OBITUARY

Carl-Ivar Brändén 1934–2004

Shuguang Zhang, Alexander Rich, Joel L Sussman & Alan R Fersht

Carl-Ivar Brändén of the Karolinska Institute died on April 28, 2004, two weeks short of his 70th birthday. Carl ("Calle" to his friends and family) was born in a tiny village in Lappland in the far north of Sweden. His father was the local schoolteacher and Carl spent his first six years at school under his own father's supervision. There were only 15 children in the school, all in the same classroom, so when one age group was in session, the other pupils were studying on their own. He learned at an early age to concentrate on his work and ignore the noise around him. The village was poor and scholarly pursuit was unheard of. The climate consisted of nine months of winter and three months of cold wind; but nature was wonderful with beautiful lakes filled with lots of fish and deep forests full of berries and mushrooms and trees to climb. Carl had the marvelous experience of being surrounded by a herd of 10,000 reindeer.

Carl left home at the age of 13 with the intent of becoming a school teacher. He won a scholarship to study mathematics and chemistry at Uppsala University. While studying in the chemistry laboratory, he was invited by the acting chairman of the inorganic chemistry department, Ingvar Lindqvist, to do a Ph.D. in his group. At the time, it was unusual for a major in mathematics to work in chemistry, but since Lindqvist's research focused on X-ray crystallography of small inorganic complexes, he was eager to obtain students with a mathematical background. Carl was extremely flattered and accepted without hesitation. As Carl aptly put it, he never had a single boring day thereafter.

In 1960, Carl attended a summer school in Manchester on modern methods of X-ray crystallography organized by D.W.J. Cruickshank. At the time, determining the structure of even a simple small molecule required intense computing power. Structure factors were calculated using mechanical calculators to add precomputed sine and cosine functions listed on so-called Beevers-Lipson strips. Most people then used a homemade analog computer for Fourier summations, the Hägg-Laurent machine, where three-dimensional Fourier sums were broken down into three successive one-dimensional summations. In a series of lectures Cruickshank described how the method of least squares could be applied to the refinement of X-ray structures and also outlined his program for one of the first digital computers in the United Kingdom.

Inspired by these lectures, Carl developed a collaboration with Stig Åsbrink of Stockholm University to write a similar program for the first Swedish electronic computer, BESK. This computer had a memory of 1,000 words, equipped with a larger magnetic drum memory, and used paper tape for input and output. There was no compiler for a higher language so the whole program was written in machine code. It took six months for Brändén to write a detailed flow chart, four months for Åsbrink to write the machine code and one year for them to debug the program. Over the next ten years this program was used by the entire Scandinavian crystallographic community and was top-rated in the use of computer time for both BESK and its much improved successor FACIT.

In order to graduate, Carl had to take another course. His choice was biochemistry. This decision completely changed his plans for the future, because he realized that he could apply his knowledge of crystallography to scientifically important and intellectually stimulating problems in biology. He wanted immediately to move into the field of protein crystallography. The obvious place to learn was the MRC Laboratory for Molecular Biology (LMB) in Cambridge, UK. As Brändén himself put it, one year as a postdoctoral fellow in the laboratory of Sir John Kendrew at LMB converted Carl from a rather ignorant chemist to a devoted molecular biologist and he studied the structure and function of proteins from that point on.

LMB was in those days one of the most exciting and stimulating places for a young postdoctoral fellow. Max Perutz had built up the laboratory with an incredible ability to choose the right people and let them work independently. Perutz himself was working on the high-resolution structure of hemoglobin. Jacques Monod visited to lecture and discuss with Perutz his latest development on the theory of allosteric transitions, for which the conformational changes in hemoglobin played an important role. Francis Crick and Sydney Brenner were working on the genetic code and attracted an endless stream of stimulating visitors. Fred Sanger's group was sequencing several large proteins and interacted frequently with the structural biologists. Aaron Klug had just moved in from London and was developing his methods of optical filtering and three-dimensional reconstruction from electron micrographs. The daily interactions with the postdocs in structural biology provided Carl with a network of colleagues who later became lifelong friends—Michael Rossmann, David Blow, Lubert Stryer, Richard Dickerson, Allen Edmundson, Ken Holmes, and Herman Watson, among others.

At the LMB, Carl rapidly engaged himself in writing a computer program for the refinement of myoglobin by the least squares method. The structure had been solved a few years earlier and higher-resolution data had been collected by David Phillips at the Royal Institution in London using a newly developed linear diffractometer. John Kendrew and Herman Watson were using these data in combination with approximate coordinates of the myoglobin structure for refinement by Fourier synthesis, but better methods were needed. Brändén and Holmes together designed a program using exact constraints and a 6×6 block diagonal matrix. It turned out to be a non-trivial problem to adopt this method to available computers. Even though they were using the fastest available machines in the United Kingdom, IBM 709 and later 7090, they had to invent several tricks to reduce computing time and storage space. The final version of the program required 12 magnetic tape stations for intermediate storage.

