Exceptional matters

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Introduction

Medical researchers in universities, research institutes and industry today are struggling to promote translational research—the catchphrase is from bench to bedside. The achievements of basic biomedical research and its potential in the postgenomic era are evident, but this Oration is in praise of clinical research. Archibald Garrod, the pioneer of clinical research of the past century, was greatly influenced by the Cambridge biologist and apostle of mendelism, William Bateson, whom I quote:

> "A word of counsel to beginners, it is: Treasure your exceptions! When there are none, work gets so dull that no one cares to carry it further. Keep them always uncovered and in sight. Exceptions are like the rough brick-work of a growing building which tells that there is more to come and shews where the next construction is to be."[1]

Garrod's Harveian Oration (1924) was entitled *The Debt of Science to Medicine*.[2] But the debt of science to medicine has in fact grown more than even Garrod might have envisaged. Fuelling this debt is the recurring theme of the recognition and elucidation of the exceptional by exceptional investigators in exceptional institutions. I have chosen examples that reflect a largely personal experience, from which astute observations of single cases or small groups of patients have been the starting point of voyages of scientific discovery—from, as it were, the bedside to the bench—but in doing so I shall illustrate the synergy between clinical and basic biomedical research that now provides medical scientists with investigative tools of hitherto unimagined power.

A rare cancer

In the 1960s, the American paediatrician Alfred Knudson, fascinated by the childhood tumour retinoblastoma, observed that whereas the sporadic form was almost invariably unilateral (and unifocal), presenting at various times in childhood, the familial form was often bilateral (and multifocal) and presented soon after birth (figure 1). Knudson deduced[3] that in the familial form there had been an inherited loss of a gene responsible for suppressing tumour formation and that during the 100 million or so cell divisions that take place in retinoblasts during differentiation, the chances of losing a second copy of this gene were sufficiently high to make the development of a tumour likely, whereas two mutations would be required to cause disease in the non-familial cases, explaining their rarity and later (unifocal) presentation. Knudson's two-hit hypothesis ignited the field of cancer genetics and heralded the era of tumour-suppressor biology. Knudson's predictions were fulfilled in a rich period of cancer research that

culminated in the cloning of the retinoblastoma (*RB*) gene in the laboratory of Robert Weinberg[4] some 14 years later, where somatic mutation of the same *RB* gene was found in the tumours of non-hereditary cases. This finding was the forerunner of the observation, now well established through the work of Vogelstein and others,[5] that patients with common cancers often have mutations in the same genes that are responsible for the familial forms.

Cancer is now recognised as a genetic disease: in most cases it is acquired as a result of a series of somatic mutations, vital clues to which have been provided by the study of familial cases. Fast-forward about 30 years from Knudson's pioneering observations: the cancer world is radically altered. The development of genomic technology has enabled the analysis and characterisation of thousands of genes in biopsy material including, importantly, archival material. Much effort is being devoted to finding gene signatures, which allow patients to be better
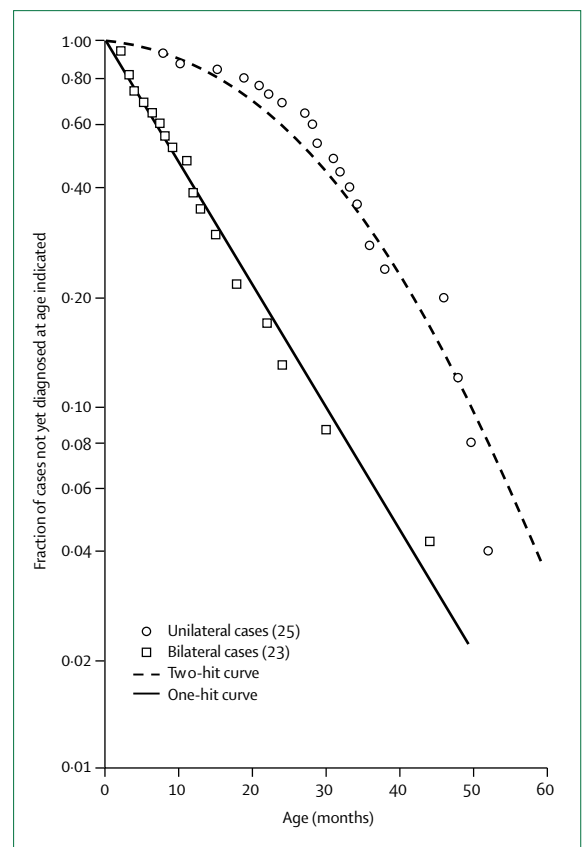

Figure 1: **Semilogarithmic plot of fraction of cases of retinoblastoma not yet diagnosed against age in months**
Unilateral and bilateral separately plotted. Kinetics of bilateral cases indicate "one hit", whereas kinetics of the unilateral cases indicate "two hits". Reprinted from reference 3 with permission of Alfred G Knudson.

characterised especially in relation to the probable prognosis and response to treatment.[6-8] The results suggest that molecular signatures do indeed predict responses to therapy. Although these early claims may have been exaggerated,[9,10] it seems likely that molecular profiling is here to stay.

Structural medicine

Max Perutz witnessed the birth of molecular haematology with the publication of Linus Pauling's classic paper in *Science* 1949,[11] describing how sickle-cell disease was due to a small change in the electric charge of haemoglobin. Vernon Ingram, working in Cambridge, showed that the alteration in charge was due to replacement of one of the 146 aminoacids in the β chain by another.[12] For many years, molecular haematology was to dominate the field of structural medicine.

