BORON AS AN ANTIDOTE TO FLUOROSIS? PART I: STUDIES ON THE SKELETAL SYSTEM

by

J. Franke, H. Runge, R. Bech, W. Wiedner, W. Kramer, W. Kochmann, A. Hennig, H. Ludke, W. Seffner, W. Teubner, M. Franke, W. Moritz, L. Barthold, and D. Geinitz

Erfurt, Bitterfield, Halle, Jena, Leipzig, GDR

SUMMARY: The antidote action of boron and aluminum sulphate in chronic fluorine intoxication was tested on 8 groups of domestic pigs over a period of 13 months. To the diet of pigs, given 7 mg NaF/kg/d, 4 and 8 mg B/kg/d and 0.1 g of aluminum sulphate was added as an antidote. NaF feeding caused chronic bone fluorosis. Although boron did not lead to reduction in fluorine storage in the skeleton, it exerted a certain detoxicating effect due to formation of less toxic boron fluorine complexes. Respecting the skeletal system, direct F action (increase of bone mass) is, at least partially, compensated by direct boron action upon bone (decrease in bone mass with reduced parathyroid activity). Aluminum sulphate reduces F absorption and F retention in the skeleton by 25 to 29%; concurrent inhibition of calcium absorption from the intestine results in secondary hyperparathyroidism and decrease in bone mass.

According to these experiments, boron or aluminum sulphate are unsuitable prophylactically in humans chronically exposed to fluoride because of individual reaction to fluoride and because of the toxic action of boron and aluminum sulphate upon bone. Short-term administration of boron for more rapid detoxication in fluorosis cases may be permissible during exposure to fluoride and after exposure to the pollutant has been discontinued.

KEY WORDS: Aluminum; Antidotes; Boron; Experimental fluorosis; Pigs; Prophylaxis; Skeletal fluorosis; Sulphate; Therapy.

Introduction

Since 1973 the authors have been searching for an antidote to reduce or prevent the toxic effects of inorganic fluoride compounds or to complex or displace the fluorine ion at its place of action, either in the metabolic process or the skeletal system. Initially, antidotes were tested such as vitamin C, aluminum, magnesium, calcium, molybdenum, copper, iron, boron (B) vanadium, selenium, glutamine, cysteine and glycocorticoids on green algae Chlorella fusca var. vacuolata, human erythrocytes and albino mice.

Because in all three tests (1) iron and boron compounds seemed to be par-

Department of Orthopedic Surgery, Medical Academy, Erfurt, DDR 5010, Regierungsstrasse, 42A. Presented at the 13th conference of the International Society for Fluoride Research, Nov. 14-17, 1983, New Delhi, India.

ticularly effective they were more intensively explored on rabbits. F^- action on teeth and skeleton was clearly reduced by concurrent administration of borax. Iron compounds were ruled out because of unfavorable side effects (2,3).

In the first experiment in the current series 48 domestic pigs were given for 1 year, 0.5 mg and 5 mg NaF/kg body-weight and antidotes B (0.15 and 3.0 mg as borax/kg body-weight), serpentine (6.75 mg/kg body-weight) and Mg O (2.75 mg/kg body-weight). F action on the skeletal system was reduced by serpentine and, particularly, by high boron doses. The F content in bone ash, however, was not affected by the antidotes. Boron alone caused osteoporosis (4 and unpublished data). A new experiment with higher F and B doses was initiated to provide conclusive answers, especially with regard to the isolated effect of high doses of boron on the skeletal system.

Material and Methods

The experiments were performed on 68 castrated male domestic pigs (mean initial weight, 59 kg) over a period of 13 months. Twelve animals (group 1) served as controls. Groups 2 - 8 (8 animals each) received the following doses of NaF plus an antidote: Group 2: 7 mg NaF/kg; Group 3: 7 mg F/kg + 4 mg B^{+++}/kg as boric acid (H_3BO_3); Group 4: 7 mg F/kg + 8 mg B^{+++}/kg; Group 5: 4 mg B^{+++}/kg; Group 6: 8 mg B^{+++}/kg; Group 7: 7 mg F/kg + 100 mg Al-sulphate: $Al_2(SO_4)_3 \times 18\ H_2O$; Group 8: 7 mg F/kg + 100 mg Al-sulphate + 4 mg B^{+++}/kg.

Aluminum sulphate, an agent known to reduce fluorosis in grazing cattle (5), was additionally included. The F/B ratio was related to $BF_2(OH)_2$, with a calculated excess of boron over fluorine of 100 or, resp., 300%.

