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Remembrance of Andreas Chrambach

The origin of the Ferguson plot

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In the late 1940s the Australian Commonwealth Scientific and Industrial Research Organization (CSIRO) decided to expand research into the production and processing of wool which at that time was Australia's major source of export income and was facing increasing competition from cotton and synthetic fibers, as well as competition for land use by beef cattle, cereals, and other crops.

Some research had been done on disease control, mineral deficiencies, and wool follicle development, but little on the basic physiology and genetics of wool growth or on the processing of wool. Partly funded by a levy on the sale of wool, six new laboratories were established for the chemistry, physics, processing, genetics, physiology, and nutrition of wool and some staff were sent overseas for postgraduate training during construction of the laboratories. At that time research degrees were not available in Australian universities.

Having graduated in veterinary science at Sydney University and joined CSIRO, I undertook courses in physiology, biochemistry, and genetics at Cambridge University before taking a PhD on the influence of the pituitary on wool growth. Hypophysectomy with a maintenance dose of adrenal steroids completely stopped wool growth, and the task was to find the pituitary hormones required to restore wool growth and to examine how far variation in secretion of pituitary hormones accounted for the several-fold variation in wool growth that occurred in the field from genetic, nutritional, and environmental causes. At that time it was not believed that there was species specificity in the various recognized pituitary hormones.

This task was continued after I returned to Australia in collaboration with my colleagues Alan Wallace and Hans Lindner. Hans examined the effect of steroids on wool growth while Alan and I searched for methods to monitor the fractionation of sheep pituitary glands. We tried paper electrophoresis and ion exchange chromatography before starch gel electrophoresis, soon after its introduction by Smithies [1]. We examined standard preparations of known pituitary hormones and found they were species-specific in electrophoretic mobility, contrary to the widespread belief. We also observed that there were multiple active forms of most of the recognized pituitary hormones. Crude extracts of pituitary glands showed as many as 70 proteins mostly of unknown function. Were these proteins hormones or intracellular compounds that were not secreted? This work was reported at the 19th Laurentian Hormone Conference [2].

This paper provided the stimulus to examine the means of discriminating between different forms of heterogeneity such as differences in molecular size or charge. The relationship of electrophoretic mobility to starch concentration, temperature, and other factors were examined. This led to the formulation of the equation subsequently called the Ferguson Plot by Dr. Chrambach. This work was reported in an invited presentation to the Brook Lodge Conference on Proteins and Polypeptides [3].

The 1950s saw the establishment of the Endocrine Society of Australia and the opportunity to examine the electrophoresis of plasma fractions from clinical cases of over- and undersecretion of pituitary hormones in collaboration with Les Lazarus at the Garvan Institute in Sydney. In a sense, this was using the human as an experimental animal for research on the sheep! However, clinical conditions caused by over and under secretion of pituitary hormones had not been explored in the veterinary and animal science literature. Plasma from acromegalic patients were examined by electrophoresis and showed multiple components not seen in normal individuals [4].

Progress toward our major objective of finding the role of the pituitary in the regulation of wool growth was reviewed by Ferguson *et al.* [5]. Progress was necessarily slow since it took months to examine the wool growth response to a single pituitary protein. Further research on the electrophoresis of pituitary hormones was interrupted by the discovery that the effect of nutrition on wool growth was largely determined by the extent to which dietary protein was deaminated in the rumen which lead to the development of means of controlling this process. These findings had implications for measuring the response of wool growth to pituitary hormones and were later summarized [6].

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I retired from active research in 1972 and was unaware that Dr. Chrambach and his associates subsequently used the equation in my 1964 paper [3] to investigate the properties of a large number of proteins, other polymers, viruses, and intracellular particles as well as their performance in a variety of gel media. He helped lay the foundation for the establishment of the journal *Electrophoresis* and the formation of a new scientific discipline from a technique originally established to characterize pituitary hormones.

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