NATIONAL ACADEMY OF SCIENCES

HANS RIS
1914–2002

A Biographical Memoir by

MICHAEL F. DOLAN AND LYNN MARGULIS

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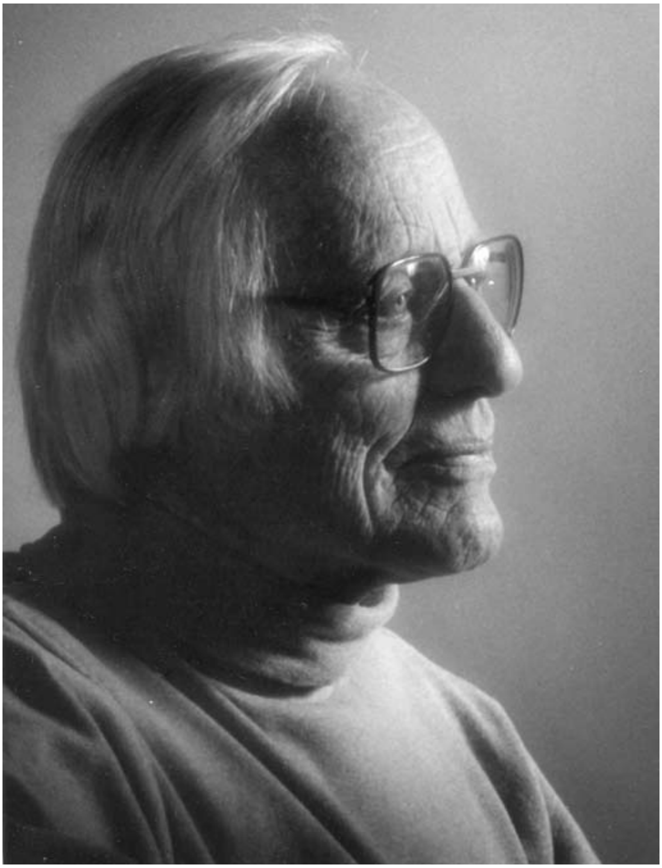

Hans Ris

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June 15, 1914–August 16, 2002

BY MICHAEL F. DOLAN AND LYNN MARGULIS

Hans Ris occupied a unique historical position: he was a cytologist (student of the cell) at the beginning of the molecular biological revolution and a superb electron microscopist when "biochemical and biophysical cytology" was renamed "cell biology." Elected to the National Academy of Sciences in 1974, he was a founder of the American Society for Cell Biology, and a winner of its E. B. Wilson Award. He led the University of Wisconsin's Integrated Microscopy Resource that boasted one of the nation's first high-voltage electron microscopes. Through his combined love of Nature and Her diverse life forms and with his dedication to the entire panoply of techniques of microscopy and biochemical cytology, he influenced the careers of scores of scientists as he generously collaborated to produce a massive collection of remarkably informative micrographs. He was a naturalist on a mission of investigation, an observer-researcher his entire life.

EARLY LIFE AND EDUCATION

Hans Ris was born in Switzerland on June 15, 1914. He grew up near a large forest on the edge of Bern. He later reflected on its influence.

From age ten I spent all my free time exploring the woods covering the hills around Bern. Observing, listening, I became fascinated by the diversity and beauty of living things…Our house was built on land where for centuries blocks of sandstone from nearby quarries were shaped into stones for the houses of old Bern and its cathedral. The leftovers had accumulated perhaps up to forty feet. Our basement on one side was open to this landscape of irregular stones, a great place for day dreaming. I enjoyed making up stories about squeezing through dark spaces to discover doors that opened into different worlds: secret gardens. And this became a guiding motive of my life, the search for doors to new secret gardens, either in the fantasy world or in the real world (1994 biographical statement).

At age 15, the same year his mother died, he bought a set of lenses and made his first microscope, using his father's cigar boxes and cardboard from old school books.

The family had lived in or around Bern's mountains for centuries. During the Protestant reformation, August Ris's ancestors, as Huguenots, fled from Alsace-Lorraine to Switzerland in 1653. The business of Hans's paternal family was knife making. Hans's grandfather, who as a youth was placed in an orphanage on the death of Ris's great-grandfather, was a talented artist whose sketch books are still in the hands of Ris's son Christopher. August Ris married Martha Egger, whose family members were farmers from the mountains outside Bern. Hans's mother, Martha, was an intelligent, active woman, a photographer who founded her own business. She was a devoted Christian Scientist. A tall, flamboyant, glamorous person, Martha Ris was owner and director of a Bern women's hat-making company. She went regularly to Paris to keep up with the latest fashions. Hans, eldest of her three sons, was interested in art and music from the beginning. As a talented young adolescent he accompanied his mother to the local Christian Science church where for almost three years on Sundays he played the organ. The year he was 15 Hans and his brothers suffered the agonizing loss of their mother to breast cancer. Her belief in the efficacy of prayer and refusal to accept medical aid probably scarred young Hans for the rest of his life. Because of his vociferous disbelief when the church people threatened him after his mother's death with eternal damnation, he refused to ever again attend Christian Science or other religious services.

