

**ANALYSIS AND EVALUATION OF FORMS OF FLUORIDE IN MILK  
FROM  
NATURALLY HIGH AND LOW FLUORIDE AREAS IN KENYA  
AND  
THE EFFECT OF THIS INTRINSIC MILK FLUORIDE ON TEETH  
DEMINERALISATION.**

**BY**

**Rebeccah W Kahama**

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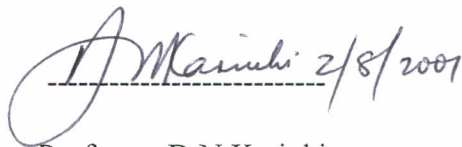
## DECLARATION

I declare that this thesis is my original work based on six publications to international refereed journals. It has not been presented for a degree in any other University.

A handwritten signature in cursive script, reading "Rebecca W Kahama", written over a horizontal dashed line.

Rebecca W Kahama

This thesis has been submitted with my approval as a University supervisor.

A handwritten signature in cursive script, reading "D N Kariuki 2/8/2007", written over a horizontal dashed line.

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## ABSTRACT

There is very little information on fluoride content in breast and cows' milk in Kenya and the African region at large. Available information on breast milk fluoride is based on samples from a single area and different analytical methods. Caries inhibiting effects of fluoride have been established but the role of both cow and human intrinsic milk fluoride on teeth demineralization has not been reported.

This study provides two reliable modern analytical methods for measuring intrinsic milk fluoride; the HMDS microdiffusion and the GLC were compared with others and validated. It describes the effect of intrinsic milk fluoride on enamel demineralisation, fluorosis in Elementaita region and the forms of fluoride in milk.

An accurate and rapid analytical method for fluoride determination in milk was developed by the comparison and improvement of four methods namely:

i) direct diffusion using HMDS, ii) Ashing followed by diffusion using HMDS, iii) gas chromatography, iv) HF microdiffusion. Fluoride levels in human and cows' milk using the established method above were analysed. The factors affecting fluoride levels in milk e.g. animal breed, season, lactation period, parity and the concentration of other ions in milk were also investigated. Forms of fluoride in milk were also evaluated and the effect of milk fluoride on teeth demineralisation was established.

The results show the HMDS microdiffusion method and the GC extraction method for total and ionic fluoride were very comparable in reproducibility and accuracy. The mean fluoride concentration in breast milk was  $0.48 \mu\text{M}$  (range  $0.2-1.05 \mu\text{M}$ ) in areas with less than  $1.0 \mu\text{g ml}^{-1}$  fluoride in drinking water and  $3.44 \mu\text{M}$  (range  $0.87-12.10 \mu\text{M}$ ) in an area with  $11.4 - 20.6 \mu\text{g ml}^{-1}$  fluoride in

drinking water. Cow milk samples span a wide range of 0.02-0.34  $\mu\text{g ml}^{-1}$  in individual animal samples. Regional averages ranged from 0.04-0.15  $\mu\text{g ml}^{-1}$  in areas with 0.01-20.6  $\mu\text{g ml}^{-1}$  fluoride in drinking water.

The forms of fluoride in milk were evaluated by determining the concentrations of diffusible and total fluoride in cow milk samples. The diffusible fluoride was determined by direct HMDS microdiffusion while for total fluoride, samples were subjected to either open ashing or digestion with proteolytic enzymes before microdiffusion. Diffusible fluoride had a range of 0.024 – 0.28  $\mu\text{g ml}^{-1}$  while total fluoride ranged from 0.05 - 0.31  $\mu\text{g ml}^{-1}$ . Fluoride levels in milk fractions indicated that the bound fraction was in the milk proteins. It was concluded that all fluoride in milk is inorganic in nature with the bound fluoride being physically or chemically sequestered in the milk proteins. The proposed method is convenient for total fluoride analysis in milk.

With special reference to children, a study was designed to establish and describe the levels of dental fluorosis and also to determine other sources of fluoride to the community. The levels of dental fluorosis were recorded using Thystrup and Fejerskov classification method (TF) for children aged between 2 - 14 years. The high fluorosis level of 95.8% was associated with the fluoride concentration in the community water supply and food which were also analysed. The effect of intrinsic milk fluoride on teeth demineralization gave a 36% reduction in calcium loss at pH 5.0 by treatment with milk with only 0.30  $\mu\text{g ml}^{-1}$  fluoride. This is an indication that intrinsic milk fluoride has some caries protective properties.