

## CRYSTALLIZED ACIDIC NUCLEOPROTEIN FROM PIG EYE LENSES

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Prof. Dr. H. Hanson dedicated to his 60th birthday.

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With a slight modification of the zinc-temperature-procedure which was elaborated for the crystallization of leucine aminopeptidase (LAP) from bovine lenses needle-like crystals were obtained from pig lenses (fig. 1). The filtrate from the temperature step (details are given in [1, 2]) is concentrated 10-fold by lyophilization and is dialysed against 0.01 M Tris-HCl buffer pH 8.0. During dialysis the crystals develop in small amounts in the course of 1–3 weeks. They do not have LAP activity. In the ultracentrifuge the crystal preparations are pure. Their UV spectrum shows a broad maximum with the highest values between 265 and 270 nm and a minimum at 247.5 nm (fig. 2). Besides a protein moiety of 96–97% all analysed preparations contain ribonucleotides or RNA of about 2–3% and perhaps a small amount of deoxyribonucleotides or DNA below 0.5%. The

crystals are soluble in 0.01 N and 0.1 N HCl resp. but are insoluble in 1 N HCl. A solution also takes place if a crystal suspension in 0.01 M Tris-buffer at pH 8.0 is frozen and thawed again. From this solution a re-crystallization is visible sometimes only after weeks. Obviously it is a protein which tends to flocculate and precipitate and which is difficult to crystallize.

The first crystals were obtained in 1964 (see also [3]). I publish these results now because recent research has shown that the protein component is an



Fig. 1. Crystals from pig lenses. Magnification: 80-fold.

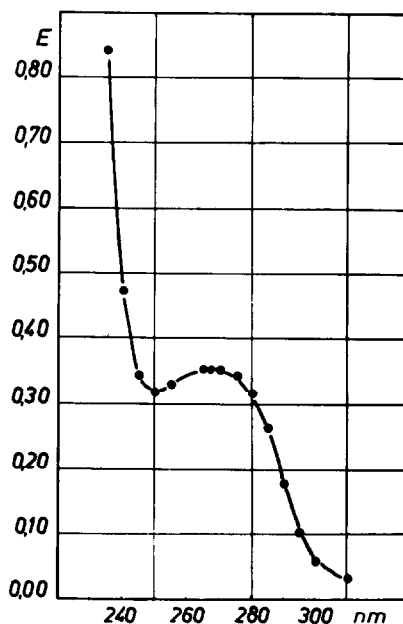


Fig. 2. UV spectrum of the pig lens crystals, solubilized in 0.01 N HCl.

acidic protein [4]. The possible importance of acidic proteins in gene regulation is now under discussion.

With the above procedure we also obtained crystals from sheep lenses which have the same form and the same solubility behaviour as the crystals from pig lenses and which is known from nucleoproteins.

The attempt to isolate similar crystals from bovine lenses in the same way was not successful. But strict control of the LAP-crystal-sediments demonstrated that during the regular LAP-crystallization-procedure in addition to the LAP crystals single beam-like crystals appear. In these crystals, which are obtained only in small amounts and are difficult to isolate, a small quantity of ribose was demonstrated.

Up to now it is impossible to state whether the isolated crystallized substances represent nucleo-

proteins which are formed *in vivo* or whether the protein and nucleotide material is complexed together randomly during the purification procedure. Further results and details will be given elsewhere [5].

## References

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- [2] H. Hanson, D. Glässer and H. Kirschke, *Hoppe-Seyler's Z. Physiol. Chem.* 340 (1965) 107.
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- [5] *In preparation.*