S.Z. is at the Center for Biomedical Engineering and the Center for Bits and Atoms, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139-4307, USA (e-mail: Shuguang@mit.edu), A.R. is at the Department of Biology, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139-4307, USA, J.L.S. is at the Department of Structural Biology, Weizmann Institute of Science, Rehovot 76100, Israel (e-mail: Joel.Sussman@weizmann.ac.il), and A.R.F. is at Cambridge University Chemical Laboratory, Lensfield Road, Cambridge CB2 1EW, UK (e-mail: Arf25@cam.ac.uk).

When the program was finished they estimated that each cycle of refinement would require 16 hours of computing time on the 7090 at the IBM computing center at a considerable cost. This type of constrained refinement was well ahead of its time, but methods very similar to what Brändén and Holmes developed in the early 1960s were rediscovered by others in the late 1970s and are now used routinely for macromolecular crystallographic refinement.

In 1962, during Carl's time at LMB, Crick, Watson and Wilkins won the Nobel Prize in medicine and Perutz and Kendrew won the Nobel Prize in chemistry. A celebration party was arranged at Crick's home, The Golden Helix, quite memorable for the fireworks arranged by Mark Bretscher. During the party, Perutz made a speech. He said that the reason why he and Kendrew had succeeded where others had failed was that they had been so ignorant about crystallography that they did not know that it was impossible to solve a protein structure. As Carl remarked later, "knowledge is fine but too much is inhibitory for breakthrough science."

After returning to Uppsala in 1963, Carl formed a long-time collaboration with Hugo Theorell and Åke Åkesson of the Karolinska Institute, who were biochemistry experts on alcohol dehydrogenase (ADH). With an equipment grant from the US National Institutes of Health, Brändén took up the structural determination of this enzyme. Awaiting lab facilities, Eila Cedergren-Zeppezauer, Carl's first graduate student, obtained the ADH crystals in her kitchen refrigerator. After ten years of active pursuit, the structure was solved. Brändén and his colleagues eventually postulated a detailed mechanism of action based on structure determinations of a number of complexes in two different conformations.

In 1966, Carl attended the first international symposium on methods of protein crystal structure determination at the Hirschegg ski resort in the Austrian Alps. The meeting was organized by Max Perutz and Walter Hoppe from Munich and had attracted 16 participants who comprised almost the entire protein crystallography community. It was an ideal size for intensive discussions on developing novel methods, both experimental and theoretical. In 1969, at a meeting in Konstanz, Germany, Michael Rossmann compared his structure of LDH with Brändén's ADH. To their delight they could identify similar features in parts of the subunits. Although no one believed their structures at the time, they had in fact discovered a new protein fold. They showed that even when there is no significant sequence identity, there could be a similar structural fold, which Michael Rossmann called a "molecular fossil", that is common in a class of functionally similar enzymes.

In collaboration with Martin Karplus, Carl's group studied the dynamics of ADH domain rotation using computer simulations and found no significant energy barrier in rotating the domains from one form to the other. Meanwhile one of Hugo Theorell's students, Hans Jörnvall, determined the amino acid sequences of several different alcohol dehydrogenases. Carl also had frequent long-term visitors from outside Sweden who were engaged in chemical or biochemical work on ADH in their home laboratories. The longest-lasting and personally most rewarding visitor was the biochemist Bryce Plapp from the University of Iowa. These extensive collaborations and friendships provided Carl with the wonderful opportunity to explore ADH structure and function from many perspectives.

Carl always liked to face grand challenges. In the late 1970s he took on an ambitious project to solve the crystal structure of Rubisco, an enzyme that catalyzes the initial carbon dioxide fixation in bacteria and plants, after reading a report from the US National Academy of Sciences. He first recruited Inger Andersson, a biochemist from Michael Zeppezauer's group in Saarbrucken, and later a graduate student, Ylva Lindqvist, whose father was Carl's Ph.D. thesis mentor. He also recruited Gunter Schneider from Hans Eklund's lab as a postdoctoral fellow. He collaborated with George Lorimer then at DuPont (Wilmington, Delaware, USA), an expert on the biochemistry of Rubisco, and obtained grams of purified Rubisco protein. Within a few months he had obtained crystals and in 1986 they published the X-ray structure of this bacterial enzyme. This structure became a turning point in the studies of Rubisco. Not only did it provide the first detailed picture of the active site of Rubisco, but it also provided the fold of the large subunit of all Rubisco molecules, which subsequently aided structural and functional interpretation of all Rubisco enzymes. In collaboration with Stefan Knight, Staffan Normark and Scott Hultgren, Carl determined the crystal structure of a bacterial chaperone protein, PapD, when the study of molecular chaperones was an emerging field.

