Preface

I keep six honest serving-men
(They taught me all I know);
Their names are What and Why and When
And How and Where and Who.

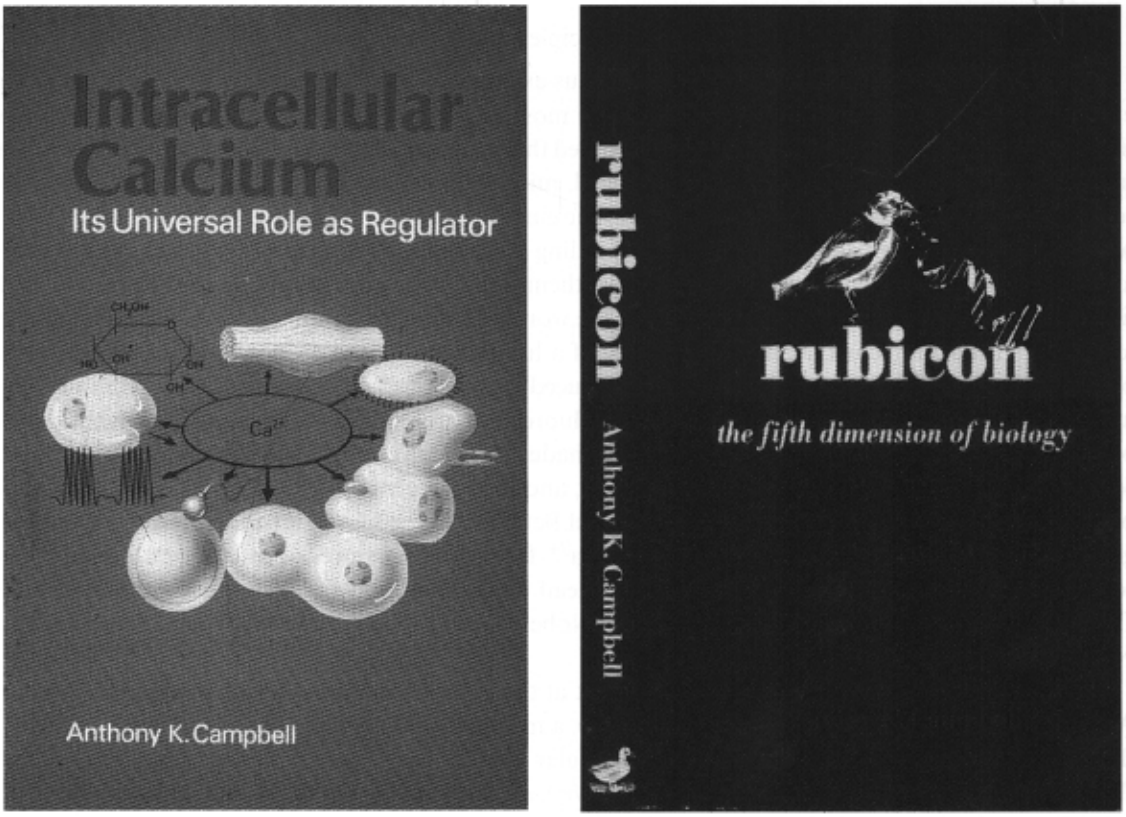
– Rudyard Kipling, *Just So Stories* (1902)

The story of intracellular calcium is a marvellous example of how the curiosity of thousands of scientists has led to an understanding of one of the most important regulatory systems in the whole of life – calcium inside cells. This curiosity has catalysed the ingenuity of scientific inventors, who have given us a wide range of molecular, electrophysiological, microscopical and imaging techniques, which have revolutionised biological and medical research. The curiosity about an apparently humble cation, Ca^{2+}, has also led to major breakthroughs in understanding killer diseases, such as heart attacks and strokes, and the consequent development of drugs to treat them. This, quite surprisingly, has produced multimillion dollar markets, with enormous benefits to the world economy and the creation of high-technology jobs. One such example is the remarkable story of a luminous jellyfish, *Aequorea*, where the curiosity, begun by Osamu Shimomura, about how it produced a green flash when touched, has given us a key indicator for intracellular free Ca^{2+} and the green fluorescent protein (GFP). Then we have the brilliance of Roger Tsien and the huge contribution he has made, first by inventing a family of fluorescent indicators for intracellular Ca^{2+}, synthesised chemically, and then the genetically engineered Ca^{2+} indicators based on GFP. The major contribution of Michael Berridge, in the search for the intracellular messenger inositol trisphosphate (IP_3) which releases Ca^{2+} from internal stores, is another example of how scientific curiosity, judgment and persistence can lead to a major discovery. Yet, interestingly, although Osamu Shimomura and Roger Tsien shared the Nobel Prize for Chemistry in 2008, there has been no Nobel Prize for intracellular Ca^{2+} as such.