A natural experiment in protein engineering

Carl-Bertil Laurell's discovery of alpha-1 antitrypsin deficiency as a cause of emphysema[13] stimulated Robin Carrell to sequence alpha-1 antitrypsin. Identification of the mutation responsible for the failure of the proteinase inhibitor to protect the lungs revealed the position of the active site that confers specificity as an inhibitor of neutrophil elastase. A similar sequence was present on another plasma protease inhibitor, antithrombin. Comparison of the two sites suggested that the specificity of inhibition depended on one aminoacid.[14] The case of a 14-year-old boy in Pittsburgh with a fatal haemorrhagic disorder, whose plasma contained an electrophoretic variant of alpha-1 antitrypsin that acted as an inhibitor of coagulation, provided confirmation. Carrell predicted and established that a methionine in the active site had mutated to arginine, changing the specificity of the inhibitor from an antineutrophil protease to an antithrombin—a dramatic natural experiment in protein engineering.[15]

Intracellular inclusions and molecular aggregates

A second example from Robin Carrell's group also relates to alpha-1 antitrypsin. David Lomas, a respiratory doctor, proposed that intracellular aggregation of the mutated molecule in hepatocytes reduced secretion of alpha-1 antitrypsin and suggested that the aggregated molecules, which could seen by electron microscopy (figure 2), were the cause of hepatocyte injury and cirrhosis, and by structural studies showed the molecular mechanisms responsible.[16] That this process might be a more general pathogenic mechanism in diseases characterised by intracellular inclusion bodies was graphically shown by a further case study. A family doctor in upstate New York insisted on an autopsy for a patient aged 56 years who died of an atypical dementia. Neuropathological examination at nearby Syracuse revealed intraneural inclusion bodies with partial sequences identical to a newly identified serpin—neuroserpin—specific to neurons. It then came

Figure 2: **Electron microscopy showing molecular aggregates of mutated alpha-1 antitrypsin molecules in hepatocytes**
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to light that the patient was a member of a family inheriting, with mendelian dominance, a trait resulting in the onset of an encephalopathy and dementia between age 45 and 60 years.[17] The similarities of the intraneural inclusions with those in alpha-1 antitrypsin deficiency led to a collaboration between the workers at Syracuse and Carrell and Lomas, who showed that every step in the pathway that Lomas had observed with the mutated alpha-1 antitrypsin was faithfully reproduced with the abnormal neuroserpin. Thus, intracellular polymerisation results in a cellular attrition, which in the hepatocyte leads to cirrhosis and in the neuron leads to encephalopathy and dementia.[18]

Triplet repeats

In 1991, Grant Sutherland, studying fragile X syndrome, the commonest cause of familial mental retardation, discovered that the disorder was associated with expansion of hereditable trinucleotide repeat sequences (CCG) giving rise to the fragile site on the X chromosome.[19] It transpired that there are several diseases characterised by triplet repeats: AGC repeats are associated with neurological disorders such as myotonic dystrophy, and in neurodegenerative disorders such as Huntington's disease the triplet repeat CAG codes for glutamine. These diseases have a striking characteristic that had no explanation in mendelian genetics: the phenomenon of anticipation, where a genetic disease becomes increasingly severe and presents earlier in successive generations. Genomic analysis provided the explanation, for in successive generations the number of repeats expands and correlates inversely with severity and age of presentation. No counterpart to these human disorders has been found in animals.[20]

Max Perutz had earlier noticed alternating positive and negative charged aminoacids in the haemoglobin of a parasitic worm, *Ascaris lumbricoides*, and concluded that the aminoacid chains had the potential to form a polar zipper.[21] He then learned of the triplet repeat expansion characteristic of Huntington's disease[22] and offered an elegant explanation of how such expansion of triplet repeats creating a long glutamine chain could, by the
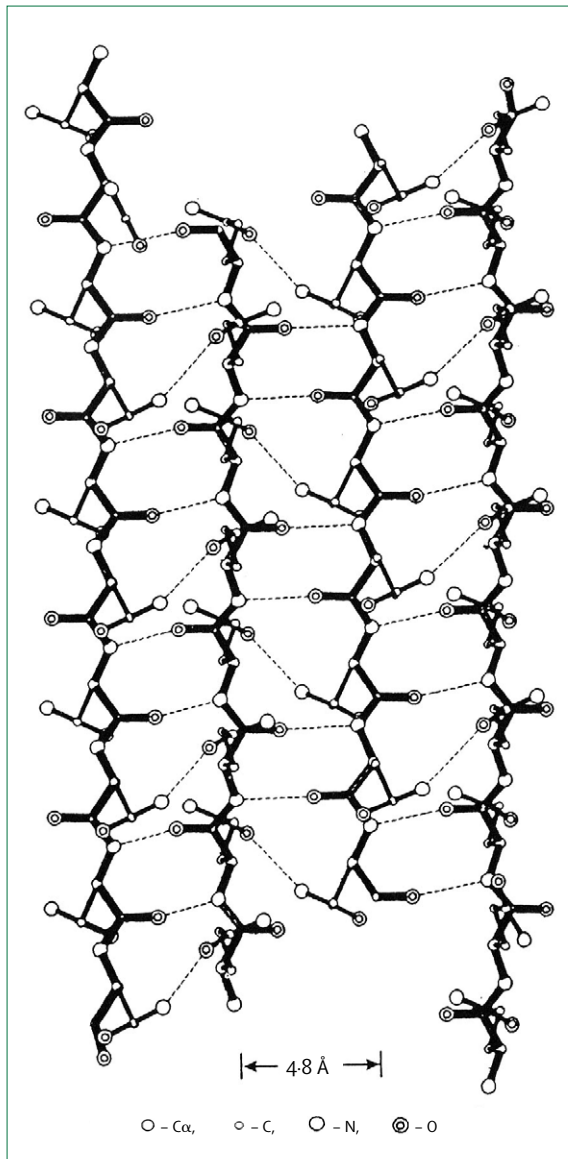
Figure 3: **Four chains of poly-L-glutamine linked by hydrogen bonds between their main chain and side-chain amides**
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action of the polar zipper, account for the formation of intracellular aggregates that characterise the disease (figure 3).[23,24]