In the mid 1980s, when protein engineering started to become a common tool in biochemistry and molecular biology, Carl realized that protein structures would not only become increasingly important, but also would be transformed from a pure academic pursuit to an important tool of molecular life sciences and beyond. Without a deep understanding of the principles of protein structure, protein engineering would be reduced to tinkering. In the midst of a lot of activities, he approached John Tooze of the European Molecular Biology Organization to be co-author of a textbook on protein structures. This beautiful book, *Introduction to Protein Structure,* with elegant hand-drawn (rather than computer-generated) illustrations and astonishing clarity has had a tremendous impact on molecular biologists, and especially on graduate students and those who wish to know more about structural biology but do not have any background in the field. Robert Gallo, the co-discoverer of the HIV virus, was so fond of this book that he once remarked at a meeting that he kept this book at his bedside and read it with great pleasure. One of us (S.Z.), while working with Alexander Rich, was attracted to the elegant book early on, and later used it for teaching a course on molecular structure of biological materials at the Massachusetts Institute of Technology (MIT). This book has stimulated numerous engineering students who had no biological background to move into biology to design new materials using the protein motifs illustrated in the book. After all, these simple molecular motifs are the diverse and remarkable structures in proteins that determine their function.

Carl maintained a keen interest in developing new methods for structural biology, a crucial part of modern biology. He realized that new technology and tools can accelerate science as a whole. He spent

five years as research director at the European Synchrotron Radiation Facility in Grenoble, France. He was responsible for establishing facilities for chemistry, biology and medicine, and managed to promote strong facilities for structural biology. He realized that this was a golden opportunity for extending the frontiers of structural biology by determining the structures of very large molecular complexes as well as collecting data from very small crystals (now down to ~10 μm^3). In addition, it was possible to study the dynamics of biochemical reactions by time-resolved crystallography in the microsecond range, obtain fiber diffraction patterns of single protein fibrils and explore novel medical applications. In the late 1990s he participated in designing the ultimate tool for structural biology, a free electron laser with femtosecond-pulse diffraction combined with three-dimensional holographic instant data collection. He was very excited to learn that a group at MIT is developing a scalable microslot probe for electromagnetic microstructure instrumentation that could complement the free electron femtosecond-pulse laser-holography system.

Carl was also keenly aware of the importance of multidisciplinary collaboration. He often collaborated with people from very different fields and promoted such collaboration on an international scale. Carl was always very warm and kind to young scientists regardless of age and background and fostered young talent, such as Alwyn Jones.

Carl's wife, Dr. Malin Åkerblom, works in an organization within Uppsala University that supports basic science projects in the poorest countries of the third world, such as Tanzania, Zimbabwe, Sri Lanka and Bangladesh. Through her work Carl had the opportunity to develop a deep insight into the problems that face science in these countries. He also had the privilege of getting to know a number of bright and devoted scientists from developing countries who struggle against difficult odds. He remarked: "From a global perspective of science policy it is a dreadful waste of human resources that the industrialized world supports mediocre scientists in large numbers while many bright young people in the Third World are deprived of the possibility to make their impact on the development of science."

Carl also spent considerable time on science policy and in the capacity of journal editor. He was one of five members of the Nobel Committee for the Chemistry Prize. He was chairman for three years of the EMBO fund committee for postdoctoral fellowships in molecular biology and often traveled throughout Europe and to Israel to interview prospective fellows. At the same time he was chairman of the Chemistry Committee of the Swedish Natural Science Research Council and a member of the board of the council as well as a board member of the Swedish Foundation for Strategic Research. He was a board member of a large consortium for functional genomics not only in Sweden but also in the United States, as well as a member of science advisory committees and evaluation committees of several institutes and synchrotron sources worldwide. Carl and Wayne Hendrickson launched the journal *Structure* in 1993.

In November 2002, Carl was diagnosed with lung cancer and he immediately resigned his activities. In typical fashion, he wrote: "I have decided that since I have devoted almost my entire life to science I also want to contribute to science towards the end of my life by participating in clinical trials based on novel compounds such as protein kinase inhibitors. I feel confident that ten years from now lung cancer will be regarded as a curable disease thanks to the steady advancement of our knowledge of the molecular biology of cancer." He also wrote: "From a human point of view it has been a fascinating experience to suddenly realize that the end of my life is in sight. Looking back, I find great comfort in the fact that I have had a full and rich life with many friends all around the world, been blessed with children and grand children and never had a boring moment thanks to my scientific activities." He was to receive a honorary doctoral degree from the Karolinska Institute on May 14, also his 70th birthday, to honor him for his lifetime achievement.

Eva Klein, a renowned immunologist at the Karolinska Institute who also discovered natural killer cells, once remarked, "I have always tried to find a great scientist, who is genuine, gentle, modest, honest and accessible. I have found the person, Carl Brändén." His passion for science, wisdom, friendship and scholarship will be missed by a much wider community for years to come.

Note: Much of this obituary is based on Carl Brändén's autobiography, which he called, "My Own Obituary, My Life in Science: from Reindeers to Synchrotrons." The autobiography will be published in Comprehensive Biochemistry ***43****: A History of Biochemistry-Selected Topics in the History of Biochemistry. Personal Recollections VIII (ed. Semenza, G., Elsevier, Amsterdam, 2004). ISBN: 0444517227.*