Fasting morning samples of blood and urine were taken the 5th, 10th 12th, and 13th month on the day of sacrifice. Only analyses dealing with the skeletal system are presented here. Bone mineral content was determined by ^{125}I-photon absorptiometry on the cleaned metacarpal bones 2 and 3 and the femur according to the Cameron and Sorensen method (6) in the bone center. X-ray photographs were taken of all 3 bones in two planes, and of the lumbar vertebral column. On the roentgenograms of metacarpals 2 and 3 and femur, the corticalis index was computed according to Exton-Smith et al. (7) (Fig. 1).

Figure I

$$\frac{\text{Cortical Area}}{\text{(bone) surface area}} = \text{Cortical Index}$$

According to Exton-Smith et al. (1969)

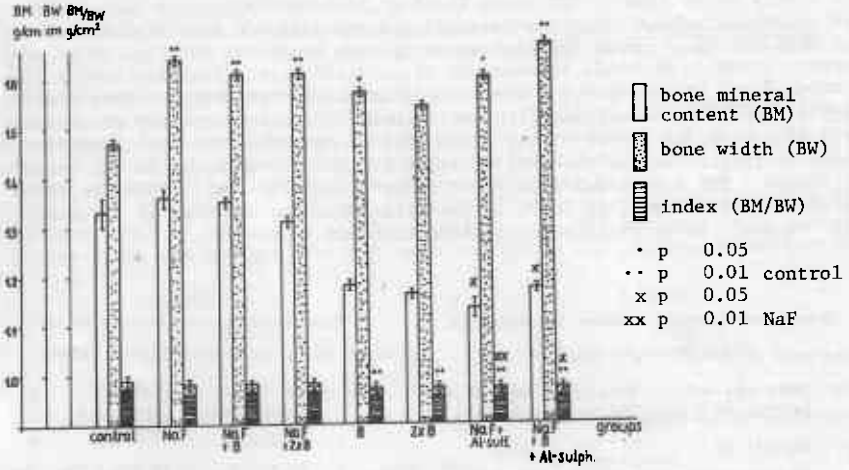
Pieces of bone, 1.5 - 2 cm long, which served for histological and histomorphometric studies, were sawed out of the right iliac crest (tuber coxae), alcohol-fixed, embedded in methacrylate and cut in non-decalcified condition. The sections were stained according to Goldner and with toluidine-blue. The spongiosa and osteoid volume were histomorphometrically determined by the point-counting method according to Chalkey (8). F in serum, urine, iliac crest and rib ash was determined by the ion selective electrode. For boron determination, bone samples were ashed in the presence of lithium carbonate: all boron was converted to $(BF_4)^-$ complexes and photometrically determined after enrichment by extraction. To determine parathyroid function, areas of 150 nuclei of the parathyroid were measured histomorphologically per animal. Increasing sizes of the nuclei indicate a stimulation of function.

Results

^{125}I photon absorptiometry (Fig. 2a and b): In group 2.the effect of fluoride was an increase in bone mineral content (BM) and bone width (BW) at

Figure 2 a

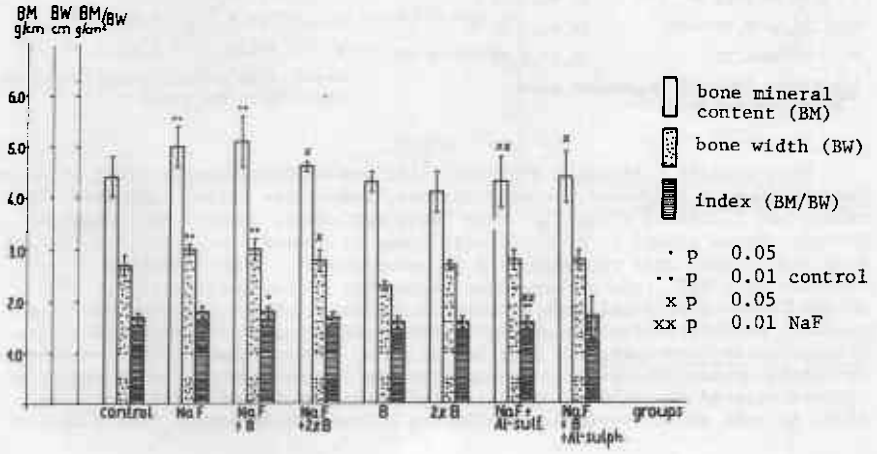
Results of the ^{125}I photon absorptiometry - metacarpal bone II



metacarpal bone II Results of the ^{125}I photon absortiometry

Figure 2 b

Results of the ^{125}I-photon absorptiometry, Femur (a.p.)



all measuring points. Addition of 4 mg B/kg (Group 5) to the diet did not prevent F action. Doses of 8 mg B/d (Group 6) almost eliminated fluoride action in all groups. Boron by itself (groups 5 and 6) caused a decrease on bone mineral content in all bones, compared to controls. Bone width remained unchanged which was more evident in group 6 (8 mg B). Addition of Al-sulphate as an antidote (Group 7) led to a significant decrease of BM compared to Group 2. All values were still below those of controls. Group 8 (F + 4 mg B + Al--sulphate) showed no substantial changes compared to Group 7.