Amyloid and protein misfolding

Mark Pepys' investigation of a calcium-dependent binding protein, the serum amyloid P component, so-called because of its binding to amyloid fibrils,[25] led to the development of a diagnostic radionuclide test for amyloid (SAP scintigraphy)[26] and allowed quantification of amyloid load in man—a requirement for evaluating and monitoring treatment—which in turn led to referral

and accumulation of patients with various forms of amyloidosis.

Rare familial cases of amyloidosis have been informative: an excellent example is a family with autosomal dominant disease where sequencing showed that the amyloid fibril was derived from a mutation of the enzyme lysozyme.[27] Lysozyme had been used as a small molecular weight (15 kDa) protein as a model for various experimental studies, notably by the late David Phillips. It was the second protein and the first enzyme to have its complete three-dimensional structure solved and has been studied extensively with respect to protein folding, especially by Chris Dobson, now in the Department of Chemistry in Cambridge, whose study of the mechanisms of lysozyme fibrillogenesis has provided a basis for the understanding of diseases brought about by protein misfolding.

Like my contemporaries of the 1960s, I had been familiar with the notion that protein-protein interactions could be pathogenic, as in immune-complex disease, but few anticipated how important a general paradigm abnormal protein folding would become. Prusiner's discovery of prions—infectious protein agents that produce their effects by inducing conformational changes in the equivalent normal brain proteins and subsequent protein aggregation—is the most dramatic example.[28]

Complement and its alternative pathway

In the 1960s and 1970s, immunology in general, and the complement system in particular, were being revolutionised by advances in protein chemistry. But one case changed thinking about the physiology of the system.

The background is that Peter Lachmann had done research on a substance known as conglutinogen activating factor (KAF) because its action was needed for conglutinin—the first described mammalian plasma lectin and peculiar to bovidae—to react with C3; the starting point was a patient in Boston (called TJ) with recurrent bacterial infections. Chester Alper and his colleagues established that abnormal breakdown of C3 in vivo was responsible for the persistent infections, and concluded from experiments in which the addition of serum or fresh plasma (but not purified C3) restored complement function and reduced C3 breakdown that there might be an inborn deficiency of a protein necessary for C3 stability in vivo and in vitro.[29,30]

What was the nature of the deficiency and how was complement being activated? That there might be alternative pathways of complement activation had indeed been suspected in the 1950s,[31] but the ideas had fallen into disrepute until the discovery of rare people (and animals) with deficiencies of C2 (or C4) whose serum still supported complement activation by gram-negative bacteria. Peter Lachmann, in collaboration with Chester Alper, showed complete absence of KAF and then, by depleting normal sera of KAF, that a pattern of activation identical to that of TJ could be produced.[32] Peter Lachmann

then proposed that what was by now termed the alternative pathway of complement was normally ticking over at a slow rate kept in control by KAF and that deficiency or depletion of KAF led to uncontrolled activation of the pathway: KAF was then renamed C3b inactivator or factor I. It was later shown by Doug Fearon that physiological activators of the alternative pathway acted by preventing complement regulators, such as factor I, and another inhibitor, factor H, from exerting their effects.[33]

Lipodystrophy and adipocytes: from immunity to metabolism and back

Had TJ not existed the intricacies of the alternative pathway would almost certainly have emerged from the investigation of another extraordinary clinical syndrome, this time not due to a single gene defect.

Study of sera from a rare form of glomerulonephritis, which histologists had termed membranoproliferative glomerulonephritis (MPGN), revealed a form of complement C3 activation not involving the classic pathway associated with a circulating factor, which caused C3 breakdown when added to normal human serum (the so-called nephritic factor, Nef).[34,35] It was later shown that Nef was an autoantibody that blocked the action of the alternative pathway inhibitors factors I and H, so that the normally labile enzyme that breaks down C3 is stabilised, thereby leading to excessive C3 consumption.[36,37] An identical system of complement activation was also found in patients with the rare disorder partial lipodystrophy (PLD), itself known to be associated with MPGN.[38,39] These clinical associations indicated that this peculiar form of autoimmunity seemed to predispose to MPGN, PLD, or both, but the underlying mechanisms, particularly of adipocyte destruction, were obscure. Only lately, by genetic technology, has it been shown in mice that knockouts of another complement regulatory factor, factor H, which physiologically inhibits the alternative pathway, develop MPGN, establishing beyond doubt the primary and causal role of unrestrained alternative pathway activation in this disorder.[40]

But what of the adipocyte? The clue came in the late 1980s with the discovery that an enzyme in adipocytes, named adipsin, was identical to an alternative pathway complement factor, factor D.[41] Elevated concentrations of factor D potentiate alternative pathway activation, and this raised the possibility that under appropriate circumstances activation of the alternative pathway, enhanced by Nef, might cause lysis of fat cells. Peter Mathieson was able to show that this could indeed take place in vitro.[42] But the finding of complement factor D in adipocytes was of more general significance, for it was the earliest unequivocal indication that adipocytes were not just storage cells for excess fat and might have other hitherto unsuspected biological functions. This leads me to the leptin story.

