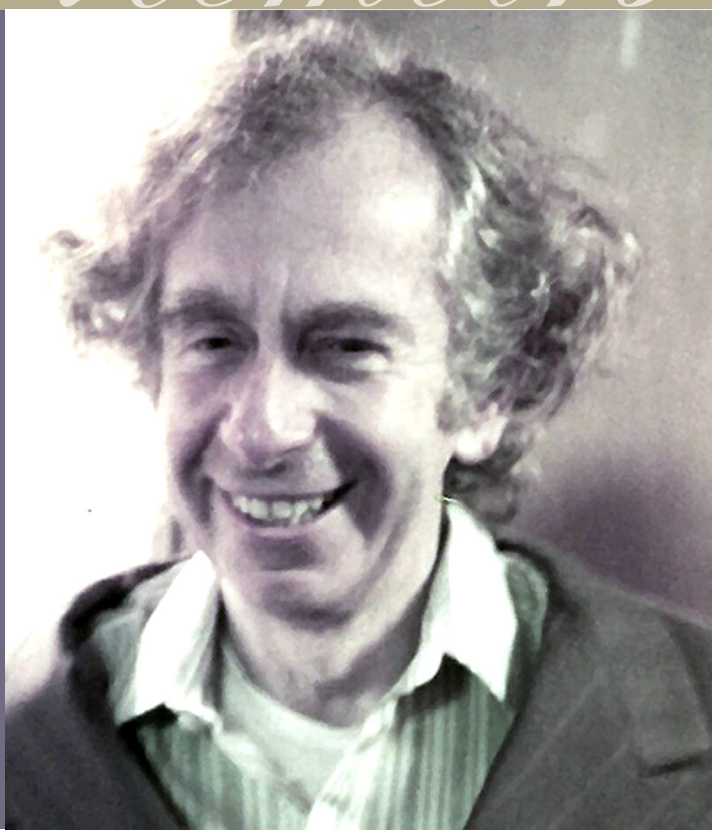
# Edwin J. Furshpan

1928–2019

BIOGRAPHICAL *Memoirs*



*A Biographical Memoir by*
*Zach W. Hall,*
*Story Landis,*
*and Richard E. Mains*

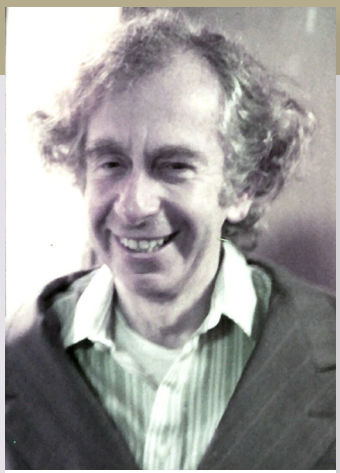


NATIONAL ACADEMY OF SCIENCES

# EDWIN JEAN **FURSHPAN**

*April 18, 1928–November 18, 2019*

Elected to the NAS, 1982



*By Zach W. Hall, Story Landis, and Richard E. Mains*

Edwin Furshpan began his career in neurobiology at a time when two principles of synaptic transmission had finally been firmly established: first, that signaling in the nervous system is unidirectional and occurs through chemical transmission at synapses; and second, that each neuron secretes only a single neurotransmitter. Through a series of influential experiments over the course of his career, many with David Potter, Furshpan extended our understanding beyond these precepts to reveal a richer world of possibility for synaptic mechanisms and for neuronal identities. His scientific papers were invariably original, addressed important questions, and were notable for their rigor and technical innovation. In addition to his scientific contributions, Furshpan, along with his colleagues David Potter and Edward Kravitz, were influential in developing new ways of teaching neurobiology and, particularly, in working to extend access for underserved student populations to medical science and neurobiology.

## Education and Postdoctoral Training

Edwin Jean Furshpan was born in Hartford, Connecticut, on April 18, 1928. His fascination with scientific research began with a project on the movement of insect wings during flight when he was an undergraduate in zoology at the University of Connecticut (UC) in Storrs. Working with Edward Boettiger at UC and at the Marine Biological Laboratory (MBL) in Woods Hole, Massachusetts, he invented a method of recording based on electrostatic charge, which allowed the study of very fast insect wing movements that were not previously observable. His undergraduate studies were interrupted by World War II, and he served from 1946 to 1948 in the U.S. Army's Panama Canal Department. After the war, Furshpan pursued his Ph.D. at the California Institute of Technology (Caltech), studying muscle stretch receptors in crayfish with Cornelius Wiersma.

E. J.Furshpan, UConn Yearbook 1949.

For his postdoctoral training, Furshpan spent three remarkably productive years in the laboratory of Bernard Katz in the Department of Biophysics at University College London. His first study followed the observation, by Paul Fatt and Bernard Katz in 1952, of spontaneous presynaptic release of acetylcholine at the frog neuromuscular junction as revealed by miniature end-plate potentials of uniform size. Fatt and Katz found that the frequency of the spontaneous potentials was affected by small changes in the osmolarity of the bathing solution. Furshpan extended this work by showing that the time course of the effect depended on the membrane permeability of the solute. Thus, membrane impermeant solutes such as NaCl or sucrose gave profound and long-lasting changes, whereas permeable solutes such as ethanol and sucrose yielded only small and transient changes in spontaneous neurotransmitter release.

