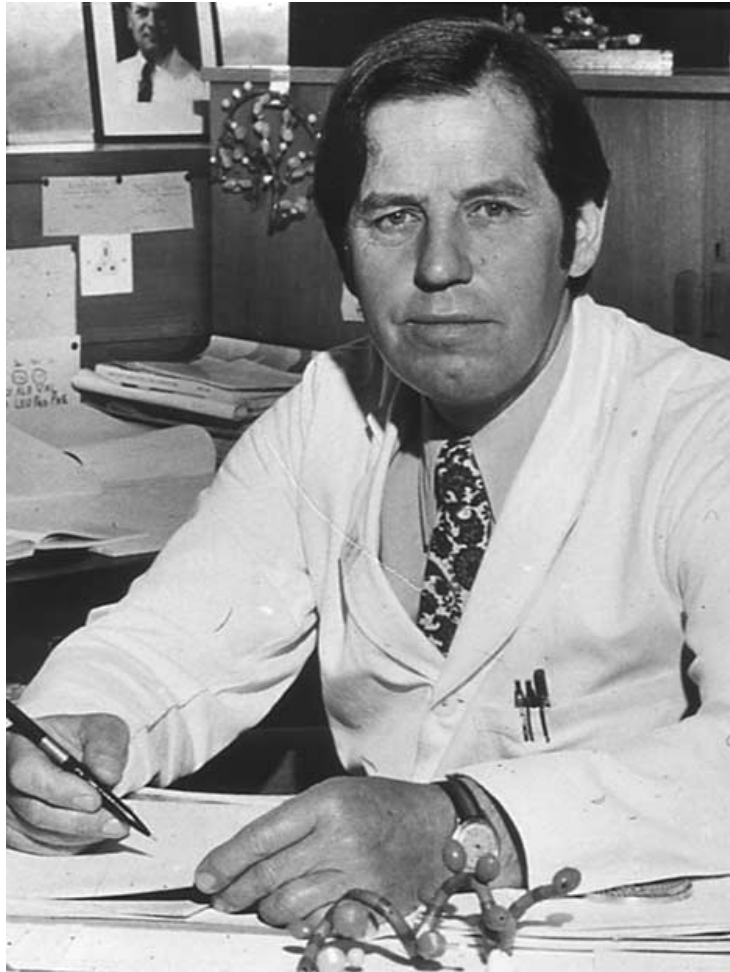


## Professor A.G.E. Pearse (1916–2003)



Professor Anthony (“Tony”) Guy Everson Pearse, a pioneering histochemist and a founding member of the Editorial Board of *The Histochemical Journal*, died in South Molton, Devon on 24 May 2003, aged 86. He inspired innumerable scientists, first with his great book, *Histochemistry: Theoretical and Applied*, and later with his hypothesis on the common embryological origin of nerves and cells producing peptide hormones (APUD cells).

Tony Pearse was born in Kent on 9 August 1916, the son of an army officer. He was educated at Sherborne School, Trinity College Cambridge and St. Bartholomew’s Hospital, London. He qualified as a pathologist, served in the Royal Navy as a Surgeon-Lieutenant during the Second World War, and then began his scientific career in the Department of Morbid Anatomy (later Histopathology) at the Hammersmith Hospital, which was attached to the Postgraduate (later Royal Postgraduate) Medical School of London, where he stayed until his retirement in 1981.

At that time, histopathological diagnoses relied mainly on Haematoxylin and Eosin staining of paraffin

sections with the addition of a few standard “special stains” such as the periodic acid–Schiff (PAS) reaction for carbohydrates and Perls’ method for iron. Pearse believed there was more than this to the study of cell chemistry. Inspired by earlier workers such as Lison, he investigated, tested and improved published methods for demonstrating cell constituents *in situ*, particularly enzymes. These studies culminated in the first edition of his textbook, published in 1953. This was quickly translated into Polish, Russian and Spanish and with its clear explanations and easy-to-follow recipes it was enormously influential, worldwide. The second edition came out in 1960, and a third, in two volumes, in 1968. The subject expanded so much that the final edition was in 3 volumes, co-edited and multi-authored. The first volume of the 4th edition was published in 1980, the second in 1985 and the third, in 1991.