Leptin is the first hormone whose role has been demonstrated in the control of appetite. The beginnings were the discovery of the genetically obese mouse, the ob/ob mouse, and the pioneering parabiotic experiments linking ob/ob mice to normal mice—establishing that normal mice provided a factor that reversed the obesity of the ob/ob mice.[43] Clearly, the ob/ob mouse lacked a factor present in the blood of normal mice, which controlled food intake and fat mass. Then, in 1995, Friedman and his colleagues identified that the ob/ob mouse was deficient in leptin, a hormone secreted by fat cells that acted on the hypothalamus.[44] Early hopes that leptin might be of value in treating obesity were dashed by the finding that under normal circumstances, obese human beings have high plasma leptin concentrations and that malnourished individuals have low plasma leptin levels, reflecting in both circumstances adipocyte mass. Stephen O'Rahilly's group were the first to identify the human equivalents of the ob/ob mouse in a series of consanguineous families in whom leptin is totally absent.[45] The children had morbid obesity characterised by uncontrollable hyperphagia dramatically reversed by administration of recombinant leptin—a remarkable (albeit extreme) demonstration of the centrality of the control mechanisms of satiety to the understanding of obesity (figure 4).[46]

But an unexpected twist to the story is that leptin turns out to be an immunoregulatory hormone: ob/ob mice[47] and the leptin-deficient children have impaired T-cell function (figure 5),[46] posing the question whether the immunity deficiency associated with starvation has its origins in whole or in part in adipocyte loss and consequent hypoleptinaemia. Further, since it is now clear that adipocytes secrete various cytokines, the converse question is the extent to which the untoward consequences of a gross excess of adipocyte tissue, which include cancer and cardiovascular disease, might be explained by subtle adipocyte dependent pertubations of the immunoinflammatory system. This is an area of research that is now attracting great interest.[48,49]

From experimental models to the clinic: plasma exchange and autoimmunity

Frank Dixon[50] had established the paradigm for pathogenetic mechanisms in glomerulonephritis—namely that there were two underlying immunological mechanisms, the first mediated by autoantibodies (especially to glomerular basement membrane, GBM) and the second due to accumulation in the glomerulus of circulating immune complexes, and that these two processes were readily distinguishable by immunofluorescent examination of renal biopsy samples.

Antibodies to GBM were usually associated with a devastating disease (Goodpasture's syndrome), in which the lung basement membranes were also damaged, causing life-threatening pulmonary haemorrhage and where kidney function was rapidly lost due to a severe proliferative nephritis (rapidly progressive glomerulonephritis, RPGN). We reasoned that if circulating autoantibodies were indeed the cause of disease, then
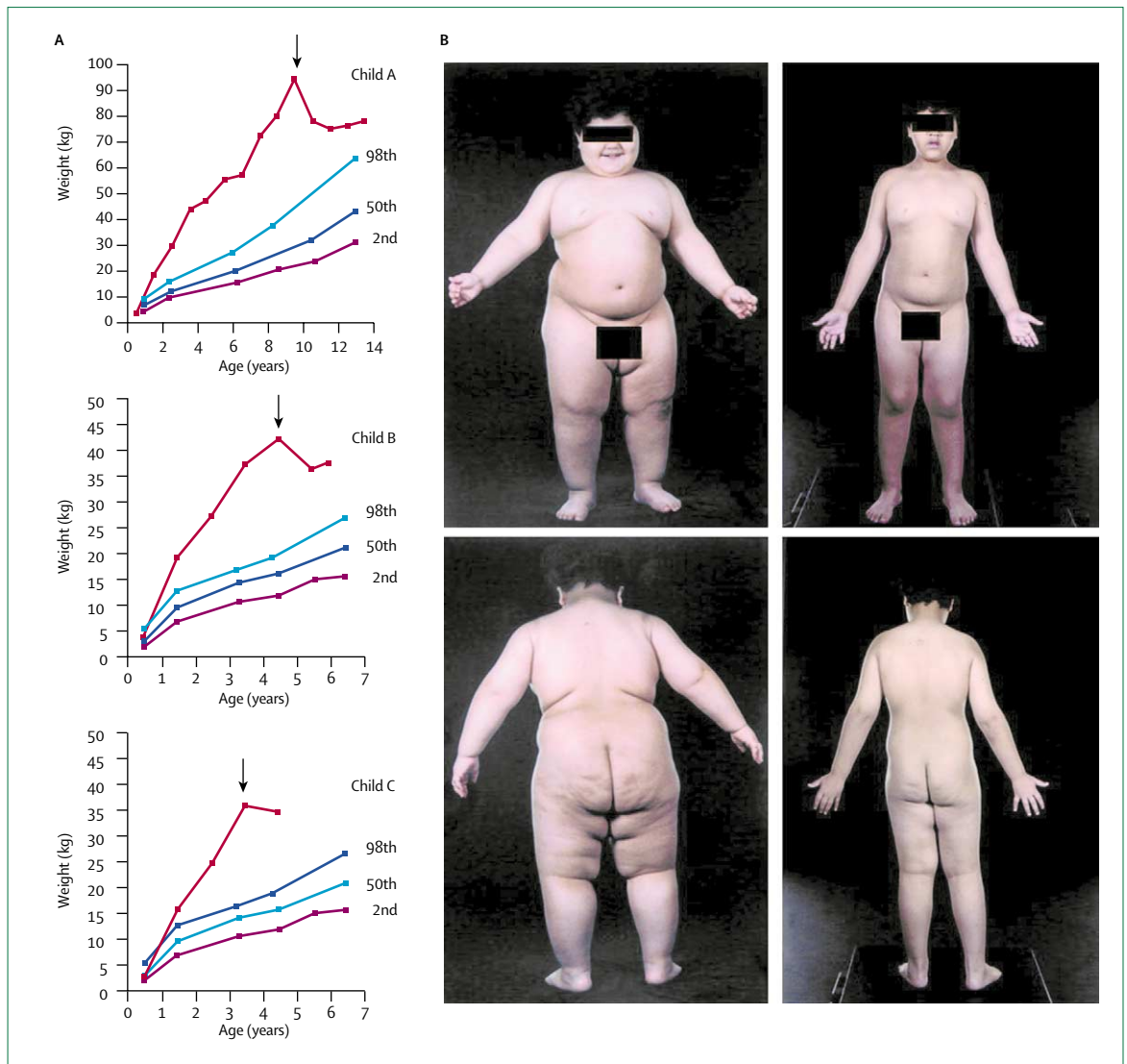
Figure 4: Effects of recombinant leptin in children with total deficiency of leptin
(A) Weights of three children compared with normal centiles for girls (upper) and boys (middle and lower). Arrows indicate start of treatments. (B) Child B before (left) and after (right) treatment. Reprinted from reference 46 with permission of the American Society of Clinical Investigation.

