THE JOURNAL OF BIOLOGICAL CHEMISTRY

Vol. 281, No. 10, Issue of March 10, p. e10, 2006
Printed in U.S.A.

Classics

A PAPER IN A SERIES REPRINTED TO CELEBRATE THE CENTENARY OF THE JBC IN 2005

JBC Centennial
1905–2005
100 Years of Biochemistry and Molecular Biology

Precocious Newborn Mice and Epidermal Growth Factor: the Work of Stanley Cohen

Isolation of a Mouse Submaxillary Gland Protein Accelerating Incisor Eruption and Eyelid Opening in the New-born Animal
(Cohen, S. (1962) *J. Biol. Chem.* 237, 1555–1562)

The Primary Structure of Epidermal Growth Factor
(Savage, C. R., Jr., Inagami, T., and Cohen, S. (1972) *J. Biol. Chem.* 247, 7612–7621)

Stanley Cohen was born in Brooklyn, New York, in 1922. He attended Brooklyn College where he majored in both biology and chemistry and graduated with a B.A. in 1943. After working as a bacteriologist in a milk processing plant to save enough money to go to graduate school, Cohen received a fellowship from Oberlin College, where he earned an M.A. in zoology in 1945. He then acquired another fellowship from the biochemistry department at the University of Michigan and obtained his Ph.D. in 1948. Cohen's graduate thesis concerned the mechanism by which the end product of nitrogen metabolism in the earthworm is switched from ammonia to urea during starvation. To do this research he had to spend his nights collecting over 5000 worms from the University campus green.

After graduating, Cohen joined Harry Gordon in the pediatrics and biochemistry departments of the University of Colorado, where he was involved in metabolic studies on premature infants. Feeling the need to gain experience in the emerging application of radioisotope methodology to biological research, Cohen left Colorado and went to Washington University in 1952 to work with Martin Kamen in the Department of Radiology. There he learned isotope methodology while studying carbon dioxide fixation in frog eggs and embryos. In 1953 Cohen joined the Department of Zoology at Washington University as an Associate Professor. There he collaborated with Rita Levi-Montalcini on the isolation of a nerve growth factor that Levi-Montalcini had discovered in certain mouse tumors. This was Cohen's introduction to the world of growth factors, which would become the focus of his scientific career.

In 1959 Cohen joined the faculty of the biochemistry department at Vanderbilt University as an Assistant Professor. At Vanderbilt, he continued to focus on growth factors. During his study of a nerve growth factor detected in male mouse submaxillary glands he noted that injection of crude submaxillary gland preparations into newborn mice elicited unexpected side effects not related to the activities of the nerve growth factor. These included precocious eyelid opening and precocious tooth eruption (1). Using these precocious characteristics as an assay, Cohen purified the "tooth-lid factor" from murine submaxillary glands. The isolation of this factor is the subject of the first *Journal of Biological Chemistry* (JBC) Classic reprinted here. In the Classic, Cohen reports, "The tooth-lid factor is a heat-stable, nondialyzable, antigenic protein, whose most distinctive chemical characteristic is the absence of phenylalanine and lysine." Subsequent histological examination showed that the precocious eyelid separation was due to the factor's ability to directly stimulate epidermal cell proliferation. Based on these observations, Cohen named the tooth-lid factor epidermal growth factor (EGF).

Cohen's development of a rapid process for isolation of milligram quantities of EGF from murine submaxillary glands in the early 1970s (2) permitted him to purify sufficient quantities for a thorough characterization. Using this material, he, C. Richard Savage, Jr., and Tadashi Inagami, determined the primary sequence of mouse EGF, as reported in the second JBC Classic reprinted here. They used automatic Edman degradation and chemical and enzymatic

cleavage to elucidate the sequence and discovered that EGF is a 53-residue polypeptide with six half-cystines that exist in disulfide linkage. Cohen later determined the positions of the three internal disulfide bonds and published the results in the JBC (3).

In 1976 Cohen was appointed an American Cancer Society Research Professor at Vanderbilt, and in 1986 he was named Distinguished Professor. He retired in 2000 and is currently Distinguished Professor Emeritus at Vanderbilt. Cohen continued to study EGF, its interaction with cell surface receptors, and the intracellular signaling pathways activated by the growth factor. In recognition of this work, he shared the 1986 Nobel Prize in Physiology or Medicine 1986 with Rita Levi-Montalcini "for their discoveries of growth factors."

In addition to the Nobel Prize, Cohen has received many awards and honors, including the National Paraplegia Foundation's Second Annual William Thomson Wakeman Award (1974), Vanderbilt University's Earl Sutherland Prize for Achievement in Research (1977), the National Academy of Science's H. P. Robertson Memorial Award (1981), the General Motors Cancer Research Foundation Alfred P. Sloan Award (1982), the American Academy of Dermatology's Lila Gruber Memorial Cancer Research Award (1983), the Gairdner Foundation International Award (1985), the National Medal of Science (1986), the Endocrine Society's Fred Conrad Koch Award (1986), and the Albert Lasker Basic Medical Research Award (1986). He was elected to the National Academy of Science in 1980 and the American Academy of Arts and Sciences in 1984 and has served on the Editorial Boards of the *Abstracts of Human Developmental Biology* and the *Journal of Cellular Physiology*.[1]

Cohen's co-author on the second JBC Classic, Tadashi Inagami (1931), has also made many important contributions to science. At the time he collaborated with Cohen on the growth hormone work, Inagami was an Associate Professor of Biochemistry at Vanderbilt University School of Medicine. He eventually became Stanford Moore Professor of Biochemistry at Vanderbilt in 1991. The majority of Inagami's research has centered on investigating hypertension. He was the first to purify renin and to elucidate its specific protease activity, and he isolated and determined the structures of multiple forms of the angiotensin receptor. Inagami also discovered the biochemical mechanism by which renin works. In recognition of his research, Inagami has received many honors including the Humboldt Foundation Award (1981), the American Heart Association's Ciba Award (1985), Vanderbilt University's Sutherland prize (1990), the Japan Vascular Disease Research Foundation's Okamoto International Award (1995), the Bristol-Myers Squibb Award for Excellence in Cardiovascular Research (1996), the Japan Society for Cardiovascular Endocrinology and Metabolism's Jokichi Takamine Memorial Award (1998), and the American Heart Association's Research Achievement Award (1994).

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[1] All biographical information on Stanley Cohen was taken from Ref. 4.