Blood plasma levels of fluoride in Icelandic sheep

JAKOB KRISTINSSON
Department of Pharmacology, University of Iceland

EGGERT GUNNARSSON
Keldur, Institute for Experimental Pathology, University of Iceland

PORKELL JÓHANNESSON
Department of Pharmacology, University of Iceland

PÁLL A. PÁLSSON
Keldur, Institute for Experimental Pathology, University of Iceland

and

HÖRÐUR PORMAR
Technological Institute of Iceland

SUMMARY
Fluoride was determined in blood plasma of Icelandic sheep kept at the Hestur Sheep Experimental Farm in Borgarfjörður in western Iceland in the autumn 1988 and in the spring 1989. The results showed a seasonal variation in the plasma levels of fluoride. The plasma levels were in the range of 12–31 ng/ml in the autumn after the animals had been grazing on an uncultivated pasture during the preceding summer. Corresponding levels the next spring in sheep, which had been fed indoors during the winter, were in the range of 80–330 ng/ml. These spring levels varied according to different fluoride content of the fodder. In the preceding 7–8 weeks before the blood samples were collected, the sheep had been fed on either hay + fishmeal or hay + concentrates. The plasma levels were, with one exception, always higher in the sheep fed on hay + fishmeal. This was indeed expected since the amounts of fluoride in the fishmeal were almost four times higher than in the concentrates.

Preliminary results with a limited number of sheep, which had been grazing in the vicinity of the aluminium smelting plant at Straumsvík southwest of Reykjavík, indicate that there is apparently no connection between plasma levels of fluoride and the occurrence of dental fluorosis. Dental fluorosis seems, on the other hand, to be related to the amounts of fluoride in bone.

Key words: dental fluorosis, fluoride in bone, fluoride in plasma, seasonal variation, sheep.

YFIRLIT
Péttni flúoríðs í blöðplasma í íslensku sauðfé
Rannsóknir á 20 kindum, sem slátráð var haustið 1989 og gengið höfðu um sumarið í nágrenni álversins í Straumvík, gáf til kynna að samhengi væri á milli þéttni flúoríðs í kjálkum og einkenna um gadd. Hins vegar virðist ekkert öruðgt samhengi vera milli þéttni flúoríðs í plasma og einkenna um gadd.

INTRODUCTION

Previously we have studied the fluoride concentration in blood plasma of human subjects (odontological students) and found it to be relatively low, as could in fact be expected, since the normal water supply in Iceland is relatively devoid of fluoride (Kristinsson et al., 1986a). It is, on the other hand, well known that the fluoride amounts of surface water accessible to domestic animals may temporarily reach high levels during volcanic eruptions.

Sheep is the species of domestic animals in this country that most often might be exposed to water or forage containing temporarily excessive amounts of fluoride. This is sometimes known to result in acute as well as more chronic toxic effects (dental fluorosis) (Sigurðsson and Pálsson, 1957).

It was accordingly thought to be of interest to study the normal levels of fluoride in plasma in the sheep. This was done in order to be better able to interpret what might be considered as toxic levels of fluoride in the plasma of these animals during or shortly after volcanic eruption.

MATERIALS AND METHODS

The sheep used for these studies were of the Icelandic breed of sheep kept at the Hestur Sheep Experimental Farm in the Borgarfjörður area in western Iceland. In addition samples were taken from twenty animals from a farm near to the aluminium plant at Straumsvík in southwestern Iceland.

Blood samples were collected in polyethylene tubes previously rinsed with 35% perchloric acid and distilled water. Heparin was added to the tubes in order to prevent coagulation. Plasma was separated by centrifugation and kept frozen at –20°C until the day of analysis. Fluoride was determined in the plasma samples with a fluoride selective electrode as previously described (Kristinsson et al., 1986ab). The results were expressed as ng fluoride per ml of plasma.

Hay samples were analysed as described by Doziner and Sígg (1976). The samples were ground to a fine powder (particle size <1 mm) and dried for at least 2 hours. An accurately weighed 1 g aliquot of the haypowder was stirred for 20 min with 50 ml of 0.1 M nitric acid. Finally this solution was adjusted to approximately pH 5.2 with 50 ml of citrate buffer containing a known amount of fluoride and the fluoride concentration determined with a fluoride selective electrode. The fluoride content of the citrate buffer was subtracted from the read values and the fluoride content of the hay calculated. The results were expressed as µg fluoride per g of a dried sample.

Samples of fishmeal, pelleted concentrate mixture and mandibles of sheep were ashed overnight in a furnace at 500°C. The ash was ground finely in a mortar and an accurately weighed aliquot dissolved in 10 ml of 0.5 M nitric acid and adjusted to approximately pH 5.2 with a citrate buffer as described above for the hay samples. Fluoride was determined in the solution as described above and the fluoride content of the samples calculated. The results were expressed as µg fluoride per g of sample.

Water samples were diluted 1:1 with a pH 5.2 buffer containing an known amount of fluoride. The fluoride concentration in the diluted sample was determined with a fluoride selective electrode. The fluoride content of the water was calculated as described for the hay samples. The results were expressed as µg fluoride per ml of sample.
RESULTS
Fluoride levels in plasma were determined in 28 individual sheep in October 1988. They had all been grazing on uncultivated pastures during the summer. Twenty-three of them were adult sheep (1–10 years old) but five were lambs born in the spring (5–6 months old). Fluoride levels ranged from 12–31 ng/ml (Figure 1). The five lambs were, however, all in the lowest bracket (10–15 ng/ml). The lambs had never been fed indoors but the adult sheep had all been kept and fed indoors the previous winter, or in the case of the older sheep, the preceding winters.