interventions aimed at reducing their generation—immunosuppression—combined with plasma exchange to remove antibody should alleviate the disease.[51]

The surprising finding was that antibody production was permanently arrested in some patients in fewer than a few weeks (figure 6). It has taken many years of research into immunoregulatory mechanisms to begin to throw light on how this might have been brought about. Only in the past decade has there been a convincing characterisation of regulatory T cells whose engagement (shifting the balance between activatory and regulatory T cells) could explain the termination of an autoimmune response. The subject of regulatory T cells is now a major area of immunological research—and devising immunological interventions specifically to promote the generation of immunological control systems to suppress unwanted immune responses, including immune responses to transplanted organs, is a major goal of modern immunology.[52]

Success in the anti-GBM syndromes led to a consideration of other putative diseases for which patients were not critically ill and easier to evaluate. The first of these was myasthenia gravis, then suspected (although not proven) to be due to autoantibodies to the acetylcholine receptor (AChR). In collaboration with John Newsom-Davis, we were able to show a dramatic response to plasma exchange in a patient with refractory disease (figure 7), later shown to correlate with the fall in circulating anti-AChR antibody concentrations.[53] This experiment established that circulating antibodies to

AChR were indeed responsible for the neuromuscular dysfunction characteristic of myasthenia gravis in man, and showed the use of plasma-exchange in determining whether circulating autoantibodies were pathogenic.

Pauci-immune nephritis

Anti-GBM nephritis characteristically presents as RPGN. But only few patients with RPGN have anti-GBM antibodies. Immunofluorescence of renal biopsy samples usually shows no evidence of accumulation of immune reactants in the kidney, and antigen-antibody complexes are not detected in the circulation, findings that led some to use the term pauci-immune. Such patients therefore did not match the Dixon experimental model, and the rationale for treating this group of patients with immunosuppressive drugs or plasma exchange was flimsy. Surprisingly, the response to the regimen of plasma exchange and cytotoxic drugs was better than that of patients with anti-GBM disease.[54] What might be the explanation? It was not until 1982 that a circulating autoantibody (whose significance was not immediately appreciated)[55,56] was detected in plasma of this category of RPGN patients, which confusingly was directed to intracellular antigens in neutrophils—antineutrophil cytoplasmic antibody (ANCA)—and later associated with Wegener's granulomatosis.[57] In due course the principal targets of ANCA were identified as myeloperoxidase and antiproteinase 3; ANCA tests proved to be of great diagnostic and practical value in monitoring therapy.[58] Further work showed that the antibody would activate neutrophils and make them potentially capable of enhancing tissue damage.[59] However, it was still not clear whether ANCA was cause or consequence of the nephritis or vasculitis. Resolution required the technology of the late 20th century, in which the gene for myeloperoxidase (MPO) was knocked out in a mouse: the knockout mice were immunised with mouse myeloperoxidase to which they made antibodies, and these antibodies were then transferred to normal mice (which had MPO in their white cells) and necrotising vasculitis and nephritis was produced.[60]

It has thus required three decades of research, moving from bedside to the bench and from man to mouse, to elucidate the scientific basis of an empirical therapeutic discovery; and in doing so a new paradigm of renovascular immunopathology was established, namely that autoantibody activation of neutrophils is the central pathogenetic mechanism in this important group of disorders.