Roentgenological findings: Pathological findings in line with fluorosis were absent. Bone tissue density varied in each group.

Cortical index: Table 1 shows the mean values from computations dealing with the femur in antero-posterior and lateral direction; the two metacarpals were also measured in two planes. Compared to controls, the administration of boron by itself (Groups 5 and 6) caused a significant decrease in the index on all bones. The action was similar in Group 7 (Al-sulphate). Fluoride alone (group 2) also caused, at least in the metacarpals, a decrease in the cortical index. No significant antidote action was apparent.

Table 1

Results of Cortical Indices According to Exton-Smith et al. (7)

Group	Femur ap. and lateral in %	Metacarpal bone II and III ap. and lateral in %
1	40.68±3.97	34.01±4.19
2	39.60±2.90	27.41±3.30 **
3	41.01±2.35	30.40±5.09
4	36.69±5.72	25.85±3.47 **
5	35.96±4.51 *	30.38±5.03
6	30.03±5.58 **	27.96±4.95 *
7	34.60±3.72 **xx	28.51±3.16 **
8	40.66±6.20	30.43±5.38

* p<0.05; **p<0.01 to control group; xx p<0.01 to NaF group

Table 2

Histomorphometric Results of the Iliac Crest Bone (Tuber Coxae)

Group	Volume of Spongy Bone %	Volume of Osteoid %	n
1	21.8±4.0	2.9±1.2	6
2	23.2±4.0	10.0±1.6 **	3
3	20.6±3.8	7.8±1.5	4
4	21.9±2.0	6.5±1.3 x	4
5	22.4±2.3	2.3±2.2	4
6	15.2±3.7 **	1.7±1.7	4
7	17.7±3.9	3.5±0.8 xx	3
8	19.3±4.2	4.2±1.3 xx	4

**p<0.01 to control group; x p<0.05; xx p<0.01 to NaF group

Histological findings - Histologically, and histomorphometrically, in Group 2, osteoid seam thickness, osteoid surface, trabeculae volume increased: bone resorption increased slightly. When boron was added, osteoid mass remained normal. Boron groups 5, 6, especially group 6, caused a strong diminution of bone and osteoid mass corresponding to osteoporosis. Administration of Al-sulphate plus NaF reduced bone mass below that of the control group. The osteoid seams were similar to those of the control group. As apparent from table 2, administration of NaF led to increased osteoid and bone mass. Additional B reduced spongiosa mass but affected osteoid quantity only slightly. Spongiosa density was reduced markedly by high boron dose (group 6) with concurrent osteoid diminution. Al sulphate distinctly diminished fluoride action; it even reduced spongiosa volume to a level below normal (Table 2).

Fluoride determination: The fluoride serum values rose, as shown in Fig. 3, in all groups in the course of the study. The serum level rose about six-fold due to NaF by itself (Group 2). With concurrent administration of boron and fluoride, F values were significantly higher (up to 40%) detectable in groups 3 and 4 only during months 5 and 10. When Al-sulphate is administered as an antidote (groups 7 and 8), the fluoride serum level dropped markedly. F concentrations in urine (Fig. 4) varied widely in the individual groups of animals with the following trends: When F was administered, F excretion rose ten to fifteen-fold, whereas excretion in each group almost doubled from the 5th to the 12th month. The constantly increasing excretion in the boron antidote groups (5 and 6) compared to NaF group 2, ranged from 3 to 71% - not statically significant. By addition of aluminum sulphate (group 7), urinary fluoride decreased insignificantly. Conversion of the F concentration to the creatinine excretion in urine (Table 3) shows the same trend. F content in pelvic bone ash (Fig.5) was, on an average, 25% above rib content. Fluoride administration during 13 months induced a rise up to ten times normal values. F values in bone of boron antidote groups 3 and 4 are in the same order of magnitude although slightly higher. Al-sulphate administration (groups 7 and 8) lead to a highly significant 25-30% decrease in F content in ash. The ash content (Table 4) was reduced in all groups compared to con-

