

# Crosslink in bone collagen in Paget's disease

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**SYNOPSIS** The crosslink in bone collagen was analysed in specimens of bone obtained at necropsy from cases of Paget's disease and compared with normal bone collagen of the same age. The specimens were stored at  $-20^{\circ}\text{C}$  before analysis.

The predominant crosslink in a normal bone collagen was hydroxylysino-hydroxynorleucine (di OH-LNL) (F1), which was designated syndesine in the past; another fraction, hydroxylysino-leucine (HLNL) (F2), much less prominent than di OH-LNL, was also noted in a normal bone collagen. Both fractions were reduced in bone tissue of advancing age. The peak corresponding to HLNL was considerably increased in Paget's disease. This abnormality was constantly seen in specimens of bone from cases of Paget's disease, but the significance of the finding could not be assessed from the present investigation.

Calcitonin has been shown to produce complete remission in Paget's disease and the crosslink pattern was found to be normal in specimens examined from a calcitonin-treated patient. This shows that calcitonin has some effect on the metabolism of collagen and a normal crosslink in such a situation lends support to this idea.

The study of crosslinks in collagen has added much to our knowledge of collagen chemistry but its application to clinical problems requires further investigation. It is generally accepted that the collagen fibres are stabilized by a system of covalent crosslinks (Bailey, 1968; Veis, 1967), and their formation is linked intimately with growth and development. Reduction with borohydride and subsequent analysis of acid hydrolysate of various collagen tissues showed that the more soluble collagen contained labile aldimine bonds (F2), whereas di OH-LNL (F1) was the only significant reducible crosslink in bone collagen (Bailey, 1970). Further investigation confirmed the identity of fraction 2 as HLNL and it was suggested that the crosslink existed in the fibre in the aldimine bond (Schiff base) as dehydrohydroxylysino-leucine (Fowler and Bailey, 1972). It was also shown that HLNL accounted for 33% of the tritium activity in the column effluent from a hydrolysate of normal bone collagen treated with  $^3\text{H}$  borohydride (Eyre and Glimcher, 1972).

Paget's disease is associated with increased turnover of bone. There is resorption and resynthesis of bone matrix. The serum acid and alkaline

phosphatase is raised and hydroxyproline excretion is increased. The defect in Paget's disease causes a disturbance in bone metabolism and for this reason it was decided to analyse the crosslink in bone of Paget's disease and compare it with normal bone of the same age. It was also intended to analyse specimens of affected bone from patients with Paget's disease who had been treated with calcitonin.

## Materials and Method

Specimens of bone from skull and pelvis were obtained at necropsy from three groups of patients between the ages of 60 and 75: (1) 15 specimens of normal bone from 10 necropsies (six men and four women) and five specimens of normal bone from three patients aged 15-20 years who died in road traffic accidents; (2) 10 specimens of bone affected with Paget's disease from eight patients (three men and five women); (3) on one occasion three specimens of bone were obtained from a 60-year-old man with advanced Paget's disease who had been treated with calcitonin for two months before his death. The specimens were collected from the affected bones located during life by bone radiography.

Soft tissue was dissected from the bone, which was then crushed and decalcified with 0.35 M EDTA





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