After he received his diploma from the University of Bern, Ris was awarded a fellowship at the University of Rochester to work with embryologist Benjamin Willier. As with his subsequent doctoral work, Ris published this as sole author. The paper, "An Experimental Study on the Origin of Melanophores in Birds," was characteristic of his writings in that it was thorough: heavily methodological, quantitative, well illustrated, and lengthy.

COLUMBIA AND ROCKEFELLER

Ris worked much of the 1940s in New York City, first as a doctoral student with Hans and Sally Schrader at Columbia University. He later returned to the City for five years at Albert Mirsky's lab at Rockefeller University. The two experiences were quite different. However, when Ris left he was already one of the nation's premiere young cytogeneticists who had a working knowledge of biochemistry.

WISCONSIN

With his move to the University of Wisconsin in 1949 to become associate professor of zoology, Ris reinvented himself as an electron microscopist. He eventually replaced his Feulgen-stained preparations on glass slides with a mammoth microscope two stories tall that required the electric power of a city block. His goal—an integrated facility in which all forms of microscopy, from low-power binocular scopes to the high-voltage electron microscope (HVEM) were simultaneously employed to observe the same material—was achieved. He constructed one of the most powerful microscopical facilities in the world. With the HVEM Ris produced transmission and scanning stereoimages. After his retirement, his successor dismantled the HVEM. Ris suffered a bitter loss.

MAJOR SCIENTIFIC CONTRIBUTIONS

CHROMATIN CONTENT OF SOMATIC AND GERM CELLS.

Working at the Mirsky lab at Rockefeller, Ris developed the use of the Feulgen reaction that stains DNA in a quantifiable fashion, and then applied it to cells of fishes and other animals. He discovered that somatic cells of a species all had the same quantity of chromatin (and the same complement of chromosomes) and that germ cells (egg and sperm) had precisely half the amount. He provided the cytological and biochemical confirmation of the concept of haploidy/diploidy of normal nucleated cells of organisms, i.e. euploidy, an idea so taken for granted by biologists that it does not seem to have needed a discoverer. A year earlier researchers in France had published the same measurements in plants, but the Ris discovery, in the excitement over the nature of chromatin and the gene, converted the finding that germs cells have half the DNA of somatic cells to an unstated assumption of textbook botanical-zoological cannon.

ANAPHASE B

Throughout his career Ris found the biological material best suited for his investigations of cell division. He was comfortable and enthusiastic with the study of mammalian cells, insects, plants, algae, insects, and cyano- or other bacteria in his incessant search for biological principles. He showed the patience to develop the best techniques for handling live, healthy material and fixing (preserving) it for detailed study at progressively higher magnifications. The discovery of what is now called anaphase II of mitotic cell division, the movement apart of the spindle poles after the separation of the chromosomes, was made by Ris in a study of spermatocytes in the barberry aphid *Tamalia.* He used direct observation rather than photographs and concluded, "We could then picture the anaphase movement as composed of two phases: in the first the chromosomes approach the poles, or in other words, the chromosomal fibers shorten. In the second phase the spindle stretches and moves the chromosomes farther apart." (1943)

CHLOROPLAST DNA AS EVIDENCE OF SYMBIOGENESIS

As he mastered the electron microscope Ris with R. N. Singh, a visiting researcher at the University of Wisconsin, explored the cyanobacteria, then called "cyanophytes," or "blue-green algae." Their work confirmed the growing realization of biologists that blue-green algae are bacteria, not plants. The cyanophytes differed fundamentally from all the algae and plants. "Our electron micrographs confirm the view that the cell of blue-green algae is different and simpler in organization than the typical plant or animal cell. On the other hand, the general pattern of ultrastructure is the same as that found in bacteria and *Streptomyces,*" Ris and Singh wrote. In his 1962 paper with Walter S. Plaut, Ris's close colleague in the Botany Department at Madison, Ris, through Feulgen and acridine orange DNA staining removed by DNase digestion, found fibrous structures similar to those in many bacteria he had studied. This confirmed his suspicion that chloroplasts contain their own DNA. By comparing the micrographs of the 20Å fibers in the nucleoplasm (later called nucleoids) of *Calothrix* and *Nostoc,* both filamentous cyanobacteria, with comparable DNA preparations of chloroplasts of the green alga *Chlamydomonas,* Ris and Plaut provided a key piece of evidence that the plastids of plants and algae evolved symbiogenetically from cyanobacteria. They concluded, The evolution of the complex cell, with its array of more or less autonomous organelles, from the simpler organization found in Monera [bacteria] is a question that has been neglected. With the demonstration of ultrastructural similarity of a cell organelle and free-living organisms, endosymbiosis must again be considered seriously as a possible evolutionary step in the origin of complex cell systems.

