position of the unknown spot on both chromatograms, the nature of the unknown can be determined. R. ROBINSON

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¹ Lumb, M., Lab. J., 8, 146 (1946). ² Williams, R. J., and Kirby, H., Science, 107, 481 (1948).

Elastin from Human Tissue and from **Ox Ligament**

THE starting material for the majority of the reported analyses of elastin has been ox ligamentum nuchæ subjected to long periods of boiling in water, the simplest of the methods described by Stein and Miller¹.

Human aorta has been treated in this fashion, and also boiled in frequent changes of 1 per cent acetic acid, a treatment which has been proved by Gross² to remove all structurally intact collagen, as demonstrated by the electron microscope. A hydrolysate of material subjected to prolonged treatment by either of these methods shows on chromatography a far greater concentration of basic amino-acids and hydroxyproline than would be expected from "pure elastin" (cf. Bowes and Kenten³, and Neuman⁴).

Similar chromatograms to those reported by Bowes and Kenten can, however, be obtained from hydrolysates of aorta tissue which has been boiled for five days with frequent changes of between 50 and 100 volumes of 40 per cent urea solution. It might therefore be concluded that certain collagenous substances are more difficult to remove from human tissue than from ox ligament; but that after the complete removal of such proteins the insoluble residue is very similar if not identical in both species, and to this the name 'elastin' could appropriately be given.

Doubt, however, is cast on the validity of this definition of elastin, since it has been shown that the resistance to solution in 40 per cent urea solution of elastin preparations from both ox and human tissues is not absolute, as reported by Stein and Miller, but is a function of the liquor ratio. The whole of an elastin preparation will pass into solution if the volume of the urea solution is raised to between 100 and 200 times that of the solid.

Similarly there are indications of heterogeneity in the residues after treatment with 50 volumes of urea solution. Differences in composition, notably with respect to proline, are observed between the material dissolved from these residues and that remaining after attack by pancreatic elastase⁵.

The definition for elastin, therefore, now becomes rather vague. Under the electron microscope, certain branched and rather ill-defined structures are identified as collagen-free elastin. Chemical analysis shows that these preparations are still contaminated with material having some of the significant amino-acids of collagen. A chemical definition of purity based on insolubility in urea is also suspect, since the amount of urea available alone determines the extent of solution.

A fuller examination of the chemical analysis of various preparations obtained by the action of urea and of elastase on both human and ox tissues is in progress. These results, together with corresponding electron microscope studies undertaken in cooperation with the Department of Biomolecular Structure of this University, will be published elsewhere.

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- Department of Medicine, University of Leeds, 2. Aug. 29.

- ¹ Stein, W. H., and Miller, E. G., J. Biol. Chem., 125, 599 (1938).
 ² Gross, J., J. Exp. Med., 89, 699 (1949).
 ³ Bowes, J. H., and Kenten, R. H., Biochem. J., 45, 281 (1949).
 ⁴ Neuman, R. E., Arch. Biochem., 24, 289 (1949).
- ⁵ Balo, J., and Banga, I., Biochem. J., 46, 384 (1950).

X-Ray Micro-radiography of Biological **Specimens**

A HGHLY loaded micro-focus X-ray tube^{1,2} was used for taking X-ray photographs of microscopic specimens. Engström and others3-5 have established a strong case for this kind of micro-radiography and have obtained remarkable photographs of microscopic sections in a wave-length range of 5-10 A. using a special tube with a focus of moderate size.

It can easily be shown that even with the fine focus now available $(4 \,\mu \times 40 \,\mu)$ it is advisable, in this wave-length range, to rely entirely on optical magnification of shadow-graphs and to place the specimen close to a photographic plate of high resolution. For, in the case of direct shadow enlargements, the definition can never be better than the dimension of the focus, and even for a point focus at a reasonable distance from the specimen, it is limited by diffraction to less than the resolving power of a maximum resolution plate. It is entirely different, of course, when optical elements proper are used, as discussed by Kirkpatrick and Baez^{6,7}.

The actual advantage a fine focus may provide over Engström's arrangement lies rather in the less critical spacing between plate and specimen and in the possibility of placing a monochromator between specimen and tube. This permits operating the tube at the normal accelerating voltage of 40-50 kV., where the output of long wave-length continuous radiation is high.

We cut out the short wave-length X-rays by total reflexion at an optical flat, which was set to the critical angle for the wave-length required. It was slowly oscillated in its plane during exposure to avoid the striations⁸, which are due to imperfections of polished surfaces.

The accompanying reproduction shows a microscopic enlargement (\times 1,000) of diatom fragments taken at a wave-length of about 7.5 A. in 40 min. on a Kodak maximum-resolution plate. Image points

