BIOGRAPHICAL MEMOIRS

Nicholas R. Cozzarelli

March 26, 1938–March 19, 2006
Elected to the NAS, 1989

A Biographical Memoir by Michael R. Botchan and Mark A. Krasnow

Figure 1 Nicholas Robert Cozzarelli.

Nicholas R. Cozzarelli—Nick to all who knew him well—was a leading authority and innovator in the study of the three-dimensional topological structure of DNA and chromosomes, and especially the enzymes that create and regulate it ("proteins that push DNA around"), their biological functions and medical significance, and the mathematical theory that underlies it. "With remarkable persistence and insight, he developed this area into an incredibly rich and complex field—one that yielded astonishing biological insights in ways I never thought possible," reflected Bruce Alberts, former President of the National Academy of Sciences (NAS).[1] His "blast ahead" spirit and zeal for science, family, and life magnified the impact of his transformative research and leadership roles, and made him one of the most engaging and effective educators, mentors, and colleagues, and a trailblazer in bridging scientific disciplines and promoting access to the scientific literature.

The discovery of the interwound double helical structure of DNA launched the field of molecular biology and has been taught in every basic biology class for nearly three-quarters of a century. Although less widely acclaimed, the higher order coiling, compaction, and catenation of DNA is nonetheless critical for efficient reading, replicating, recombining, repairing, and resolving the unwieldy chromosomal DNA molecules that encode the instructions for making and operating an organism. Nick Cozzarelli boldly entered this field just as the first enzymes that can alter DNA topology were being uncovered by James C. "Jim" Wang, James J. Champoux, and Martin F. Gellert and found to either uncoil ("relax") or twist up ("supercoil") DNA and dubbed DNA topoisomerases.[2]

In studies of bacterial antibiotics, Cozzarelli and colleagues demonstrated through elegant genetic and biochemical experiments that certain quinolone (such as nalidixic acid and ciprofloxacin) and coumarin (such as novobiocin) compounds directly act on topoisomerases. They and others showed that the target of nalidixic acid is GyrA and the target of novobiocin is GyrB, the two subunits of bacterial DNA gyrase, the first and as yet only topoisomerase that actively introduces negative supercoiling. The powerful combination of pharmacology, genetics, biochemistry, and mathematics employed by Cozzarelli and co-workers were seminal in elucidating gyrase's deft mechanism—wrapping the substrate DNA around its surface such that two distant DNA strands cross, introducing a break in one of them at the crossing ("node"), then passing the other strand through the break and resealing it, without ever letting go of the DNA strands or broken ends until ATP hydrolysis resets the enzyme for another round.

This strand passage mechanism inverts the topological sign ("sign inversion") of the node created by the crossing strands, explaining how gyrase curiously introduced two supercoils in each catalytic cycle. The model also explained how gyrase was able—by similarly passing strands from different DNA molecules—to catenate and decatenate DNA rings and untangle chromosomes, like the interlocked and seemingly inseparable daughter chromosomes that result from replicating double helical DNA. This ability resolved a longstanding mathematical argument against the double helical structure of DNA,[3] rendering it "ethereal" Nick would muse. The strand passage mechanism divided topoisomerases into two classes: type II enzymes, such as gyrase**,** that break and pass double-helical DNA segments, introducing (or removing) two supercoils per cycle; and type I enzymes that break and pass single-stranded DNA segments, affecting one supercoil per cycle. The analysis by Nick's team of mutants and inhibitors showed that topoisomerase-targeting antibiotics block or slow bacterial growth by altering supercoiling or blocking daughter chromosome untangling. Most importantly, they also found that some antibiotics turn the topoisomerase into a "poison" that kills the bacterium by stalling the enzymatic reaction mid-cycle, thus making permanent the transient DNA breaks, and thereby chopping the genetic material into worthless fragments. The same mechanism applies to some of the most important cancer chemotherapeutics that similarly convert human topoisomerases into "cell suicide machines" that cleave the DNA in tumor cells.[4] Mutant analysis assigned specific functions in DNA topology in the cell to individual topoisomerases and expanded understanding of the cellular roles of supercoiling and other geometric features of chromosomes.

