ASPECTS OF PROTEIN SYNTHESIS

IN THE

HAIR FOLLICLE

A thesis submitted by

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SUMMARY

There have been few investigations on the mechanism of protein synthesis in the hair follicle although such information is fundamental to an understanding of the processes of hair growth. This thesis reports investigations in vivo and in vitro on the synthesis of proteins in the hair follicle of the guinea pig.

Cellular and cell-free systems were prepared from follicle tissue that was harvested by the wax-sheet method. Attempts to separate distinct fractions of subcellular components like nuclei and mitochondria were only partly successful. However, ribosomes were isolated from follicle tissue for the first time. Their physical properties were characteristic of ribosomes from other mammalian tissues. Aggregates of two to fifteen ribosomes (polysomes) were separated from follicle tissue by sucrose gradient centrifugation and observed in the electron microscope. It was assumed that the ribosomes were held together by mRNA because of the disaggregating effect of added ribonuclease. The authenticity and distribution of polysomes in vivo and in vitro and hence the sizes of the templates involved in the synthesis of follicle proteins have been considered.

Proteins were extracted from follicle tissue and fully-developed fibres. They gave similar band-patterns when studied by starch-gel electrophoresis. Although dilute salt solutions extracted smaller amounts of protein from follicle tissue, the starch-gel patterns were similar to those extracted by 8M-urea. These findings encourage future physico-chemical characterization of keratins in the absence of denaturing agents like urea.
A number of biochemical techniques were used to study the incorporation of labelled amino acids into specific cell fractions and the soluble proteins of follicle tissue. The results of these studies indicate that the accepted pathways for protein synthesis in other mammalian systems are also present in the hair follicle. Evidence was obtained which suggests that the mRNA molecules in the follicle are relatively stable \textit{in vivo}. There is no direct evidence from these studies to suggest that this is the only mechanism of protein synthesis in this tissue. Further, it cannot be assumed that the mechanism studied is that involved in the specific synthesis of keratins and other structural proteins.

The synthesis of proteins containing citrulline, an aspect unique to certain cells of the hair follicle, was also investigated \textit{in vivo} and \textit{in vitro}. Although free citrulline could be derived from free arginine via the urea cycle in this tissue, citrulline is not incorporated into protein \textit{de novo}. It is derived from arginine at a stage subsequent to the incorporation of the latter into a polypeptide.

The manifestation of an active ribonuclease in follicle tissue prompted attempts to inhibit its adverse effect on the incorporation capacity of this system and to locate its cellular origin. The activity of the enzyme was slightly reduced with non-specific inhibitors. From histochemical studies it was concluded that the ribonuclease was extracted from epidermis or sebum as a contaminant during the initial harvesting technique.

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REFERENCES