

EFFECT OF FLUORIDE ON COLLAGEN CROSS-LINK PRECURSORS OF RABBIT TISSUES

A.K. Susheela and Y.D. Sharma

Fluorosis Research Laboratory, Department of Anatomy, All India Institute of Medical Sciences,
New Delhi - 110 029, India

Paper received: 5th August, 1981

Earlier reports have provided evidence to suggest that fluoride inhibits collagen biosynthesis (1, 2). Efforts have been made to study the nature of the collagen laid down, by investigating the saturated peptide-bound aldehydes which are

Effect of sodium fluoride on saturated aldehyde content of salt soluble collagen of rabbit tissues (means \pm SD, n = 5)

	80 days	154 days	176 days
Bone			
Control	0.42 \pm 0.03	0.63 \pm 0.01	0.78 \pm 0.02
Experimental	0.24 \pm 0.03	0.34 \pm 0.03	0.21 \pm 0.03
Tendon			
Control	0.42 \pm 0.02	0.54 \pm 0.04	0.72 \pm 0.03
Experimental	0.12 \pm 0.03	0.12 \pm 0.03	0.15 \pm 0.03
Trachea			
Control	0.30 \pm 0.05	0.42 \pm 0.04	0.51 \pm 0.03
Experimental	0.22 \pm 0.03	0.34 \pm 0.01	0.12 \pm 0.03
Skin			
Control	0.27 \pm 0.03	0.60 \pm 0.02	0.57 \pm 0.08
Experimental	0.12 \pm 0.03	0.20 \pm 0.02	0.40 \pm 0.04

Values are expressed as μ M of acetaldehyde/100mg of collagen. $P < 0.001$ (Student's *t* test) for all comparisons with respective control tissues.

known to be the cross-link precursors. The present communication therefore reports the status of the saturated peptide-bound aldehyde content in salt soluble collagen following excessive fluoride ingestion. Tissues such as bone, tendon, tracheal rings and skin were investigated as they are representative of different types of collagen.

One group of animals used as experimental samples was given 50 mg NaF/kg body weight, at every 24 hours through intragastric route. The other group of animals was used as controls. Experimental animals and age matched controls were killed on the day 80, 154 and 176. The neutral salt soluble collagen was extracted and purified as described by Kang *et al.* (3). The collagen samples were then dissolved in 0.1 M glycine buffer (pH 4.0) and denatured at 60 °C for 20 minutes. The saturated aldehyde associated with the salt soluble collagen was measured spectrophotometrically at (312 nm) according to the method of Paz *et al.* (4) using N-methylbenzothiazolone hydrazone (METH) reagent.

Results and discussion: The results obtained for aldehyde content are given in the table. Although salt soluble collagen is a minor fraction of total tissue collagen (5), this fraction was preferred for the present study as this contains mostly tropocollagen molecules. The data obtained on the 4 different tissues of the control animals revealed that the saturated aldehyde content increased with the duration of the experimental phase, possibly indicating increased cross-link precursors with advancing age. Age related changes in collagen cross-links are known to occur (6).

It is also evident that fluoride ingestion led to significant reduction in saturated aldehyde content. Although, the aldehyde contents of bone and tendon tissues are (collagen type I) in the same range in the control animals, the effect of fluoride ingestion is more pronounced on tendon collagen, which is evident from the higher percentage reduction. The aldehyde contents of trachea (collagen type II) and skin (collagen type I + III) in control animals are less compared to those of bone and tendon collagen. Fluoride also induced reduction in the aldehyde content in both the tissues. In bone and tendon, the percentage reduction of aldehyde contents showed a linear increase with the duration of fluoride ingestion. However, in trachea and skin the pattern was not the same.

The results of the reduction of the saturated aldehyde content further substantiate the view that the effect of fluoride ingestion is variable among the different tissues having different types of collagen. The reduction in aldehyde content could be due to the impairment in its formation. It is known that the copper dependent enzyme, lysyl oxidase, which is responsible for the formation of aldehyde, is affected in fluoride toxicity (7). It is also known that fluoride reduces the copper content of various tissues (8). It is therefore suggested that due to excessive ingestion of fluoride the tropocollagen molecules with reduced number of aldehydes are likely to produce inadequately cross-linked collagen fibers. It may be concluded that fluoride ions interfere with the normal metabolism and maturation of collagen by exerting an adverse effect on the saturated peptide-bound aldehyde.

1. Rao, K. and Susheela, A.K. (1979) *Fluoride*, 12, 65-71
2. Susheela, A.K. and Mukherjee, D. (1981) *Toxicol. Eur. Res.*, in press
3. Kang, A.H., *et al.*, (1966) *Biochemistry*, 5, 509-515
4. Paz, M.A., *et al.*, (1965) *Arch. Biochem. Biophys.*, 109, 548-559
5. Levene, C.I. and Gross, J. (1959) *J. Exp. Med.*, 110, 771-790
6. Bailey, A.J., Robins, S.P. and Balian, G. (1974) *Nature*, 251, 105-109
7. Machoy, Z. (1980) *Fluoride*, 13, 39-41
8. Tao, S. and Suttie, J.W. (1976) *J. Nutr.*, 106, 1115-1122

We wish to thank the Indian Council of Medical Research and Department of Environment (Government of India) for financial support.