Some years ago I gave a lecture about my work at the Karolinska Institutet in Stockholm, Sweden. At an enjoyable supper afterwards, with his group, a member of the Nobel Committee asked me who I thought should win the Nobel Prize for intracellular Ca^{2+}. I was flattered to learn that he had used the first version of *Intracellular Calcium: Its Universal Role as Regulator* (Figure 1) to make a presentation to the committee. He was very discrete. I said that Roger Tsien and Michael Berridge were obvious candidates. But my actual answer was the two people whose pictures are in the frontispiece. Lewis Victor Heilbrunn was deceased, but Setsuro Ebashi was still alive at the time. His discovery of the first Ca^{2+}-binding protein, troponin C, and the first intracellular Ca^{2+} store, the sarcoplasmic reticulum, really triggered the explosion in the study of intracellular calcium in the latter part of the twentieth century. The Nobel Prize system is an inspiration to us all. Important as it is to recognise seminal contributions of individuals, the story of intracellular calcium highlights the problem of the prize system.

Too many people have made seminal contributions and have made major discoveries. Thank goodness for that, otherwise we might as well all give up!

There have been dozens of multiauthor books on intracellular calcium published since my first book, *Intracellular Calcium: Its Universal Role as Regulator*, was published by Wiley in 1983. In my first book, the aim was to document as well as I could the evidence that intracellular calcium was indeed a universal regulator in living systems. It led me to realise that Ca^{2+} is both a digital switch and an analogue regulator, depending on the phenomenon concerned. This is the basis of my Rubicon hypothesis (Figure 1). In the present book, my main aim is to explain how Ca^{2+} actually works inside cells and, crucially, the evidence for this. In particular, I aim to use what we have learnt about the molecular and cellular biology of intracellular calcium, to show why Nature has selected particular components for specific tasks. Why, for example, has muscle chosen to use calsequestrin in the sarcoplasmic reticulum, as its main Ca^{2+} sink, whereas non-excitable cells such as the liver use calreticulin? *Natural history* is about describing



Intracellular Ca^{2+} as a universal signal (a)

Digital versus analogue signals in living systems (b)

Figure 1 *(a)* Intracellular Calcium: Its Universal Role as Regulator *(Campbell, 1983). Front cover reproduced with permission from John Wiley & Sons. (b)* Rubicon: The Fifth Dimension of Biology *(Campbell, 1994). Campbell, 1994. Front Cover reproduced with permission from Gerald Duckworth & Co. Ltd.*

what goes on in the Universe. *Natural science* is about understanding *how* the Universe works. My aim has been to bring together these two essential approaches to scientific endeavour.

To my knowledge there are no other books on intracellular calcium written by one person. Quite a challenge! Multiauthor books provide detailed information on highly focussed topics written by world experts. A single-author book offers the opportunity to develop themes within and between chapters. It also allows the author to develop individual creativity, whilst still retaining the consensus view. Since I was a boy I have had three intellectual passions: a love of nature, natural history; an insatiable curiosity about how nature and man-made things work, natural science; and music, as a tenor, viola player and conductor. This book sings the music of intracellular calcium. Everywhere you look, smell, taste, hear and feel, intracellular calcium is involved. This book is focussed on molecular mechanisms. But, it also aims to focus on the real problems that nature has given us. What really matters is not what happens to an artificial tissue culture cell system in the laboratory, but rather how cells in nature work. Thus, throughout I have addressed the questions about Ca^{2+} signalling in the natural physiology and pathology of the cells involved. This gives us a great opportunity to enjoy and marvel at the beauties of nature.

I have tried to emphasise two key scientific principles throughout the book. First, to show how intracellular Ca^{2+} acts as a switch, to activate a wide range of cellular events, and how an analogue mechanism can be superimposed on this digital signalling process, to alter the timing and strength of the cell event. Secondly, in the tradition of Charles Darwin and Alfred Russel Wallace (note his baptism document in the church of St Mary, up the road in Llanbradoc where he was born, shows he was christened Russell with two 'l's because his father misspelt a friend's name), the molecular biodiversity of the components of the Ca^{2+} signalling system is highlighted, upon which their BIG idea of evolution by Natural Selection critically depends. These themes are a development of two of my previous books (Figure 1). *Rubicon: The Fifth Dimension of Biology* provided evidence to support the hypothesis that life, throughout 4000 million years of evolution, has depended critically on the evolution of digital events in cells, organisms and ecosystems.

Most importantly, at a cultural level, the story of intracellular calcium has revealed the beauty of molecular biodiversity throughout the animal, plant and microbial kingdoms. Yet, why is this story so poorly dealt with in schools, and even many university curricula? In fact, I have found major mistakes in school exam revision books, including in one physics book – the emphasis on potassium and not calcium in the regulation of the heart beat! As one of the founders of the renaissance, Albrecht Dürer (1471–1528), wrote 'Be guided by Nature and do not depart from it thinking you can do better yourself. You will be misguided, for truly art is hidden in Nature and he who can draw it out possesses it'. I believe this philosophy is crucial when we teach students at school and university, and when we try to communicate our work to the general public, or even politicians!