Monoclonal antibodies

Monoclonal immunoglobulins were first recognised in patients with myeloma. Much of our knowledge of immunoglobulin structure, function, and genetics comes from the studies of myeloma proteins by Kunkel,[61] Putnam,[62] and others, an earlier example of knowledge transfer from bedside to laboratory. This work also
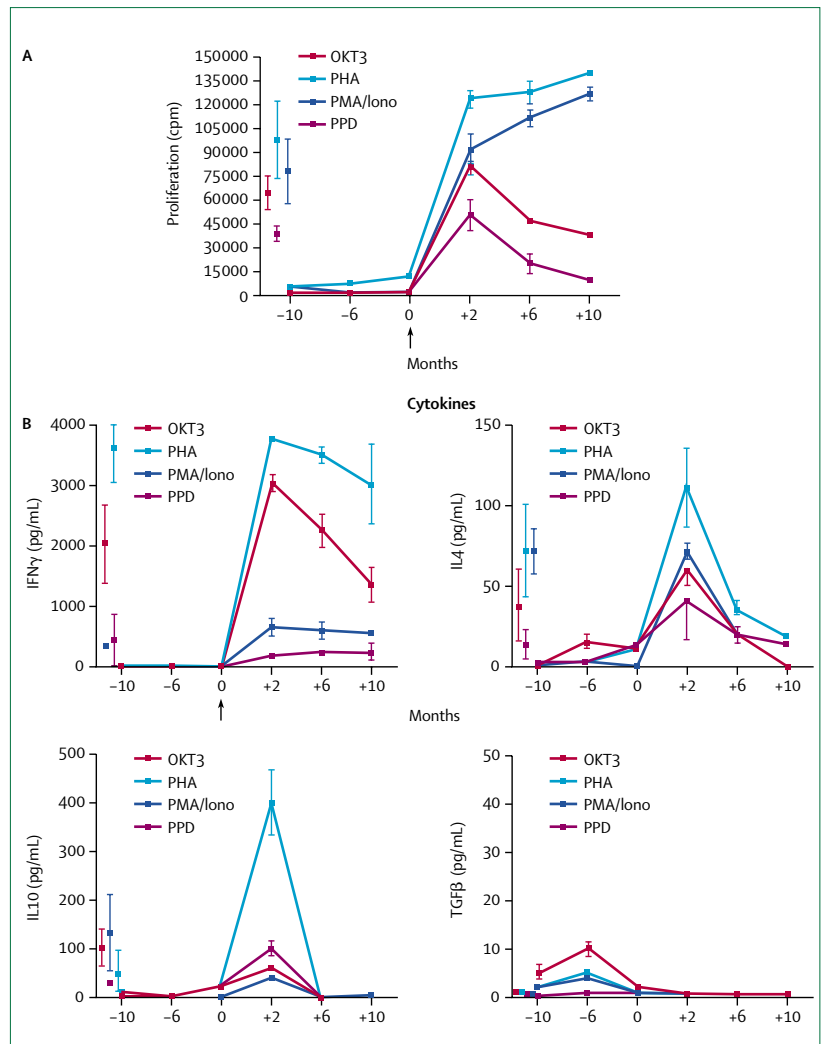

Figure 5: **Effects of leptin on assays of cell mediated immunity**
(A) proliferation assays and (B) cytokines interferon γ, interleukin 4, interleukin 10, and transforming growth factor β. Leptin therapy reverses the T-cell hyporesponsiveness. Reprinted from reference 46 with permission of the American Society of Clinical Investigation.

provided the basis of the technology devised by Milstein and Kohler for the production of monoclonal antibodies in mice by fusion of lymphocytes with myeloma cells.[63]

By the mid 1980s there was great interest in the potential use of monoclonal antibodies in man, in particular in their use in the control of transplant rejection and in the treatment of immunological malignancies and lymphoma. A major limitation for the treatment of chronic disease was that human beings would mount an antimurine immune response. The breakthrough was the production by Greg Winter in the MRC Laboratory of Molecular Biology of humanised antibodies in which the combining site of a mouse or rat antibody was genetically transplanted onto the framework of a human antibody, so that it could be repeatedly used in man with a greatly reduced chance of evoking an immune response.[64]
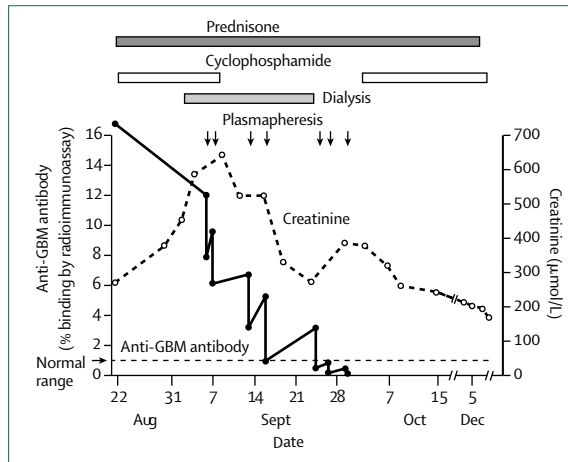
Figure 6: Effect of plasma exchange and immunosuppression on anti-GBM antibody and renal function
Autoantibody production ceased and never recurred. Reprinted from reference 51 with permission of the BMJ Publishing Group.

