V8

Hydroxyapatite Crystals Orientation during a Pathological Calcification of Human Tendons

A. BIGI, A. RIPAMONTI, N. ROVERI

Istituto Chimico 'G. Ciamician', Università di Bologna, Bologna, Italy

L. DOVIGO and M. SCARAMELLI

Divisione di Reumatologia, Arciospedale Sant'Anna, Ferrara, Italy

The structural relationship between collagen fibers and apatite deposits in mineralized tissues has been widely investigated by electron microscopy and X-ray diffraction techniques [1]. For these studies oriented samples, such as osteons and turkey leg tendon at different degrees of calcification, have been particularly useful [2–4]. However many features of the problem are still obscure. The availability of oriented samples containing collagen fibers at low degrees of calcification is an essential condition to obtain information about the sites of the initial deposition of the apatite crystals in the collagen fibers structure using diffraction techniques. Tendon affected by diffuse idiopatic skeletal hyperostosis, which is a disease characterized by calcification and ossification of tendons and ligaments [5], is a suitable tissue because calcium hydroxyapatite is laid down within the tissue to different degrees along its length and to different degrees with the development of the disease.

We have examined fragments of human tendons which showed radiographic evidence of this pathological calcification at the sites of attachment to olecranon by small and high angle X-ray diffraction techniques in order to investigate the inorganic deposits and their interaction with collagen fibers.

X-ray diffractometric analysis carried out on powdered mineralized samples permitted the recognition of the mineral deposit present in the tissue as hydroxyapatite. This inorganic phase exhibits a good degree of crystallinity and cell parameters $a = 9.40(1)$ Å, $c = 6.89(1)$ Å very close to those of bone apatite.

High angle X-ray diffraction analysis carried out by means of a flat camera on fragments dissected from several zones of the tendons revealed a different orientation of the apatite crystals as a function of the degree of calcification of the tissue. The apatite crystals appear to be preferentially oriented with their c-axis parallel to the tendon axis in the zones at a low degree of calcification. This orientation reduces progressively as the degree of calcification increases and no preferential orientation can be observed in the fully calcified zones. The X-ray reflections characteristic of collagen molecular structure appear superimposed on those of the inorganic phase and show that collagen fibers orientation is always coincident with that of the c-axis of the hydroxyapatite crystals.

The small angle X-ray diffraction pattern recorded on fragments at a low degree of calcification shows the characteristic reflections of collagen fibrillar structure which appear oriented parallel to the tendon axis. No diffraction reflection due to collagen can be observed in the small angle X-ray diffraction pattern obtained from fully calcified fragments.

These results show a clear relationship between apatite crystals deposition and collagen fibers distribution and orientation. In fact at a low degree of calcification apatite crystals are aligned parallel to the collagen fibers of the tendon whereas at a high degree of calcification no orientation of collagen fibers as well as of apatite crystals could be revealed. This agrees with the morphological observations which show that at a high degree of calcification the collagen fibers lose their characteristic orientation parallel to the tendon axis and assume an isotropic distribution in a structure closely resembling that characteristic of cancellous bone. Furthermore it must be noted that the collagen molecular packing does not seem to be affected by the mineralization process, at least during the initial stage of the deposition.

1 A. Engstrom, in 'The Biochemistry and Physiology of Bone', G. H. Bourne (ed.), Academic Press, New York, 1972, pp. 237–256.
2 A. Ascenzi, E. Bonucci, A. Ripamonti and N. Roveri, *Calcif. Tissue Res.*, *25*, 133–143 (1978).
3 A. Ascenzi, E. Bonucci, P. Generali, A. Ripamonti and N. Roveri, *Calcif. Tissue Int.*, *29*, 101–105 (1979).
4 S. W. White, D. J. S. Hulmes, A. Miller and P. A. Timmins, *Nature*, *266*, 421–425 (1977).
5 D. Resnick and G. Niwayama, *Radiology*, *119*, 559–568 (1976).

V9

Cu(II) Adriamycin Complexes. Identification and Interaction with DNA

HELOISA BERALDO, LUCIA TOSI

Département de Recherches Physiques, LA N° 71, Université Pierre et Marie Curie, 75230, Paris, France

and ARLETTE GARNIER-SUILLEROT

Laboratoire de Chimie bioinorganique, UER de Médecine et Biologie Humaine, Université Paris XIII, 93000 Bobigny, France

Adriamycin (Adr) is an anthracycline antibiotic widely used in the treatment of various human cancers. Once in the cell, adriamycin localizes in the nucleus and is believed to act by inhibiting both DNA replication and RNA transcription [1]. Since metal