

CUTANEOUS ACID MUCOPOLYSACCHARIDES IN PSEUDOXANTHOMA ELASTICUM*

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The fibers taking elastic tissue stains in pseudoxanthoma elasticum (PXE) are more similar to elastin than collagen (1). Associated with these elastotic fibers are tinctorially demonstrable increased acid mucopolysaccharides (AMPS) as demonstrated by the Mowry colloidal iron and alcian blue stains (2, 3). Also, PXE dermis has twice as much hexosamine on a dry weight basis as normal human adult skin with most of the increase representing glucosamine. (4)

This paper presents quantitative studies on acid mucopolysaccharides in unexposed adult human skin compared with skin diseased with PXE.

MATERIALS AND METHODS

Skin from covered areas of the body carefully trimmed free of fat was obtained at postmortem examination from seven human adults aged 23 to 67 years and from a 46 year old white female† who died from cerebrovascular complications of PXE. After grinding with liquid nitrogen in a Wiley mill and defatting with acetone and ether, the skin was enzymatically digested, deproteinized and the acid mucopolysaccharides precipitated with cetylpyridinium chloride (CPC)‡ according to the method of Schiller *et al.* (5). Acid mucopolysaccharides were eluted with 0.4 M NaCl, 1.2 M NaCl, and 2.1 M NaCl and quantitative measurements for sugar components performed on these eluates freed of CPC by treatment with 0.1 M potassium thiocyanate (6). De-CPCed, dialyzed fractions were analyzed for uronic acid by the orcinol (7) and carbazole procedures (8) and for hexosamine by the Boas modification of the Elson Morgan method. (9) The identity of the hexosamine components was confirmed by ninhydrin degradation followed by paper chromatography (10). The latter method also permitted qualitative resolution of glucosamine and galactosamine.

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RESULTS

Table I indicates the AMPS content expressed as μm uronic acid (via orcinol)/g dry weight of skin. Hexosamine to orcinol ratios were quite close to 1 in all fractions. The carbazole/orcinol ratio was 1.2 ± 0.2 for the 0.4 M NaCl eluates and 0.39 ± 0.17 for the 1.2 M NaCl eluates of the controls. The carbazole/orcinol ratio for the 1.2 M PXE fraction was 0.48. Paper chromatography revealed glucosamine to be the major amino sugar in all 0.4 M fractions while galactosamine was the major component in all 1.2 and 2.1 M fractions.

DISCUSSION

The advantage of the CPC precipitation method of Schiller *et al.* (5) in isolation of AMPS is that it makes possible identification of AMPS from small amounts of tissue (3–5 g dry weight) while the older classic methods of Meyer *et al.* (11) require 50 g or more of dry weight skin for identification of the AMPS.

The 0.4 M NaCl eluate represents hyaluronic acid. This is confirmed by the 1:1:1 ratio of orcinol:carbazole:hexosamine and the identification of the major amino sugar component as glucosamine. The 1.2 M NaCl fraction represents chondroitin sulfate but does not distinguish between chondroitin sulfate A, chondroitin sulfate B, and chondroitin sulfate C. The finding of carbazole/orcinol ratios much less than one with galactosamine as the major amino sugar component suggests that chondroitin sulfate B is the major AMPS present in the 1.2 M NaCl fractions (12).

The finding of galactosamine as the major amino sugar in the 2.1 M NaCl eluates suggests that chondroitin sulfate also is present in this fraction but inadequate amounts of material for analysis make identification impossible.

SUMMARY

The histologic finding of increased dermal acid mucopolysaccharide in pseudoxanthoma elasticum was confirmed utilizing quantitative isolation. The major cutaneous acid mucopolysaccharide in pseudoxanthoma elasticum is

TABLE I

Acid mucopolysaccharides in pseudoxanthoma elasticum (μ m uronic acid (via orcinol)/g dry weight skin)

| | PXE | Control (7 aged 23-67 years) |
|---------------------|-----|------------------------------------|
| Hyaluronic Acid | 3.3 | 0.9 \pm .28 |
| Chondroitin Sulfate | 2.1 | 1.3 \pm .58 |
| Total AMPS | 5.8 | 2.4 \pm .67 |

TABLE II

Carbazole/orcinol ratio for 0.4M and 1.2M NaCl elutes

| NaCl | Controls (7) | PXE |
|------|-----------------|------|
| 0.4M | 1.2 \pm 0.2 | 1.0 |
| 1.2M | 0.39 \pm 0.17 | 0.48 |

hyaluronic acid with lesser amounts of chondroitin sulfate much of which is probably chondroitin sulfate B.

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