Figure 3

Fluoride Content in Serum During Experiment

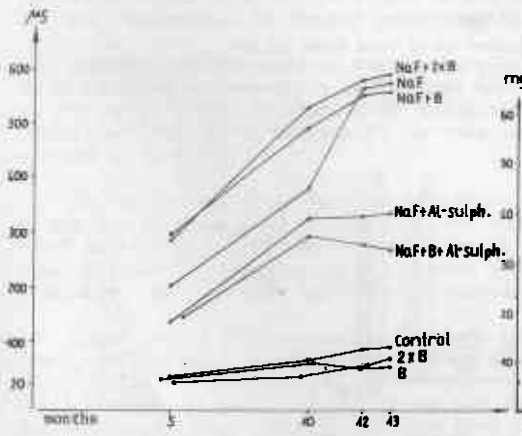
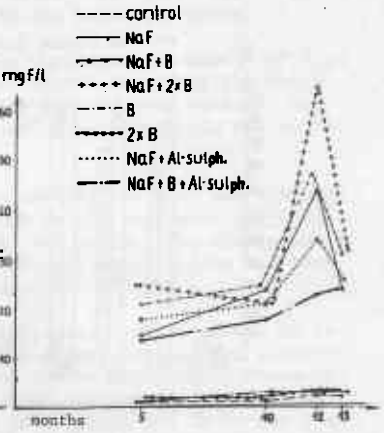


Figure 4

F⁻ Content in Urine During Experiment



trols and to NaF group 2; reduction was most marked in boron groups 5 and 6 and in Al-sulphate groups 7 and 8 (Table 4). Boron content in bone failed to increase in any of the groups. The marked fall in activity of the parathyroid glands (Table 5) in boron groups 5 and 6 is highly significant and the increase in activity in the two Al-sulphate groups 7 and 8 statistically significant. No hyperactivity in NaF group 2 was observed (Table 5).

Discussion

The 13-month duration of administration of 7 mg F /kg/d to young pigs resulted in mild skeletal fluorosis. Macroscopically some exostoses and hyperostoses were observed. Apposition rings in the subperiosteal region were ap-

Table 3

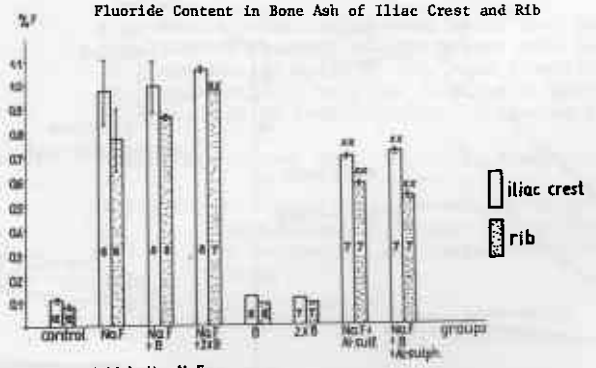
F Content in Urine Calculated to Creatinine Content in Urine (mg F /mmol creatinine)

Group	5th Month	n	10th Month	n	12th Month	n	13th Month	n
1	0.149±0.062	12	0.216±0.042	11	0.109±0.019	10	0.137±0.043	12
2	1.578±0.659**	8	2.114±0.897**	6	2,470±1.562	7	1.472±0.309	8
3	2.254±0.725	8	2.272±1.491	8	2.014±0.499	8	1.748±0.529	7
4	2.468±0.507^{xx}	8	2.330±1.079	8	3.969±2.719	7	1.807±0.291^{x}	8
5	0.112±0.038	8	0.212±0.045	8	0.149±0.031	7	0.153±0.045	7
6	0.124±0.038	8	0.211±0.047	8	0.178±0.063	8	0.195±0.089	8
7	1.650±0.476	8	1.115±0.526^{x}	8	1.527±0.527	7	1.368±0.960	8
8	2.217±1.084	8	1.043±0.261^{x}	8	1.711±0.571	7	1.376±0.456	8

**p<0.01 to control group; ^{x}p<0.05; ^{xx}p<0.01 to NaF group

Figure 5

Fluoride Content in Bone Ash of Iliac Crest and Rib



xx p<0.01 to the NaF-group

parent on the humerus cross-section (9,10). I^{125}-photon absorptiometry showed an increase in mineral content and bone width, which is also typical of human industrial fluorosis (11). Morphometrically obtained cortical indices decreased since the medullary canal is enlarged by increased endosteal resorption. Radiography revealed no reliable fluorosis signs at the femur, the lumbar vertebral column and the metacarpals. Histologically the increase in surface osteoid and osteoid seam thickness at the iliac crest with a slight increase in trabecula thickness and of resorption, is typical of beginning fluorosis (10, 12-15). At a dosage of 0.5 mg F/kg/d, the F level in serum, name-

ly 575 µg/l is almost three times that recommended by some for osteoporosis therapy (16-18). Fluoride values in iliac crest bone ash reached 0.97%, a level equivalent in humans to stage III fluorosis, according to Roholm (10, 13, 14, 19, 20).