DINOFLAGELLATE DIVISION WITH DONNA KUBAI

A further step Ris took to investigate the evolutionary origin of eukaryotic cells was a project he conducted with his Ph.D. student Donna Kubai to explain mitosis in the dinoflagellates. The unique appearance of dinoflagellate chromosomes had been known since the work of Eduard Chatton (1887-1945). Consistent with their peculiar condensed, mitotic-like state during interphase, they apparently lacked histone proteins. Their chromosomes were composed of 25Å units diameter fibers, similar to those of prokaryote nucleoids. In contrast with most forms of mitosis Kubai and Ris found no spindle microtubules directly involved in chromosome movement. Instead microtubules form bundles that pass through cytoplasmic channels that are exterior to the closed nuclear membrane. The dinoflagellate chromosomes attach to the inside of the nuclear envelope. They are moved as the nuclear envelope grows or flows—another similarity to bacterial cell division. The hypothesis that dinoflagellates were an intermediate (mesokaryotic) form between prokaryotes and eukaryotes was briefly asserted in the textbooks. Subsequent studies showed unambiguously that dinoflagellates are eukaryotes, but like many other protists they are variant in the details of their mitotic behavior. Fascinated by their nuclei, chromosomes, and mitotic division behaviors Ris continued investigation of the unique cell biology of protists for his entire life. Among his papers he left superb but unpublished micrographs. He documented idiosyncratic unipolar, persistent mitotic spindles of the colonial radiolarian *Collozoum*, for example. His unpublished work undoubtedly holds new discoveries (see Archives section below).

30 NM CHROMOSOME FIBER

Ris, a leading early investigator in the use of the electron microscope, studied the cyclical morphological changes in chromosomes. He reported as "the smallest unit obtainable without removing or dislocating the histones (1956)" a 10 nm chromatin fiber. He later realized that the naturally occurring state of chromatin comprises 30 nm supercoiled solenoids. He established the 30 nm (not the 10 nm) fiber as the defined secondary structure of chromatin in vitro. "It became clear that the 10-nm fibers seen in sections and isolated nuclei were produced by the buffers used and did not represent the native state," he reported in 1979. As with the so-called microtrabecular network in cells, which he debunked, Ris pioneered the study of adequate techniques to separate real, persistent structures from artifacts caused by different methods of fixation and microscopic preparations.

THREE-DIMENSIONAL STRUCTURE OF THE NUCLEAR PORE COMPLEX

Ris crowned his remarkable career with a discovery that revealed the power of his beloved Integrated Microscopy Resource in Madison. By use of high-voltage electron microscopy and low-voltage high-resolution scanning electron microscopy, where the same material was subject to observation at many size scales with each type of microscopic technique, he demonstrated that the nuclear pore complex, the site for transport of large molecules from the nucleus to the cytoplasm, that had previously been considered a conventional two-dimensional structure, was really an elaborate three-dimensional complex that resembles a fishtrap (now called the "nuclear basket"). Still working away in his laboratory, Ris at age 75 again showed the great value of integration of results from different imaging techniques in research on living organisms.

> These stereoimages, which represent some of the most spectacular and informative electron micrographs found in the literature to this day, have started a new chapter in the history of NPC [nuclear pore complex] structure. In fact, these remarkable ultrastructural data have challenged many researchers in the field—including ourselves—to replicate, scrutinize, and elaborate on these novel views of NPC architecture. Last, but not least, Ris' morphological data also had a profound impact on the molecular dissection of NPC structure (Lim et al., 2006).

PUBLIC SERVICE

Ris was a founding executive committee member of the American Society for Cell Biology in 1960. He received the society's E. B. Wilson Award in 1993. Ris cherished Wilson's book *The Cell in Development and Inheritance* (Wilson, 1925), and read it aloud to his advanced cytology students. He served on a National Academy of Sciences committee that visited China to investigate the professional employment prospects for Chinese students who returned from the United States.

Thoru Pederson, who served with Ris on the Council of the American Society for Cell Biology, recalled in his 2005 Ris obituary,

> While he was always open to new ideas (and was a good listener—patient and never interrupting), at the same time he had a very astute sensitivity to people who were self-promoting. Hans was a seeker of truth, and the only times I witnessed anger on his otherwise smiling face was when he suspected deception or distortion in someone. What a wonderful trait, and how mightily it adds to our respect of him.

FAMILY LIFE

Ris married Hania Wislicka (June 16, 1913-March 25, 1998), a Zürich medical student who became a pediatrician. Hania, a Polish Jewess, had lost both her parents in the German death camps of World War II. After the couple married and immigrated to the United States, Hania did her residency at Johns Hopkins University, Baltimore. She bore him two children: Christopher (b. March 28, 1949) and Anet (b. June 24, 1953). Hans is survived by his second wife, the artist Theron Caldwell Ris; his son and his wife and their daughter of Fairfax, California; and his daughter and her husband of Portland. Oregon.

ARCHIVES

Hans Ris's archival materials are found at three institutions. His papers dealing with his career at the University of Wisconsin were deposited in the University Archives at the Steenbock Library. His research and other professional papers were donated to the American Philosophical Society Library. His collection of 3,000 micrographs, the best of which are those that Ris called his "treasure," are electron micrographic images of nuclei, chromatin, and cytoskeletal structures that come from a diverse array of organisms. Many are of protists, and most are unpublished. This entire collection is archived in the library of the American Museum of Natural History.

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