Cozzarelli and his colleagues extended their multidisciplinary approach to elucidate the mechanism of other enzymes that affect the higher-order structure of DNA, such as: recombination enzymes that they discovered tangle their substrate DNA into knots and catenanes in the process of rearranging and restoring its continuity**;** condensins that compact and organize chromosomal DNA during cell division**;** and**,** bacterial FtsK, which pumps newly replicated DNA from one part of the cell to another in preparation for chromosome segregation. They developed advanced techniques for characterization of DNA and chromosome structure, including methods for assigning the absolute stereostructure of DNA knots and catenanes, and they formalized the mathematical basis for using such information to infer dynamic events in an enzymatic mechanism and to provide fresh insights into the dynamics of DNA and chromosome structure in the cell. Nick's science "was always original and impeccable."[5]

Nick was born on March 26, 1938, in Jersey City, New Jersey, and his life typifies an American success story. His parents were immigrants from Castelnuovo di Conza in southern Italy. His father was a shoemaker and his mother edited telephone books, and they encouraged their son to take seriously the advantages of study. Nick did so and earned admission to Princeton University on a full scholarship, intending to prepare for a career in law. His curiosity and thirst for fundamental knowledge developed as his career path transitioned from classical professional trades to scholarship. He became fascinated by science and graduated *magna cum laude* with a bachelor's degree in biology in 1960. He began medical school at Yale University but after a year there switched to graduate training in the Division of Medical Sciences at Harvard Medical School. For his doctoral dissertation, Nick studied the genes and pathways of glycerol metabolism in *Escherichia coli* under the guidance of Edmund Chi Chien Lin, earning his Ph.D. in biochemistry in 1966. Nick then pursued postdoctoral training with Arthur Kornberg, the discoverer of DNA polymerase and esteemed Nobel laureate at the Stanford University School of Medicine, where Nick began his lifelong work on DNA and its enzymes. Nick purified T4-phage-induced DNA ligase, which stitches DNA strands together, deriving its energy from the hydrolysis of ATP. Nick's ligase was used by Mehran "Micky" Goulian in the construction of enzymatically synthesized DNA of the bacterial virus phiX174, which was able to enter host cells and yield progeny virus.[6] This feat was hailed in the popular press as "synthesis of life in the test tube" and "the greatest scientific achievement of 1968," just ahead of the first successful human heart transplant.

Nick began his independent career in 1968 with his appointment to the faculties of the Departments of Biochemistry and of Biophysics and Theoretical Biology at the University of Chicago, where he initiated work on topoisomerases and site-specific recombination enzymes.[7] In 1982, he joined the faculty at the University of California, Berkeley (UC Berkeley) and spent the rest of his career there as a professor in the Department of Molecular Biology, where he served as chair and head of the Virus Laboratory, a state-funded organized research unit, from 1986 to 1989, and then, after a reorganization of the biological sciences, as a professor in the Division of Biochemistry and Molecular Biology of the Department of Molecular and Cell Biology.

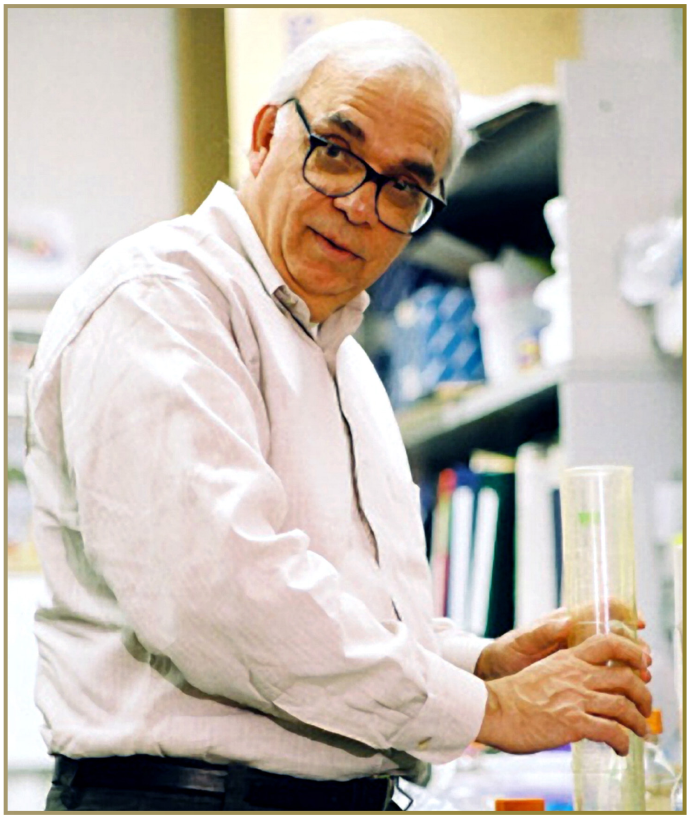

Figure 2 Nick Cozzarelli at the bench in his laboratory at UC Berkeley.