Table 4

Ash Content in % to Dried Bone

Group	Iliac Crest%	n	Rib%	n
1	28.3±3.6	12	53.4±4.0	12
2	28.7±3.8	6	52.0±2.5	6
3	27.3±2.9	8	50.3±3.3	8
4	27.6±3.6	8	50.6±5.1	7
5	26.7±3.9	8	48.6±5.6	8
6	26.4±2.3	7	46.0±6.1 *	7
7	26.2±2.2	7	47.0±5.5 *	7
8	29.8±3.4	7	47.7±4.6 *	7

*p<0.05 to control group

Table 5

Parathyroid Activity: Average Area of 150 Parathyroid Nuclei per Animal

Group	μm^2	n
1	18.65±1.79	12
2	18.87±1.23	5
3	19.50±2.57	8
4	18.47±2.60	8
5	13.23±1.08 **	8
6	15.97±1.44 **	7
7	21.05±1.56 **	7
8	21.72±2.08 **	7

**p<0.01 to control group

Boron given in two different dosages as an antidote yielded the following results in the skeletal system. At bone seams, photon-absorptiometrically, histologically and chemically (ash content), fluoride action was cancelled or alleviated by concurrent administration of boron, especially at high dosage (group 6). Elevated F level in serum, associated in both boron groups 5 and 6, with constantly increased F excretion in urine, pointed towards the antidote action of boron. The formation of F-B complexes, which are preferably excreted through the kidney and are less markedly stored in bone (21), theoretically might explain this phenomenon. $[BF(OH)_3]^-$, $[BF_2(OH)_2]^-$, $[BF_3OH]^-$ and $[BF_4]^-$ are the complexes involved: the first three form fairly rapidly but are less stable than the fourth. F analyses of iliac crest and rib ashes, however, failed to show any antidote action of boron. In keeping with the higher F values in serum, F values in bone were slightly higher than those in F group 2. Boron analysis did not prove that boron was stored in bone. This contradiction is explained by analysis of boron action on bone in boron groups 5 and 6 which showed clearly reduced mineral values in femur and metacarpals. A similar effect was shown by the decrease in cortical indices in the roentgenogram and, histologically, by osteoporosis with diminished bone formation, especially in group 6 (8 mg B). Boron causes osteoporosis by its immediate action on bone metabolism. This direct boron action on bone (decrease in bone mass) cancels, at least partially, the direct F action on bone. Since fluoride storage in bone is not affected by it, no genuine antidote action of boron by formation of B-F complex in bone is involved. The raised F levels in serum, the slightly raised F content in bone and the increased F excretion in urine suggest boron-improved F absorption from the intestine, perhaps as (B-F) complex. Elsair et al. (22) proved by acute fluoride intoxication in rabbits (60 mg F /kg/d), an increased F digestive utilization coefficient with increased F excretion in urine and increased fluoride storage in bone, by the use of fluoride balances. The decrease in parathyroid activity in the two boron groups

(5 and 6) may be the key to boron action. An increase in parathyroid activity due to a relatively high NaF dose (15.4 mg/kg/d) could not be proved by determination of the average area of nuclei, though a secondary hyperparathyroidism was actually found in animal experiments involving 200 ppm (23, 24) and in South India in human endemic fluorosis (25-27). In the last-mentioned cases malnutrition, calcium and vitamin D deficiencies associated with a high F intake (up to 65 mg F/d) are some of the causes involved. Secondary hyperparathyroidism in rabbits, acutely and subacutely intoxicated by 60 or 40 mg F/kg, is caused by inhibition of calcium absorption from the intestine (22). Nevertheless, the toxic effect of fluorine seems to be alleviated by concurrent administration of boron (28), as shown in the literature and by our former studies due to varying toxicity of NaF and BF complexes. The oral LD_{50} for potassium fluoroborate in rats is 2,000-3,000 mg/kg according to Hodge and Smith (29); for NaF, the authors found an LD_{50} of 250 mg/kg in rats (30), for fluoroborates toxicity was ten-fold lower. With the ion-sensitive electrode, fluorides as $(BF_4)^-$ complexes can be determined only after destruction of this complex (total fluorides). In the rabbit experiment, after feeding boron and fluorine, ionized fluoride content and total fluoride content coincided; $(BF_4)^-$ complexes were not being formed but rather the stages $[BF(OH)_3]^-$ to $[BF_3OH]^-$ might be assumed.

