BOOKS: BIOCHEMISTRY

Heroic Battles of the Protein Wars

David Eisenberg

Ideas that today seem obvious were once the subjects of angry controversy. Discoveries that seemed narrow, such as the finding that one enzyme is a protein, could open scientific polemics as well as scientific vistas. This was especially true in the development of protein science, as we learn from the spirited history that Charles Tanford and Jacqueline Reynolds (a husband and wife team of emeritus biochemistry professors at Duke) offer in *Nature's Robots*. Compared with the molecules analyzed and synthesized by 19th-century chemists, proteins are enormous, complex, and honed by evolution to a vast variety of specific functions. The paths to the discoveries of their structures, syntheses, and functions were often lonely and arduous, requiring new tools and news ways of thinking.

Nature's Robots
A History of Proteins
by Charles Tanford and Jacqueline Reynolds
Oxford University Press, Oxford, 2001. 312 pp. $27.50, £18.99. ISBN 0-19-850466-7.

Take the experience of James Sumner who, as a new junior faculty member at Cornell, set out to discover the chemical nature of an enzyme. Tanford and Reynolds quote Sumner's recollections from his 1946 Nobel lecture:

> I wish to tell next why I decided in 1917 to attempt to isolate an enzyme. At that time I had little time for research, not much apparatus, research money or assistance. I desired to accomplish something of real importance. In other words, I decided to take a "long shot." A number of persons advised me that my attempt to isolate an enzyme was foolish, but this advice made me feel all the more certain that if successful the quest would be worthwhile.

Well, why not be ambitious? Sumner had already attained his faculty post despite having been told early on that his disability—he had only one useful arm—barred him from a career in chemical research. He chose urease from jack bean as the enzyme to isolate, and he followed its activity to successively fractionate "globulins" from the plant seed. After working essentially alone for nine years, he described the purification and crystallization in a seven-page paper [*J. Biol. Chem.* **69**, 435 (1926)], which documented positive tests for protein and negative tests for fats, carbohydrates, and other possible contaminants. Over the following six years, Sumner further characterized the crystalline protein in another 18 papers; these included isoelectric point measurements, antibody reactions, and digestion by proteolytic enzymes to show concomitant loss of activity and protein structure.

One might think that these findings would have been enough to convince scientists that enzymes could be identified with proteins, but hostility to that notion remained. The German Nobelist Richard Willstätter and others favored the "carrier theory," which held that a small absorbed molecule was the agent of the catalytic activity. They were fooled by their work with minute samples: These showed enzymatic activity (because of the immense specific activity, then unknown, of enzymes), but the available chemical tests failed to detect proteins within them. Willstätter was so confident of the conclusion that enzymes are not proteins that he traveled to Cornell to dismiss Sumner's idea in front of his colleagues and students. Tanford and Reynolds are at their best in describing such sharp intellectual collisions and the cataclysmic changes in thinking that followed. They note that after Sumner's discovery, "the number of distinct proteins would rise by leaps and bounds: every biochemical reaction needed a unique enzyme."

Serum separators. For his doctoral research with The Svedberg, Arne Tiselius (in lab coat) developed electrophoresis to separate proteins on the basis of their electrical charges. This photograph was taken in 1926, the year Svedberg was awarded the Nobel Prize in Chemistry; Tiselius would win the same prize in 1948.

The authors also relate an earlier dispute of monumental importance, that between the physician-scientist Gerrit Mulder and the analytical chemist Justus Liebig. Following a suggestion from Jacob Berzelius, Mulder named proteins in a 1838 paper. He adopted the Greek πρωτειοξ, which means "in the lead" or, in the authors' vernacular translation, "we're number one." Tanford and Reynolds explain that Mulder did much more than name proteins. His elemental analysis of phosphorus and sulfur in proteins revealed that different proteins had similar atomic compositions. For egg albumin, he found an elemental formula of $C_{400}H_{620}N_{100}O_{120}P_1S_1$; for serum albumin, he found the same composition but with an additional atom of sulfur. Wheat albumin had a nearly identical composition as well, and Mulder concluded that "the main mass of animal matter is delivered directly from the plant kingdom."

Liebig initially accepted Mulder's idea that all proteins are minor modifications of a single substance, but on finding a greater nitrogen content in fibrin than in egg albumin, his "admiration for Mulder turned overnight to unprincipled contempt." The resulting dispute, Tanford and Reynolds conclude, planted the idea that proteins held some secret of life. "When it turned out that every protein was different, that dozens of them could be identified, similar but distinct, physiologists began to discern a link between unique proteins and unique physiological functions."

The heroes in *Nature's Robots* tend to be analytical and physical chemists; depicted as their wrongheaded opponents are colloid chemists, numerologists, and speculative philosophers, such as Dorothy Wrinch of Smith College. However, even the reaction evoked by Wrinch's unrealistic structural models helped to advance ideas on protein stability. Tanford and Reynolds laud Theodor Svedberg for the ultracentrifuge, Arne Tiselius for electrophoretic separation, and Edwin Cohn and John Edsall for establishing that proteins are bristling with charges. They applaud the researches of J. D. Bernal and Max Perutz on three-dimensional structures, and the studies of Emil Fischer and Fred Sanger on chemical structure. The au-

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thors emphasize the huge advance achieved by Sanger's decade of work on the amino acid sequence of insulin, which showed that proteins have a definite, iron-clad chemical structure. From this, the idea of a genetic code easily followed. Almost as important was Sanger's additional feat of finding that although the β chains are identical in beef, pig, and sheep insulin, the α chains differ by substitutions in positions 8 to 10: essentially the discovery of orthologs. Could this be considered the first step in bioinformatics?