Cozzarelli was one of the first scholars to understand the importance of interdisciplinary collaborations and worked to build academic structures to facilitate such cross-fertilization.[8] For example, in 1988 he founded and directed the National Science Foundation-supported Program in Mathematics and Molecular Biology. This program encourages collaboration between mathematicians and molecular biologists and helped to explain the topological transformations, such as the pattern of knotting and unknotting of DNA molecules introduced by topoisomerases and recombination enzymes. As Bruce Alberts recalled, Nick was the "first person I knew who successfully connected leading mathematicians to first rate molecular biologists, resulting in valuable contributions to both fields. Although common today, they were quite novel when he began his collaborations in DNA topology in the 1980s."[9]

Nick was always at home in the lab, whether at the bench in his early days or later in his office embedded in the lab and from which he regularly emerged to check on his trainees. "Any neeeeews, Marko?" he'd say as he approached my (Krasnow) desk, raising up on his toes and clasping his hands while gazing down at me with the trademark Cozz grin in anticipation of an exciting new result. As recalled by former postdoc Steven A. Wasserman, "Nick relished the output of experiments as much, if not more, than their elegant and insightful design.[10] Nick built a full day each week around one-on-one meetings with his lab members, sitting down to dissect, discuss, and debate data. "He had a sincerity and complete commitment to whatever he was doing, whether it was teaching, giving a lecture, or just through interactions in the lab. It created a great lab environment. It made everything you were doing seem cutting edge and important," said his former graduate student Patrick O. Brown.[11] Another former graduate student, Michael F. Carey, said, "To this day I still hear Nick's voice. I use his phrases—good science, blast ahead, make a contribution—and I base my scientific decisions on guiding principles laid out in his training."[12] Nick created a dynamic laboratory environment that was a fertile training ground, spawning leaders in diverse areas of molecular biology.

Weekly "Cozz lab" group meetings and journal clubs were filled with scientific sparring over the latest experiments or combing through a new paper in the field. Nick relished a lively debate. His zeal for science and knowledge of the literature showed through, always with his insistence on experimental rigor. He had a sharp analytical mind and, most importantly, an ability to use his keen insight to frame new findings in the broadest context and exalt them with Cozzarelli flair. "He had a natural gift for seeing the important question and for squeezing much more information and insight out of data than most scientists thought was possible," recalled former postdoc Roland Kanaar.[13] He never shied away from sharing entertaining personal anecdotes about investigators and the history of the field, teaching us that not only the science, but also the personalities of the scientists, had a powerful influence on progress, as it clearly did with Nick.

Nick was a masterful, charismatic communicator. Some of this talent came from his passion for science. "The de facto motto of the lab, 'Blast Ahead,' was an expression of his enthusiasm, joy, and dauntless spirit of adventure," said Patrick Brown.[14] "Unlike most of us, he always had a smile on his face when he spoke, and I thoroughly enjoyed every talk of his that I heard and every paper of his that I read," reflected Alberts.[15] Nick also had a love of language and a flair for turning a phrase that manifested in both talks and writing. He knew the value of effective communication in science, worked hard to achieve it, and passed it along to his trainees. Lab manuscripts would go through many cycles to get the writing right. "Drafts would pass between Nick and the primary authors, covered with Nick's comments and remarks, often written on extra pieces of notepad paper slipped in between the manuscript pages," noted Ronald Kanaar.[16] Talks too were tuned to perfection. Kanaar recalls: "During the practice talk you could see when it wasn't quite right—his arm would go up and reach for his hair, he would bend forward, rest his elbow on the table, support his head with his hand, and, fixing his eyes on his yellow notepad, he would fill the page with difficult to decipher, but extremely useful, comments. Usually the talk was completely rearranged, inevitably for the better."[17] Nick's own talks were dynamic, often including him manipulating large plastic-ribbon DNA models in wizardly performances of the topological transitions mediated by the enzymes he was describing.