There are 13 chapters. Chapter 1 aims to arouse curiosity about what could be special concerning calcium inside cells. Chapter 2 lays down some key principles and identifies important issues about how we name things – nomenclature. Chapter 3 provides an historical overview, starting with Ringer's famous experiments on frog heart at the end of the nineteenth century. Chapter 4 discusses how we can study intracellular Ca^{2+} and Chapter 5 summarises how Ca^{2+} is regulated inside cells, so that it can carry out its unique regulatory role. Chapter 6 describes how Ca^{2+} works in cell and what is unique about the chemistry of intracellular Ca^{2+}. Chapters 7, 8 and 9 deal with the cellular events in animal, microbial and plant cells, which are triggered by a rise in intracellular Ca^{2+}. Chapters 10 and 11 relate to medical and pathological problems, first cell injury and then drugs which affect the Ca^{2+} signalling system. Chapter 12 is focussed on the evolution of Ca^{2+} signalling. There is some speculation here. But, hopefully this not too far fetched and, in any event, able to catalyse new thoughts about this fascinating aspect of intracellular Ca^{2+}. The final chapter summarises what we know and what we do not know about

intracellular Ca^{2+}. I also discuss the importance of intracellular calcium in the curricula at school and university, and why it is important for professional scientists to engage with schools and the public. We all need to show how curiosity has led to the major discoveries and inventions which have revolutionised all of our lives.

When I give talks to schools or the public I often start by asking the audience what do they think is the greatest gift that evolution has given us? Let's keep sex out of this for a minute! For me, the greatest gift is curiosity. We are the most curious organisms in this planet. I have even been labelled the 'curious Professor'. No one, except me, is going to read this book from page 1 to the end. Each chapter stands on its own, so there is some repetition between chapters. But I hope by delving into parts you will catch a little of the inspiration I have had from writing it and reading the several thousand references at the end. Please forgive me if I have left out one of your treasured publications. If you feel I have omitted a key paper, or made a mistake, do please email me (campbellak@cf.ac.uk), and I will try to add these to a web page and any further editions. The references and other supplementary material will be made available as Endnote files on www.wiley.com/go/campbell/calcium. There will be a student edition - *Fundamentals in Intracellular Calcium* - to be published 2015/2016, with supplementary material such as hands-on demonstrations for schools and lecturers.

There are many people I'd like to thank. First, my wife, Dr Stephanie Matthews, with whom I have collaborated for over 30 years, and my five wonderful children, David, Neil, Georgina, Emma and Lewis, who have been a great inspiration. My mother died before this book was completed and was a major force in my life. My sister too, Professor Caroline Sewry, who gave me some microscopy pictures for the book. Our dear, late mother Jennet Campbell gave us our musical genes and was amazed that she had produced two science Professors! I have been lucky to have collaborated with many enthusiasts, and to have had many highly able Post-docs and PhD students in my group over the past 40 plus years. Currently, discussions with two colleagues, Ken Wann and Barry Holland, have been vital. Thanks. I thank Barry Holland for his collaboration over 15 plus years and for essential feedback on Chapters 8, Tony Trewavas FRS for feedback in Chapter 9, and Ken Broadley for feedback on Chapter 11. But any errors or omissions are my responsibility. I am particularly grateful to all in the School of Pharmacy and Pharmaceutical Sciences for their tremendous support over the past ten years. I also thank all those who have worked so hard in Pembrokeshire to make the Darwin Centre such a success there, and the many members of my group over 40 years who have helped me investigate intracellular calcium and bioluminescence. Many people at Wiley have worked hard to make this book a success. I thank Paul Deards for his initial enthusiasm for the project, my editor Jenny Cossham, Sarah Tilley Keegan, my first contact at Wiley who has been so supportive and encouraging, and Beth Dufour of RSSP for her vital work on copyright permissions. I would also like to thank Jasmine Kao and Rebecca Lim at Wiley, for their involvement and support with this book. I also would like to thank Ray Loughlin, the copyeditor for the book and Aishwarya Daksinamoorty, Project Manager and her colleagues in SPi Global, who helped craft the various documents into final pages of print which you now see within these covers. I have been lucky enough to have my research funded from a wide range of sources. I thank particularly the MRC, BBSRC (formally SRC and AFRC), NERC, The Wellcome Trust, The Arthritis and Rheumatism Council, The Multiple Sclerosis Society, The British Diabetic Association, The Waterloo Foundation and The Royal Society.

Curiosity inspires, Discovery reveals.
Bon appétit.

Anthony K. Campbell
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