The following findings from earlier experiments as well as the results of part II confirmed that BF complexes reduced the toxic effect of fluoride. The growth-retarding effect of NaF on green alga Chlorella fusca var. vacuolata is markedly reduced by sodium borate. The glycolysis rate of human erythrocytes, which is reduced by NaF, is raised again by borate addition (1), but normal values were not attained. In the rabbit experiment (2) the reduction in serum iron and iodine level by NaF is prevented by concurrent administration of boron; the same behavior was observed with cholinesterase but boron addition failed to induce normal values. In our first experiment with yearling domestic pigs during 12 months, lowered serum iron level and cholinesterase activity as well as increase in the glucose-6-phosphatase activity caused by 5 mg NaF/kg/d were restored to normal by 0.35 mg B/kg/d.

Administration of boron concurrently with NaF aerosol inhalation in Syrian golden hamsters induced a decrease in cholinesterase and caused transaminases to increase (31). Elsair et al (32) also found that hemostasia disorders in the rabbit, caused by 40 mg F/kg/d, were inhibited by 15.4 mg B/kg/d. The same team, testing liver homogenates in vitro, observed a correction of increased oxygen consumption caused by NaF (33). In acute F intoxication of rabbits (60 mg F/kg/d), administration of boron after discontinuation of fluoride administration accelerates F excretion (detoxication) (34). Negative calcium and phosphorus balances, due to high F doses leading to hypocalcemia and secondary hyperparathyroidism, are corrected by concurrent boron doses: F storage in the skeleton is unaltered or increased (22,28,35). As early as 1965, Hasek (36) found that 3 g of boric acid/100 kg of body weight, administered to fattened bulls, failed to prevent dental fluorosis. On the other hand, the prevailing opinion in the literature is that aluminum salts reduce fluorine retention in bone. For grazing cattle, Gründer (5) reported a reduced absorption quota of 30 - 40% due to various Al-compounds; the bone fluorine content was reduced by about 20%. In poultry, F absorption due to Al-sulphate was significantly reduced (F content in intestine increased by 63%) (37). In humans, F absorption was reduced by 30% due to high F doses of aluminum hydroxide and by 57.6% with low F doses (38).

In our experiments, F storage in bone was reduced 25-29% by Al-sulfate. A distinct pathological change in bone was apparent: bone mass decreased in comparison to the control group. Increased parathyroid activity in both Al--sulfate groups might provide an explanation. Al-sulfate in the intestine leads to precipitation of calcium and phosphorus. The resulting calcium deficiency may then trigger secondary hyperparathyroidism and/or phosphorus deficiency of 1α-hydroxylase in the kidneys. Combinations of Al-sulfate and borate failed to yield any additional information.

Conclusion

Boron acts as an antidote in F intoxication, probably due to formation of less toxic boron-fluoride complexes; F content in bone, however, is slightly increased with a concurrent increase in F serum level and in urinary F excretion. Boron by itself, in the doses used, leads to osteoporosis associated with reduction in parathyroid activity. The boron effect, decrease in bone mass also, at least partially, cancels the F effect on bone, namely, increase in bone mass.

Boron prophylaxis is not recommended. It may even be dangerous, since strong individual differences were observed in chronic fluoride intoxication of humans caused by industry or by high F content in drinking water, and since only a certain proportion of exposed persons are affected by fluorosis (39-41). According to Elsair et al (22) boron is only suitable for short-term use to attain rapid detoxication (curative) after discontinuance of exposure to fluoride. The situation is similar when aluminum sulphate is involved. By reducing F absorption from the intestine and F storage in the skeleton, it leads to secondary hyperparathyroidism due to deterioration in calcium absorption. The mechanism of boron and Al-sulphate action on the development of chronic fluorosis differs. Al-sulphate reduces F absorption; boron acts directly on bone and forms less toxic boron fluoride complexes.