Soon after arriving at Berkeley, Cozzarelli took on the job of departmental graduate adviser. This caused a notable rise in the spirits of the graduate students thanks to his attention to their individual needs, strengths, and problems. He was never shy to proffer sage advice. "If you want to be a leader in science, you must be creative, think in an original way. The good scientist knows the literature, whereas the really good scientist knows when to forget the literature," to quote one of the many Cozzarelli gems from his several-year stint in the Ask-a-Scientist online forum overseen by the Howard Hughes Medical Institute.[18] In an interview about the forum, Nick said, "I love communicating science as much as I did when I was a college student—probably more."[19] Nick was also universally admired for and had a similar impact in his classroom teaching of introductory molecular biology for graduate students. He chose classic and current papers for students to read and to analyze in detail. Students never felt overloaded with work, but they were inspired as they were transformed from novices to near-experts. He was a brilliant teacher who, as Brown recalled, "by the force of his personality and his total engagement with the subject, made anything he was teaching in a classroom or a seminar seem, for that moment, as if nothing could be more important or interesting. He put a phenomenal amount of care and time and effort into preparing his lectures. He was the best classroom teacher I've ever seen—I'm sure there are dozens of students who were inspired to become scientists by having Nick as a teacher."[20]

In 1995, at age 57, Nick shifted his focus to scientific service. Given his gift for scientific communication, his scientific accomplishments, and his interdisciplinary bridge-building, he was invited to become editor-in-chief of the *Proceedings of the National Academy of Sciences* (PNAS), the prominent journal of the NAS he had been elected to six years earlier. At the time, the journal was in many ways unchanged since its creation in 1914 and had not adapted to the changing landscape of scientific publishing, and he was charged with returning the stodgy gray journal to its former esteemed reputation. It was a herculean task, but Nick relished the challenge and over the next eleven years remade the journal in his image. He had risen from humble beginnings to scientific prominence, and so he pushed for similar opportunities for others. "Sometimes he pushed the Academy

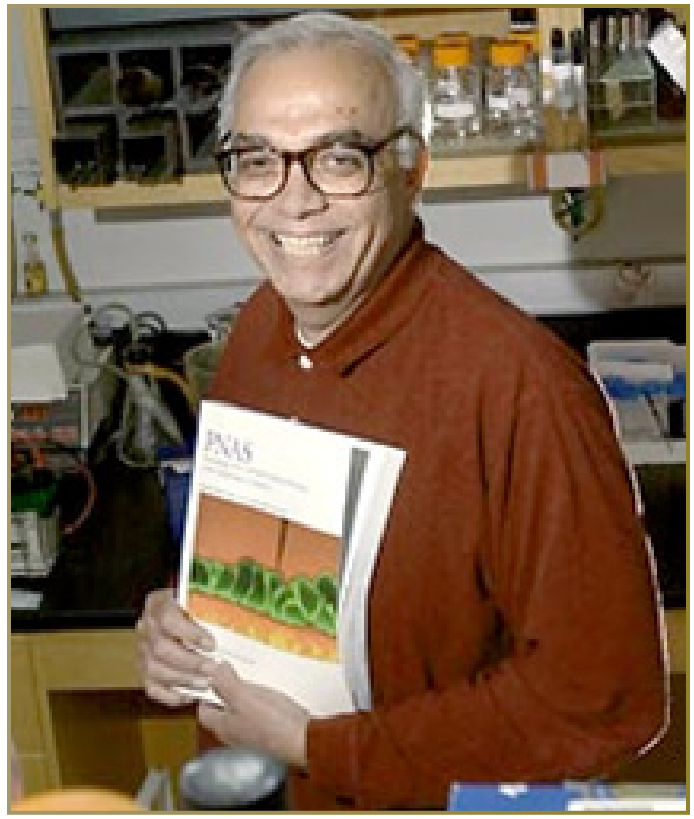

Figure 3 Nick Cozzarelli shortly after being named Editor-In-Chief of the *Proceedings of the National Academy of Sciences*.

faster than I thought it could go. Conversations would usually start with him saying 'Now, Bruce, do you really believe that.…' But always in a good-humored way that I could see him smiling, even at the other end of the phone," recalled Alberts.[21] Through sustained effort and classic Cozz cajoling, he opened the journal to manuscript submissions by all authors (Track II, Direct Submission), not just those communicated by academy members, which increased submissions to thousands a year and enhanced the quality and variety of published material.[22] He greatly expanded the editorial board and its expertise and increased publications outside the biological sciences, making it scientifically diverse, just like him. Nick also thrust PNAS into a leadership role in the expansion of open-access publishing and copyright. He believed ready access to the scientific literature was important for both science and society and so made PNAS articles free online after six months and free immediately to more than 140 developing nations. He added new sections to make content more accessible, such as highlights of several exceptional papers in the issue and biographical profiles of recently elected NAS members. He even enlivened its bland cover with striking images of the science inside. "He was the finest journal editor with whom I have ever dealt, and it is generally accepted that he was the best editor of the PNAS since its inception," said