By the mid-fifties, Pearse’s progressive attitude towards histopathology had earned him a Readership in Histochemistry, a tiny office off the departmental secretariat and a separate laboratory of one-and-a-half rooms on the second floor of Hammersmith Hospital. Here he accommodated an international community of aspiring histochemists who came to learn and experiment. Phosphatases, esterases, dehydrogenases, oxidases and other enzymes, proteins, carbohydrates, lipids, nucleic acids – if any of these was present in a cell it could be localised, and its appearance in normal tissues compared with that in pathologically or physiologically altered states. The crowded conditions encouraged communication and many lasting friendships were formed among his colleagues. The available space was diminished by the presence of several deep freezes and cryostats necessary for the production of frozen sections for enzyme histochemistry. The prototype cryostat, later commercialized as the Pearse–Slee cryostat, was affectionately known as “Wheezy” and was in daily use for many years. Designed by Pearse and built with the help of the excellent engineering department at the Hammersmith, it was an innovative improvement on earlier models. The microtome (a Cambridge Rocker) was placed inside a refrigerated chamber and all the controls were externalized, which helped to maintain the chamber at a constant temperature of  $-20^{\circ}\text{C}$  and to prevent frostbitten fingers. An anti-roll plate allowed sections to be flattened on the knife blade. Not least of the skills acquired by the histochemists of the time was the making of a good guide plate. All subsequently designed cryostats have followed the same principles.

Pearse was fortunate in that the expanding science of histochemistry coincided with the development of electron microscopy, which was then revealing so much about the inner construction of the cell, so that mitochondria, lysosomes, Golgi vesicles, etc. could be identified as the source of histochemical reactions. Many of the methods could be adapted for sub-cellular localization. The second edition of Pearse’s book (1963) had a six-page chapter on electron histochemistry and the third edition, 41 pages.

In 1965 Pearse was appointed to a personal Chair and became the first Professor of Histochemistry. His Department moved to more spacious accommodation on the fifth floor of the new Commonwealth Building but in accordance with Parkinson’s Law, the bench allotted to each worker soon contracted to half a bench as more and more people came to learn from and work with him. Over his 36 years at the School, Pearse’s laboratory was host to well over 200 visiting scientists from more than 40 countries. Those who came had had to work hard to win grants or scholarships to fund their time in England, and were thus already the *crème de la crème*. Very many went on to become eminent in their particular fields.

Pearse agreed readily to the proposals of his visiting research fellows, who had a wide variety of interests, matching his own inquisitive mind. Experiments were not confined to human tissues. Over the years, in addition to the usual laboratory mammals, the laboratory housed incubating hen’s eggs, and subsequently chicks, quails, terrapins, frogs and toads, fishes of many species, lampreys and hagfishes, amphioxus, sea squirts, giant African snails, pond snails, locusts, cockroaches, nematode worms and, rather more dangerously but mercifully briefly, an alligator and a puff adder. These varied creatures contributed both to basic histochemical concepts, such as muscle enzymology, and to the comparative neuroendocrinology associated with the APUD concept.

By the mid-sixties, enzyme localisation had become standard practice, diagnostically useful, for example, in muscle diseases and Hirschsprung’s disease. Pearse’s attention was then caught by the subject that was to become his second major contribution to the science of histochemistry: the chemical and ultrastructural similarities between cells producing peptide hormones. The seed of his idea were sown by the discovery of calcitonin, the peptide hormone of the thyroid gland that lowers blood calcium levels. When Pearse showed that calcitonin was produced by thyroid interfollicular cells, subsequently called C cells, he realised that these cells had common properties with other peptide hormone-producing cells such as the pituitary corticotrophs, adrenal medullary cells, pancreatic islet cells and some gut endocrine cells.

They all had high levels of  $\alpha$ -glycerophosphate dehydrogenase and non-specific esterase or cholinesterase, and characteristic electron-dense secretory granules that showed masked metachromasia and stained with lead haematoxylin. They also, in common with nerve cells, had a high content of biogenic amines such as noradrenaline or dopamine, and the ability to take up from the environment artificially supplied amine precursors and decarboxylate them to amines (Amine Precursor Uptake and Decarboxylation, APUD). Pearse's hypothesis, first put forward in 1966 and elaborated in the subsequent years, was that these similarities were due to a common origin for the two types of cell in the embryonic neural crest. Using chicken embryos as a convenient experimental model, he then set out to show the embryonic origin of the calcitonin cells from the neural crest by their APUD ability. Fortunately, fuelling the flame, cells from the neural crest were shown to migrate to the ultimobranchial bodies, the source of calcitonin in birds. Later work, by other groups, showed that most of the cells of the group did not originate in the neural crest.