The potential of this approach was shown by one patient with an autoimmune disease whose condition was seemingly cured by administration of immuno-suppressive monoclonal antibodies.[65] The patient, a retired doctor with a 20-year history of refractory vasculitis unresponsive to various conventional cytotoxic and immunosuppressive drugs, was given a combination of monoclonal antibodies (antiCD52—Campath—and antiCD4) based on a protocol devised for induction of immune tolerance in mice by Herman Waldmann. Sustained remission followed and there was no vasculitic relapse. He died some years later of colon cancer: treatment had arrested the autoimmune response. Campath is now in advanced trials in an important
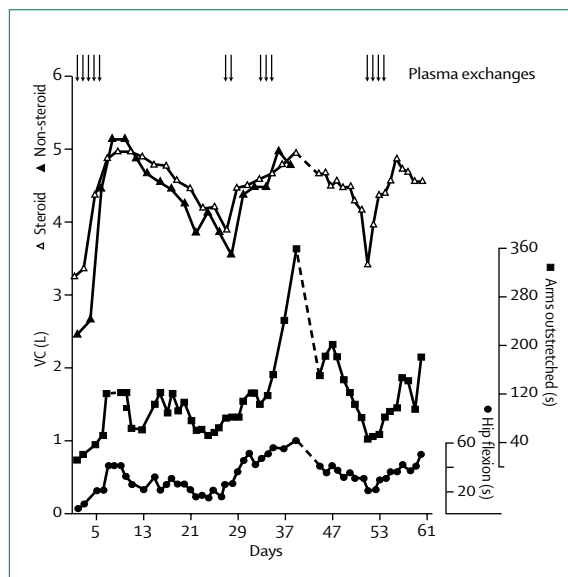

Figure 7: Effect of plasma exchange in a patient with myasthenia gravis
Improvement in neuromuscular function is dramatic. VC=vital capacity. Reprinted from reference 53 with permission.

autoimmune disease, multiple sclerosis, in a collaboration between Alastair Compston and Herman Waldmann.[66]

But even more unexpected was the dramatic benefit discovered by Maini and Feldmann of the effects of monoclonal antibodies to tumour necrosis factor α (TNFα)—this work owed its origins to the finding of high levels of TNFα in synovial tissue in rheumatoid arthritis.[67,68] Although the potential of interfering with immunologically driven inflammation by inhibiting defined mediators had been the object of research for decades, few anticipated the scale of benefit that was found clinically by Maini and colleagues and now confirmed by many others.

Antibody-based therapies now subtend a multibillion-dollar industry, with many drugs in advanced clinical trials or approved for use in man in conditions ranging from lymphoma to angina.

Academic medicine and the NHS

In the late 1980s, a small group of us moved from the Royal Postgraduate Medical School to the Clinical School at Addenbrooke's Hospital in Cambridge, driven by conviction of the need for a closer partnership between clinical and basic biomedical research, especially molecular and cell biology, which had to a considerable degree been pioneered in the MRC Laboratory of Molecular Biology on the Addenbrooke's site. We sought to link academic and service developments at Addenbrooke's, a 1000-bed hospital, the provider of hospital care to a substantial population of East Anglia. A priority was the development of a culture of clinical research, a key to which is provision of advanced medical technology including, in particular, imaging technology. A further goal was promotion of population-based medical research, and with the help of the NHS we established the Institute of Public Health, incorporating the MRC Biostatistics Unit, the University department, and NHS public health groups.

There is a pleasing development in epidemiology: the gap is narrowing between the intensively investigated hospital patient and the study of large-scale populations. Medical technology, through the use of such techniques as automated biochemistry, genomic technology, imaging, and endoscopy, allows research to be undertaken on populations that not so long ago could only be done on small groups of patients in a clinical investigation unit. The well-worked-up patient is being replaced by the well-worked-up community!

We have constructed laboratories to enable clinical and basic biomedical scientists to work side-by-side, a brain-imaging centre, a clinical research facility that has revitalised clinical research, a clinical research unit for a major pharmaceutical company, and a laboratory for the emerging discipline of genomic epidemiology. Of paramount importance has been the culture change at Addenbrooke's NHS Trust, reflected not least in a plan developed in conjunction with the MRC and the

University for a biomedical campus with the aims of improving clinical services, providing outstanding facilities for teaching and research, and maximising the wealth-generating opportunities the NHS offers.

The theme of health and wealth generation has been given momentum nationally by the publication and government acceptance of reports by the Academy of Medical Sciences[69] and the Biotechnology Innovation Growth Team,[70] chaired respectively by Prof John Bell and Sir David Cucksey. The Academy Report emphasised the need for what it termed experimental medicine, the detailed study of patients by state-of-the-art techniques, encompassing the latest imaging modalities and appropriate biochemical, genomic, and proteomic technologies. For the biotechnology and pharmaceutical sector, experimental medicine can facilitate target choice and accelerate drug development. The pharmaceutical and clinical academic communities are concerned that the infrastructure for this type of medical research urgently needs to be strengthened.

Exceptional opportunities: the role of the clinical investigator

Over the past 25 years, many—including influential leaders of western medicine—have expressed concern that the clinical investigator is an imperilled species.[71,72] But it may be surprising that there is not a greater spirit of optimism, considering that research tools are now of a power hitherto inconceivable: within weeks of the SARS outbreak the responsible coronavirus was identified and fully sequenced (compared with a comparable period of years for this to be achieved for HIV); brain imaging is capable of elucidating the neuronal basis of normal cognitive function and is revolutionising research in psychiatry; gene array technology is creating a new paradigm for cancer therapy.