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References

1. Kochmann, W., Bech, R., Grunewald, A., Domesle, E., Kramer, W., Hentschel, H., Wiedner, W., Böhm, H., Schrandt, D., Franke, J., Runge, H., and Brox, u.D.: Ergebnisse experimenteller Untersuchungen an verschiedenen biologischen Testobjekten mit anorganischen Fluoriden und organischen Fluorverbindungen unter Einwirkung mehrerer Gegenmittel. Wissenschaftl. Hefte d. Pädagog. Inst. Köthen, 2:283-302, 1973.
2. Baer, H.P., Bech, R., Franke, J., Grunewald, A., Kochmann, W., Melson, F., Runge, H., and Wiedner, W.: Ergebnisse tierexperimenteller Untersuchungen an Kaninchen mit Natriumfluorid unter Einwirkung von Gegenmitteln. Z. ges. Hyg. 23:14-20, 1977.
3. Grunewald, A., Kochmann, W., Bech, E., and Wiedner, u.W.: Untersuchungen über das Verhalten von Natriumfluorid im Warmblüterorganismus unter Einfluß von Bor und Eisen. Z. ges. Hyg. 24:526-528,1978.
4. Seffner, W., and Tuebener, W.: Antidotes in Experimental Fluorosis on Pigs: Morphological Studies. Fluoride, 16:33-37, 1983.

5. Gründer, H.D.: Fluorimmissionswirkung auf Rinder. Zbl. Vet. Med. A 19:229-302, 1972.
6. Cameron, J.R., and Sorenson, J.: Measurement of Bone Mineral in vivo. Science, 142:230-232, 1963.
7. Exton-Smith, N.A., Millard, P.H., Payne, P.R., and Wheeler, E.:Method for Measuring Quantity of Bone. Lancet, 2:1153-1157, 1969.
8. Chalkley, W.H.: Method for the Quantitative Morphologic Analysis of Tissue. J. Nat. Cancer Inst. 4:47-53, 1943.
9. Shupe, J.L.: Fluorine Toxicosis and Industry. Amer. Ind. Hyg. Assoc. J., 31:240-247, 1970.
10. Roholm, K.: Fluorine Intoxication. A Clinical-Hygienic Study. H.K. Lewis & Co., London, H.K., 1937.
11. Runge, H., Franke, J., Geryk, B., Hein, G., Fengler, F., Paul, H., Bismarck, M., and Schmidt, C.W.: Bone Mineral Analysis in Persons with Long-Time Fluoride Exposure. Fluoride, 12:18-27, 1979.
12. Franke, J.: Histological Changes of Human Fluorosis, Experimental Fluorosis in Animals and Osteoporosis Following Sodium Fluoride Therapy. Fluoride, 5:182-198, 1972.
13. Franke, J., Rath, F., Runge, H., Fengler, F., Auermann, E., and Lennart, G.: Industrial Fluorosis. Fluoride, 8:61-83, 1975.
14. Franke, J., Runge, H., and Fengler, F.: Endemic and Industrial Fluorosis. In: B. Courvoisier, A. Donath and C.A. Baud (Eds.), Symposium CEMO II: Fluoride and Bone, Genève: Editions Medecine et Hygiene, 1978, pp. 129-143.
15. Shupe, J.L., Miner, M.L., Greenwood, D.A., Harris, L.E., and Stoddard, G.E.: The Effect of Fluorine on Dairy Cattle: II. Clinical and Pathological Effects. Amer. J. Vet. Res., 24:964-979, 1963.
16. Taves, D.R.: New Approach to the Treatment of Bone Disease with Fluoride. Feder. Proc. 29:1185-1187, 1970.
17. Fuchs, C., Dorn, D., Hauswald, C., Henning, H.V., Köbberling, J., Kubosch, J., McIntosh, C., Unger, H.D., and Scheler, F.: Fluorid-Spie-gel im Serum bei der Osteoporose-Behandlung mit NaF. Verh. dt. Gesellsch. Inn. Med., 82:910-912, 1976.
18. Sluys Veer, J.J., vanKesteren, G., Backer-Dirks, O., and Flissebaalje: Serum Fluoride Concentrations in Osteoporotic Patients During NaF Treatment. In: B. Courvoisier, A. Donath, and C.A. Baud (Eds.), Symposium CEMO II: Fluoride and Bone, Genève, Editions Medecine et Hygiene, 1978, pp. 293-397.
19. Franke, J., and Auermann, E.: Die Bedeutung der Beckenkammpunktion mit histologischer und mikroanalytischer Untersuchung des gewonnenen Knochenmaterials bei der Diagnostik der Fluorose. Int. Arch. Arbeitsmed, 29:85-94, 1972.
20. Hodge, H.C., and Smith, F.A.: Occupational Fluoride Exposure. J. Occupat. Med., 19:12-39, 1977.
21. Largent, E.J.: Fluorosis, Columbus Ohio State Univ. Press, 1961.
22. Elsair, J., Merad, R., Denine, R., Reggabi, M., Benali, S., Hamrour, H.M., Azouz, M., Khalfat, K., Tabet Aoul, M., and Nauer, J.: Action of Boron Upon Fluorosis, An Experimental Study. Fluoride, 15:75-78, 1982.
23. Faccini, J.M., and Care, A.D.: Effect of Sodium Fluoride on the Ultrastructure of Parathyroid Glands of the Sheep. Nature, 207:1399-1401, 1965.
24. Faccini, J.M.: Fluorine and Bone. Calcif. Tissue Res., 3:1-16, 1969.
25. Teotia, S.P., and Teotia, M.: Endemic Skeletal Fluorosis in Children

Evidence of Secondary Hyperparathyroidism. In: Frame, B., Parfitt, A.M., and Duncan, H. (Eds.): Clinical Aspects of Metabolic Bone Disease, Excerpta Medica, Amsterdam, 1973, pp. 232-238.