Every history book has to stop somewhere; this one finishes with interesting historical sections on the links of proteins to physiological functions and to genetics. Readers may feel deprived of hearing about the achievements of genome sequencing and our ability to enumerate nearly every protein produced by an organism. But there is so much of interest here, described with scholarship and punch, that they will enjoy *Nature's Robots* even without the genomic catharsis.

NOTA BENE: ANIMAL BEHAVIOR

Creature, Heal Thyself

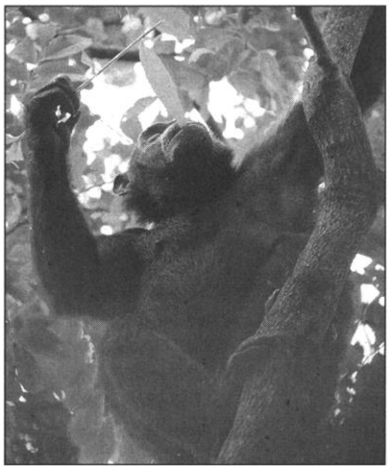
"Leave your drugs in the chemist's pot, if you can heal your patient with food," advised Hippocrates in the 5th century B.C. Living in the industrialized world, we seem to forget this advice each time we reach for the latest pharmaceutical wonder. In contrast, wild animals are experts in the art of staying well: not only do they carefully select a nutritious diet, but they also treat ailments by eating plants with medicinal properties. Indeed, watching what animals ate when they became ill helped our ancestors to concoct their herbal remedies.

Wild Health
How Animals Keep Themselves Well and What We Can Learn from Them
by Cindy Engel

Houghton Mifflin, Boston, 2002. 288 pp. $24. ISBN 0-618-07178-4. Weidenfeld and Nicolson, London, 2002. £20. ISBN 0-297-64684-2.

Cindy Engel, a lecturer in environmental sciences at the Open University in the United Kingdom, has written a fascinating account of the numerous behaviors that help animals to maintain their health and stay well in the wild. Determined not to anthropomorphize, Engel argues that the survival strategies of wild animals—seeking plants from "Nature's pharmacy" to treat illness, and altering their diets to prepare for migration, hibernation or reproduction—do not reflect innate animal wisdom, but rather are the result of millions of years of natural selection.

Plants synthesize an astonishing array of secondary compounds that fend off attack by herbivores, protect against pathogens, prevent growth of competing plant species, and attract animal pollinators. Although toxic at high doses, many of these compounds are medicinal if taken in small quantities. How do animals "know" which plants to eat to alleviate their unpleasant symptoms? Engel details the evidence for self-medication by animals, much of which comes from long-term studies of wild chimpanzee colonies in Tanzania, such as Jane Goodall's work at Gombe over the last 40 years and research in neighboring Mahale by Toshisada Nishida and Michael Huffman.



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More than ten years ago, Huffman reported on a sick female chimp that recovered her health after she sucked out the bitter pith of *Vernonia amygdalina* (a plant not normally eaten by healthy chimps), but his observation was met with skepticism. However, this self-medicating behavior was subsequently witnessed in other chimps, and *Vernonia* is used by the local Tongwe tribe as a herbal remedy. Chemical analysis revealed that the pith contains several sesquiterpene lactones that have activity against internal parasites. Goodall's group and Huffman have also observed chimps scouring their guts of parasitic worms by carefully folding and swallowing whole leaves selected from plants that they would normally ignore (see photo above). The leaves—with their rough texture, tiny hooks, and folded concertina shape—act like velcro, scraping off loose worms from the gut interior. Bears, wolves, tigers, and snow geese are also known to swallow rough leaves or grass to get rid of intestinal worms.

Many mammals (including gorillas, chimps, and elephants), some birds, as well as certain indigenous tribes eat soil with a high clay content. Such clay-rich soils not only contain essential minerals but also bind to plant toxins and stop diarrhea. We still follow this ancient practice when we take anti-diarrheal preparations containing kaolinite clay. Engel describes many other striking behaviors of wild animals that seem to be ways to maintain health: Muriquis monkeys in Brazil alter the plants they eat to regulate their fertility, so that they will reproduce when food is plentiful. Elephants and gorillas cover their dead with earth and vegetation, which hastens decomposition and reduces infection. Capuchin monkeys rub the toxic secretions of millipedes into their fur to repel biting insects.

In a delightful digression, the author reveals that humans are not the only creatures to enjoy certain vices. Apparently, many animals and birds have a strong predilection for alcohol, which they obtain by eating fermented fruit; they often overindulge and become inebriated. Engel proposes that a taste for alcohol may have been maintained by natural selection—despite the dangers associated with inebriation (accidents and predation, for example)—because alcohol is both rich in calories and a stress reducer. This attraction to alcohol can have disastrous consequences: In 1985, a herd of 150 thirsty elephants stormed an illicit still in a West Bengal village, gorged themselves on moonshine, and then pulverized seven concrete buildings in a drunken stampede. Animals also partake in certain stimulating refreshments. Coffee is reputed to have been discovered 1500 years ago when a goatherd noticed that his goats became excessively energetic after feeding on the red berries of a small shrub. Ancient Peruvian Indians, observing that their llamas chewed coca leaves when carrying heavy loads on long journeys, took up the habit themselves.

Combining scientific observations and anecdotes of wild animal behavior together with our own traditions of folklore and herbal medicine, Engel has produced an enticing, well-referenced (although poorly illustrated) narrative that should be easy for any reader to digest. She concludes this entertaining book by sensibly proposing that long-term research into animal behavior in the wild could provide valuable insights into ways to keep ourselves and the captive creatures that depend on us—pets, livestock, and zoo animals—in good health.

—**ORLA SMITH**