The native or newly-formed amines could be identified by their wavelength-specific, formaldehyde-induced fluorescence (FIF). This could be seen diffusely in formalin-fixed, paraffin sections, but was sharply localized in sections of tissue that had been freeze-dried, exposed to formaldehyde vapour from heated paraformaldehyde and then embedded in hot wax under vacuum. Mucosubstances and antigenic for peptides were also brilliantly localized in these preparations. To facilitate the technique, Pearse put his instrument-designing skills to work again, and came up with a tissue freeze-dryer. This, too, was commercially produced (the Pearse-Edwards Speedivac tissue dryer). It was a bench-top apparatus, much smaller than conventional freeze-dryers used for lyophilising tissue. Snap-frozen tissue was placed on a thermoelectrically cooled plate and maintained at  $-40^{\circ}\text{C}$ , a temperature that, under vacuum, allowed adequately rapid sublimation of water molecules with minimal ice crystal damage within the tissue. A further instrument, a microspectrofluorimeter, was also designed, with the help of Dr (later Professor) Fred Rost, to identify and measure the fluorescence of the APUD cells.

The seventies were the heyday of the development of the APUD concept, which gave rise to an enormous quantity of work both in experimental embryology and in the definition of the diffuse neuroendocrine system with its increasing complement of regulatory peptides. Immunocytochemistry was gradually accepted as a safe and reliable way of identifying cell contents. Pearse, always a pioneer, had already begun to use this method in the early sixties to look at pituitary hormones, and then to identify gastrin cells. Now, in collaboration with the Endocrine Unit of the Department of Medicine, peptides were purified and antibodies were raised that could be used for the radioimmunoassay and immunocytochemistry of these novel hormone-like substances. In Pearse's Department, they were localized to cells in many organs, but principally along the gastrointestinal tract where they were assigned to many previously uncharacterised APUD cells. The common origin theory was strengthened by the finding that many of the newly discovered regulatory peptides were present in nerves as well as endocrine cells (the brain-gut peptides) and that a "neuron-specific" enolase could be found in endocrine cells of the series. However, it had to undergo several modifications and was finally abandoned in the light of embryological research (notably by Andrews' and Le Douarin's groups), which showed definitively that pancreatic islet cells and many gut cells were not derived from the neural crest nor even from neuroectoderm. Cell culture and mouse chimaera studies also showed that gut cells of all types, including endocrine cells, could grow from single endodermal stem cells.

Pearse's hypothesis had inspired global research and great insight into the origins and diagnosis of endocrine diseases, including multiple endocrine neoplasia, and also into the evolution of the diffuse neuroendocrine system. Peptides essentially similar to those of mammals, at least in immunoreactivity, were shown in animals from the lowest to the highest along the phylogenetic scale, but in invertebrates they are predominantly present in nerves, suggesting that the endocrine cells of the vertebrate system had their evolutionary, if not their embryological origin in the nervous system. A spin-off from the APUD studies was the discovery that single cells often produced more than one regulatory peptide. This led to the concept of cell plasticity and could be said to have laid the foundations for modern stem cell research.

Without exception, Pearse's staff and alumni held him in the highest esteem and affection. He was always approachable and never seemed to be too busy to discuss results and problems, provide explanations and suggest new paths of investigation with scientific insight, sympathy and humour. He greatly appreciated the collaboration of his visiting Research Fellows and was unfailingly generous in acknowledging it. He was honoured by many foreign universities and had friends and colleagues all over the world, some of whom

shared his enthusiasm for sailing and gardening. In his time he wrote numerous papers with enviable clarity, served as Editor or on the editorial board of many Journals and was President of the Royal Microscopical Society from 1972 to 1974. He was a kind and modest man who avoided institutional politics and whose main objective was for the work to progress in a rigorously scientific manner.

This article is reproduced with kind permission from Springer Science and Business Media". It was originally published as "Professor A.G.E.Pearse (1916 – 2003): A Tribute", by Susan Van Noorden in The Histochemical Journal, **34**; 2002, 617-619, Kluwer Academic Publishers.