NAS member and subsequent editor Solomon H. Snyder.[23] He was a "transformational journal editor, just as he was a transformational scientist ... fearless in confronting the difficulties in changing the journal, just as he was fearless in experimental work.... He knew when to forget the convention and start a new direction, both in science and publishing," according to Alberts.[24] He achieved his goals through his "boundless energy, enthusiasm, dedication, and uncompromising standards that characterized his own science," said Jack Halpern, inorganic chemist and Cozzarelli's former colleague at the University of Chicago.[25] Honoring Nick for his service, the NAS and the PNAS established the Cozzarelli Prize, which is "awarded annually to six research teams whose PNAS articles have made outstanding contributions to their field" and who share Nick's "uncompromising scientific standards, his dedication to advancing scientific knowledge, and his enthusiasm for an 'upstream swim.'"[26]

Throughout his career, there were few things that could tear Nick away from the lab and his science. The most significant, of course, was his wife, Linda, and daughter, Laura. At the end of a long day in the lab, Nick would abruptly rush home for the family dinner and, when Laura was young, her bedtime story, only to return after lights out to join us for a late-night lab session while Linda remained to complete notes from her day's patient therapy sessions. When Laura was older and a student at UC Berkeley, she'd slip into the lab to see Dad, and when she later became a special education teacher, he'd slip out for an occasional science talk to her class. Only Laura could dissolve, if just momentarily, Nick's legendary scientific intensity and bold personality, drawing out the sweet, gentle, caring soul inside. We saw this again, for an entire night, at her beautiful wedding to Brian Wood.

Sports could also tear Nick away from the lab, at least on Sunday afternoons. In the mid-1980s during the San Francisco 49ers Super Bowl runs, Nick held an additional lab meeting each week beyond the regular one-on-one science sessions. This was an hour or more slot exclusively with postdoc Wasserman for some equally intense Monday morning quarterbacking—"analyzing the data" from the previous day's football game. Nick's need for such analysis ran deep. When Steve initially took a faculty position out of state, Nick simply replaced the weekly in person meeting with a phone call, overnighting the data (game tape) to Steve for review just ahead of their session.

Then there was food, an essential part of the infectious joie de vivre that characterized Nick's research and interactions with others and indeed every aspect of his life. Lifelong friend I. Robert "Bob" Lehman, a faculty member during Nick's postdoc at Stanford (and with whom Nick competed on discovery of DNA ligase), reflected: "Nick was a great gourmet. He and his wife Linda joined Sandy and me monthly at gourmet restaurants in San Francisco that Nick had carefully chosen."[27] When Nick and family moved from Chicago to Berkeley, everyone naturally assumed it was for the new scientific opportunities and colleagues. But some of us wondered if the attractions might be other vistas, like their splendid minka-inspired home in the Berkeley hills with a "three-bridge view" (one more than Jim Wang had when he was on the faculty at UC Berkeley, which Nick had longingly admired), or the exalted Berkeley food scene replete with the Cheese Board and Chez Panisse? Nick and Linda regularly hosted students and colleagues, initially intimate affairs with seminar speakers, at their Hyde Park condo (a few blocks from Muhammed Ali) in Chicago but expanded that practice in Berkeley to include large receptions for as many as fifty prospective students that featured Nick and Linda's refined taste in food, wine, and art. They also hosted intimate dinner parties, none more special than their annual Christmas Eve dinner for family and friends, especially those like us without our own Christmas tradition. Linda worked days preparing the courses for the traditional Italian "Feast of the Seven Fishes" (with modern Berkeley gourmet twists) in their designer kitchen while Laura served and Nick poured paired Italian wines late into the night, the immense and lovingly decorated tree sparkling nearby. Nick sweetly bedded the children down in the master suite, leaving adults at the table continuing stories and laughter into the wee hours, tiramisu and espresso fueling their drive home.

"Nick was an original, both in personality and as a scientist," as Alberts recalled.[28] He died too young, struck down by Burkitt's lymphoma on March 19, 2006, just before his sixty-eighth birthday while still fully engaged in science. Even when desperately ill, he remained hard at work writing letters, finishing papers, and dealing with authorship issues. He was brought down from within, alas the only obstacle Nick couldn't blast through, despite the chemo-therapeutics his work spawned on hand and the *godersi la vita* he still held for science and living, especially the prospect of his grandsons, Benjamin and Alexander Nicola.

Cozzarelli was elected to the National Academy of Sciences in 1989, received the CIBA-GEIGY/Drew Award in Biomedical Research in 1990, became a fellow of the American Association for the Advancement of Science in 1999, and was elected to the American Academy of Arts and Sciences in 2000.

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