In May 1989 blood samples were taken for analysis of fluoride in plasma from nineteen ewes that had been fed indoors during the winter. During the last 7–8 weeks before the samples were collected, eleven of the ewes had been fed on hay and fishmeal. The others were fed on hay and concentrates (pelleted concentrate mixture consisting of 43% maize, 30% grassmeal, 20% fishmeal, 5% sugar and 2% of minerals and vitamins). The plasma levels of fluoride in these ani-

mals ranged from 80–330 ng/ml (Figure 2). They were, with one exception, always higher in the ewes fed on hay + fishmeal than in those fed on hay + concentrates. These levels were uniformly higher (six to twelve-fold) than was the case in the sheep that had been grazing on uncultivated pastures during the summer (Figure 1 and 2).

Twenty sheep (7 adult sheep and 13 lambs) from a farm in the vicinity of the aluminium smelting plant at Straumsvík southwest of Reykjavík were killed in October 1989 after having grazed in that area during the summer. Six of the adult sheep showed dental fluorosis. Fluoride was determined in plasma (obtained immediately before slaughtering) from all of these animals, as well as in their mandibles. The results are shown in Figure 3. From the figure it is clear that the fluoride levels in plasma were similar to those found in the sheep fed on hay + concentrates (Fig-

![Figure 1. The distribution of plasma levels of fluoride in 28 Icelandic sheep grazing on uncultivated pastures. The plasma samples were collected in October 1988. The mean level of fluoride in the samples was 19.9 ng/ml.](image1)

![Figure 2. The distribution of plasma levels of fluoride in 19 Icelandic sheep kept indoors during the winter 1988–1989. The plasma samples were collected in May 1989 before the sheep were let out for grazing. The shaded bars represent sheep fed on hay + concentrates (mean level 120 ng/ml) and the open bars sheep fed on hay + fishmeal (mean level 252 ng/ml).](image2)


there was apparently no connection between the levels of fluoride in blood and dental fluorosis. However, dental fluorosis seemed to be correlated with the amount of fluoride in bone (Figure 3).

Fluoride content was determined in the hay used for feeding the sheep at Hestur as well as in the concentrates and the fishmeal. The drinking water in the sheep house was also analysed. In the hay the fluoride concentration was 0.7 μg/g, 61 μg/g in the concentrates, 229 μg/g in the fishmeal and less than 0.05 μg/ml in the drinking water.

DISCUSSION

The fluoride levels in sheep that had been grazing on uncultivated pastures during the summer were uniformly low or in the range of 12–31 ng/ml (Figure 1). However, only two of the adult sheep had fluoride levels in the lowest bracket. The other were lambs that had never been housed. The question thus arose whether feeding of the adult sheep indoors during the winter (or winters) before might influence this “autumn level” of fluoride in the blood. This was even more pertinent since Richards et al. (1985) have shown that in the pig the biological half-life of fluoride is remarkably long (about 60 days) and for that reason fluoride ingested in the feed during winter and subsequently accumulated in bone might influence the amounts of fluoride found in plasma during the following summer. In the present studies we therefore determined the levels of fluoride in plasma in the spring after the sheep had been kept indoors and fed during the winter.

These studies show that the levels of fluoride in plasma were uniformly higher in the spring than in the autumn. The highest spring levels of fluoride in the plasma of adult sheep were thus up to twenty times higher than the lowest autumn levels. Available data (Smith, 1966) indicate that naturally occurring dietary fluoride is rather well absorbed (61%) by the sheep. It seems therefore reasonable to conclude that the difference in spring and autumn levels of fluoride in the sheep was caused by different fluoride content of the diet in winter and in summer, respectively. This conclusion is further supported by the fact that the highest plasma levels of fluoride in the spring were, with one exception, always found in the sheep receiving the highest amounts of fluoride in the fodder. It is therefore quite possible that the high plasma levels of fluoride found in the autumn in adult sheep was the result of a high intake of fluoride during the previous winter.

Good records were kept of all the sheep and it was obvious that the high amounts of fluoride in plasma as shown in Figure 2 had no acute detrimental effect on the health of the animals. This was in fact remarkable since the spring levels were so much higher than the autumn levels (Figure 1 and 2). The question therefore emerged: What is the acute toxic level of fluoride in the plasma of the
sheep? In this connection it is of interest to note that French and Moroccan workers could not detect any acute toxic effects of ingested fluoride in sheep at plasma levels approximately 1300 ng/ml and only when plasma levels were in the range of approximately 5000 ng/ml was some restlessness and loss of appetite observed (Kessabi et al., 1985). It should furthermore be noted, that only one or two of the animals used in our studies developed signs of dental fluorosis. Experiments are now in progress in order to study the acute and chronic effect of fluoride in Icelandic sheep and how these might be reflected in higher plasma levels of fluoride.

Preliminary results obtained from a limited number of sheep indicate that there is apparently no connection between the levels of fluoride in plasma and dental fluorosis. Dental fluorosis, on the other hand, seemed to be correlated with the amounts of fluoride in bone (Figure 3). It should be noted, however, that the plasma levels of fluoride in most of these animals were similar in the autumn to those found in the spring in sheep fed on hay + concentrates thus indicating a definite exposure to fluoride during the grazing period (Figure 1, 2 and 3). The main objective of further experiments should therefore be to determine which concentrations of fluoride in plasma would indicate later development of dental fluorosis in the sheep. If this is known it should be possible to avoid economical losses on farms in areas suffering from contamination of volcanic ash, by moving the sheep to safer areas in time. This could for instance have been relevant during two relatively recent eruptions of Mount Hekla in 1947–1948 and 1970 (Sigurðsson and Pálsson, 1957; Georgsson and Pétursson, 1972).

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REFERENCES