So why do clinical academics feel so threatened? Clinical research is inherently difficult. Our scientific training teaches us to formulate hypotheses, set up experiments to test them, rigorously controlling variables so as to minimise confounding factors. Humans are genetically heterogeneous and when they are ill there are many confounding variables. The reality of much clinical research is that the starting point is a series of observations in which the astute clinical observer notices something exceptional or, more usually, has technology that is being harnessed for the first time in a clinical setting.

As a member of peer-review research committees, I have often observed the destructive criticism of clinical research applications, usually by clinical peers with a seemingly infinite capacity to identify problems over which the hapless would-be researcher has no control and dismissing worthwhile exploratory proposals as fishing expeditions. Basic biomedical scientists witnessing these deliberations can be forgiven for concluding that the only respectable clinical research methodology is the randomised controlled trial. "Research is the art of the soluble" (Medawar), so gifted clinical investigators flee to the laboratory where they are in control of their experiment by, for example, experimenting on mice genetically constituted to order. And in the laboratory, disease-oriented research is increasingly becoming the domain of non-medically qualified research workers— who require a shorter period of university education and can devote themselves fully to medical research without the distractions of clinical practice. The clinical research worker may be seen as expensive, not wholly engaged and addressing problems that are difficult and messy. Funding agencies faced with the choice of rigorous proposals for basic biomedical science versus research on patients behave predictably, but in so doing may forget that it is the real-life messy problems that need solutions.

The challenge of complexity: the need for new methodologies

Much of what I have presented is where analysis of a seemingly complex problem has revealed a conceptually relatively simple explanation, such as a genetic mutation producing functional change in an enzyme—the single gene disorders that led Garrod to his classic description of inborn errors of metabolism. Satisfyingly, this approach often elucidates normal biological functions and new targets for treatment. But how should we approach the much commoner diseases, the diseases of maladaptation, where our genetic constitution relates uneasily to the changed environment in which most of us exist? Examples are obesity and diabetes, high blood pressure, allergy, and autoimmunity, for which the combined individual small effects of many genes are the basis of susceptibility.

I shall use the example of insulin-dependent diabetes, a disease caused by the autoimmune destruction of islet cells. Early hopes that predisposition to this disorder was attributable to a fairly small set of HLA genes were soon dashed. John Todd's *Nature* paper[73] elucidated the small contribution of one gene, that coding for CTLA4. This publication involved more than 10 000 people in 16 centres, 53 authors, who included geneticists (of mice and men), immunologists, computational biologists, and specialist statistical geneticists supported by a 5-year programme grant costing more than £20 million!

Problems of this complexity pose an exceptional challenge. Biological complexity is now being addressed in leading universities under titles such as systems biology or integrative biology, with computational biology as its principal methodology. Paul Nurse, in his Harveian Oration *Great Ideas in Biology*,[74] discussed ways in which the complex molecular interactions of the cell might be transformed into logical informational structures and processes more familiar to those in disciplines such as mathematics and physics. Data are being generated on an unprecedented scale by techniques such as gene arrays and the various "omics"; this in turn is driving the

development of new statistical and computational methodologies. There are also vast largely untapped datasets in the patient records, to which, in due course, will be added the output of initiatives in genetic epidemiology. Substantial resources, extensive collaboration, long-term commitment, and in particular, removal of cultural barriers or funding mechanisms that discourage interdisciplinary and interinstitutional activity, are required.

Where does this leave the doctor scientist? My response is—centre-stage. The medically qualified research worker has one outstanding advantage: there is no system of university education that teaches human biology as effectively as a medical school does. But we need to do better by present and future generations of medical students. Of concern is the tendency for teaching and research to become distinct activities: research is increasingly undertaken in purpose-built laboratories, entry to which is restricted (not least because of animal terrorists). Medical students, who by-and-large are exceptionally gifted, can reasonably expect to have contact with the best researchers in their medical schools, as I did nearly half a century ago. But the competitive pressure of research and of the Research Assessment Exercise means that many university staff, especially those wholly funded by research grants, believe they cannot afford to spend time teaching. Several UK medical schools are now developing MB/PhD programmes to provide exceptional students an early opportunity to engage in research before the constraints of professional training occur. It is clear from the Cambridge programme, which started in 1990, that a majority of its graduates are committed to academic careers and are better prepared to address the complex issues ahead.

I remain optimistic. The UK government is committed to substantial investment in the science base over the next decade. Medical science is one of the country's great academic strengths and we have powerful and research intensive pharmaceutical, biotechnology, and health-care industries. Most importantly, the role of the NHS in medical research is recognised, and the government's endorsement of the Academy of Medical Sciences and the Biotechnology Innovation Growth Team reports through setting up of the Clinical Research Collaboration,[75] is of great international interest.

But I must leave the last words to my distinguished predecessor, Clifford Allbutt, Regius Professor of Physic 1892–1925. I quote from his Harveian Oration (1900):

"It was in Padua that medicine, long degraded and disguised, was now to prove her lineage as the mother of natural science, and the truth of the saying of Hippocrates, that to know the nature of man one must know the nature of all things."[76]

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