMONG
PATIENTS IN A PEDIATRIC DENTAL PRACTICE
J A Lalumandier and R G Rozier
Columbus, Ohio, USA

Abstract from Pediatric Dentistry 17 (1) 19-25 1995

Seven hundred eight patients aged 5-19 years in a pediatric practice in North Carolina were selected using a random-start, systematic sampling procedure and enrolled in a case control study to determine risks for fluorosis. Subjects were examined by four trained examiners using the Tooth Surface Index of Fluorosis (TSIF). Information on fluoride exposures and other explanatory variables was obtained through parent interviews and mail questionnaires. Fluoride exposure was confirmed by fluoride assay of community drinking water samples. Bivariate associations for the entire sample were tested using MH chi2 statistic. A secondary analysis controlling for fluoride in drinking water was performed using logistic regression for 233 subjects (116 drinking fluoridated water; 117 drinking fluoride-deficient water) who were lifetime residents at the same address. Nearly 78% of subjects had a TSIF score of > 0; 36.3% > 1; and 18.9% > 2. Twenty-two variables found in bivariate analyses (P < 0.15) to be associated with fluorosis were included in multivariate analyses. For subjects drinking fluoride-deficient water, fluorosis (1 or more positive TSIF scores) was associated with dietary fluoride supplement frequency (OR = 6.5) and age of the child when brushing was initiated (OR = 3.0). For subjects drinking fluoridated water, fluorosis was associated with age of child when brushing was initiated (OR = 3.1).

Key words: Child; Enamel; Epidemiology; Fluorosis.
Reprints: J A Lalumandier. Bureau of Dental Health, Ohio Department of Health, Columbus, Ohio, USA.

A great amount of the voluminous literature on fluoride effects in dental journals is reiterative, and repetitive of assumptions based on earlier research claims. Value-laden terms abound (like "fluoride-deficient" in the above abstract). Often little original research is reported. In our Abstracts section we aim to present an overview of fluoride research within various scientific disciplines. However it is simply not possible to reproduce all the dental abstracts. The following is a list of some other recent articles. - Editor.


TRENDS IN CARIES PREVALENCE IN NORTH AMERICAN CHILDREN. B A Burt. *International Dental Journal* 44 (4 Suppl 1) 403-413 1994. Reprints: School of Public Health, University of Michigan, Ann Arbor, Michigan 48109-2029 USA.


CELEBRATING 50 YEARS OF FLUORIDATION: A PUBLIC HEALTH SUCCESS STORY. M W Easley. *British Dental Journal* 178 (2) 72-75 1995. Reprints: Department of Health Services, Kentucky, USA.

---

**FLUORIDE-INDUCED CHANGES IN THE ACTIVITIES OF SUCROSE METABOLIZING ENZYMES IN RELATION TO STARCH ACCUMULATION IN SORGHUM CARYOPSIS, RAISED THROUGH LIQUID CULTURE**

B Asthir and R Singh
Ludhiana, Punjab, India

Abstract from *Plant Physiology and Biochemistry* 33 (2) 219-223 1995

Effect of sodium fluoride (10-50 mM) on the activities of sucrose metabolizing enzymes and alkaline inorganic pyrophosphatase in relation to the transformation of free sugars to starch was studied by culturing detached ears of sorghum (*Sorghum bicolor*) in complete liquid medium for 4 and 7 days. Addition of fluoride to culture medium reduced the contents of dry weight, total free sugars and starch of the grains. This ion stimulated the activities of sucrose metabolizing enzymes *viz*, soluble acid-, neutral invertases (EC 3.2.1.26) and sucrose synthase (EC 2.4.1.13). However, the activity of sucrose phosphate synthase (EC 2.4.1.14) remained unaffected. The activity of alkaline inorganic pyrophosphatase (EC 3.6.1.1) decreased with fluoride and this was concomitant with an increase in the contents of inorganic pyrophosphate (PPI) and reducing sugars. Culturing detached spikelets in 10-50 mM fluoride and pretreating isolated endosperm in presence of 50 mM fluoride drastically reduced the incorporation of C-14 into starch from supplied [U-C-14] sucrose or glucose or fructose. Based on these results, we infer that low activity of alkaline inorganic pyrophosphatase by fluoride led to an increased accumulation of PPI in grain amyloplasts which in turn inhibited the production of starch.

Key words: Alkaline inorganic pyrophosphatase; Ear culturing; Invertase; Sucrose metabolizing enzymes; Sugars to starch transformation; *Sorghum bicolor*.

Reprints: R Singh, Punjab Agricultural University, Department of Biochemistry, Ludhiana 141004, Punjab, India.