26. Teotia, S.P.S., and Teotia, M.: Secondary Hyperparathyroidism in Patients with Endemic Skeletal Fluorosis. Brit. Med. J. 1:637-640, 1973.
27. Makhni, S.S., Sidhu, S., Singh, P., and Singh, G.: The Parathyroid in Human Fluorotic Syndrome. Fluoride, 14:17-19, 1980.
28. Elsair, J., Merad, R., Denine, R., Reggabi, M., Alamir, B., Benali, S., Azzouz, M., and Khelfat, K.: Boron as a Preventive Antidote in Acute and Subacute Fluoride Intoxication in Rabbits: Its Action on Fluoride and Calcium-Phosphorus Metabolism. Fluoride, 13:129-138,1980.
29. Hodge, H.D., and Smith, F.A.: Biochemical Effects of Inorganic Fluorides. In: Simons, J.H. (Ed.), Fluorine Chemistry Vol. IV., Acad. Press, New York, London, 1965.
30. Franke, J., Runge, H., Fengler, F., and Wanka, Ch.: Beitrag zur experimentellen Knochenfluorose. Int. Arch. Arbeitsmed. 30:31-48,1972.
31. Rathmann, D., Bech, R., and Baer, H.P. Beitrag zur tierexperimentellen Fluorose nach Inhalation von NaF-Aerosolen. Z. ges. Hyg. 23: 632-633, 1977.
32. Elsair, J., Merad, B., Denine, R., Reggabi, M., Benali, M., Alamir, B., and Ali Rachedi, M.: Effects of Fluoride Intoxication of Several Months on Hemostasis in Rabbits in the Presence and Absence of an Antidote (Boron). Fluoride, 12:136-143, 1979.
33. Elsair, J., Merad, R., Denine, R., Reggabi, M., Alamir, B., Benali, M., Khelfat, K., and Ali Rachedi, M.: Effect of Fluoride and an Antidose (Boron) on Respiration of Liver Tissue in Rabbits, Fluoride, 12:172-176, 1979.
34. Elsair, J., Merad, R., Denine, R., Regabbi, M., Benali, S., Azzouz, M., Khelfat, K., and Tabet Aoul, M.: Boron as an Antidote in Acute Fluoride Intoxication in Rabbits: Its Action on the Fluoride and Calcium-Phosphorus Metabolism. Fluoride, 13:30-38, 1980.
35. Elsair, J., Merad, R., Denine, R., Azzouz, M., Khelfat, K., Hamrour, M., Alamir, B., Benali, S., and Reggabi, M.: Boron as Antidote to Fluoride Effect on Bones and Claws in Subacute Intoxication of Rabbits. Fluoride, 14:21-29, 1981.
36. Hasek, A.G., Narozny, J., Hluchan, E., and Blaho, U.: Erforschung der Maßnahmen zur Einschränkung der Fluorose bei Mastbullen. Veterinarni Med., 10:605-614, 1965.
37. Cakir, A., Sullivan, T.W., and Mather, F.B.: Alleviation of Fluoride Toxicity in Starting Turkeys and Chicks with Aluminum. Poultry Sci., 57:498-505, 1978.
38. Spencer, H., Kramer, L., Norris, C., and Wiatrowski, E.: Effect of Aluminum Hydroxide on Fluoride Metabolism. Clin. Pharmacol. Ther. 28:525-539, 1980.
39. Franke, J.: Wirkungen von Fluor auf das Skelettsystem unter besonderer Berücksichtigung der Industriefluorose und der Natriumfluoridbehandlung der Osteoporose. Med. Diss. (B), Halle/S., Martin Luther Universität, 1976.
40. Franke, J.: A New Concept of the Effect of Fluoride on Bone. Fluoride, 12:195-208, 1979.
41. Fluoride Effects on Vegetation, Animals and Humans. Eds. Shupe, U.L., Peterson, H.B., Leone, N.C., Paragon Press, Salt Lake City, 1983, pp. 227-232.

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