## A New Mechanism of Synaptic Transmission

In London, Furshpan began a lifelong scientific partnership with David Potter, a postdoctoral student from Harvard University who was also in the Katz lab. Together, they published three historic studies on the synapses made by giant motor neurons of the crayfish abdominal cord onto motor neurons, a synaptic connection that mediates the rapid escape response. The first paper established that the giant axons excite motor axons not through a chemical transmitter, but via an electrical synapse in which current passes directly from presynaptic to postsynaptic neurons. Importantly, Furshpan and Potter demonstrated that the synaptic junction was rectifying, meaning that a depolarizing positive current could easily pass from a depolarized giant axon to the motor neuron, but not in the opposite direction. Their description of a rectifying electrical junction was groundbreaking and was regarded as an almost heretical idea in 1957, but this mechanism of neurotransmission is now described in all modern textbooks of neurobiology. The second paper (1959) demonstrated

Clockwise from top left: Ed Furshpan, Steve Kuffler, David Hubel, Torsten Wiesel, Ed Kravitz, Dave Potter.

that transmission at these electrical synapses had a negligible synaptic delay, much shorter than that found at chemical synapses. They also showed that a hyperpolarizing, but not a depolarizing, pulse passed readily from the postsynaptic neuron to the presynaptic fiber, elegantly establishing that the electrical junction functions as a conventional diode. The third paper (1959) examined another mechanism of synaptic transmission at the junction of the giant axons onto the motor neuron, in which a chemical transmitter, likely gamma amino-butyric acid (GABA), produces an inhibitory postsynaptic potential.

## Further Varieties of Intercellular Signaling

Following their time in London, Furshpan and Potter were recruited by Stephen Kuffler to the Neurophysiology Laboratory at the Wilmer Eye Institute at Johns Hopkins University Medical School in Baltimore. Shortly thereafter, the laboratory, consisting of Kuffler, Furshpan, Potter, David Hubel, and Torsten Wiesel, moved to the Pharmacology Department of Harvard Medical School (HMS). This group would later form the nucleus of a new Department of Neurobiology. At Harvard, Furshpan, working with Takahisa Furukawa, made another landmark observation by describing a novel mechanism of electrical inhibition in the Mauthner cells of goldfish. In a series of definitive experiments, they established that goldfish Mauthner cells receive two types of inhibition: a conventional inhibitory chemical signal mediated by a neurotransmitter that activates chloride channels in the Mauthner cell, plus a huge and heretofore unexpected inhibitory effect of nearby axons firing action potentials. The train of action potentials results in passive hyperpolarization of the axon hillock of the Mauthner cell, thus blocking the formation of action potentials that arise at that site. The inhibitory effect could be mimicked by a hyperpolarizing current from an external electrode placed close to the axon hillock. This mechanism of "excitation causing inhibition," previously unknown, is now believed to occur in many regions of the mammalian brain, but this was the first and experimentally the most compelling demonstration of this phenomenon.

In a later series of experiments with Potter and Edwin Lennox, Furshpan examined a very different type of electrical connection, the low-resistance gap junctions between cells that are now known to occur in many tissues. Their first study, in 1966, established that virtually all of the cells of the developing squid are electrically coupled via tight junctions, now often called gap junctions, which readily pass currents in both directions between cells. Such junctions are widespread among the cells of many developing embryos (for example, cardiac myocytes uncouple from the rest of the body when the heart starts to beat), and they are widespread in early neuronal development, especially during synapse

formation. That same year, Furshpan was named the R. H. Pfeiffer Professor in the Department of Neurobiology.

## Synaptic Transmission in Developing Neurons in Culture

Furshpan continued his collaboration with Potter in an extended series of studies of the synapses formed by sympathetic neurons in cell culture. During this period, they attracted and assembled a remarkable group of talented trainees whose diverse skills allowed them to extend their studies beyond electrophysiology and pharmacology to include ultrastructure and biochemistry. A characteristic feature of Furshpan's and Potter's work during this period was that they refused to put their name on any paper in which they did not perform a preponderance of the experiments. Edward Kravitz, a neurochemist and one of Furshpan's closest colleagues in the HMS Neurobiology Department, commented that this policy was "a shame because [Ed and Dave] were so intimately involved in the thinking behind the studies and in guiding every step of their execution by their students and post-docs. They inherited this practice from Bernard Katz, who was rigid in his belief that investigators should not share in the authorship if they hadn't done most of the experiments in the paper." Thus, the Furshpan-Potter laboratory contributed many papers in which the only attribution to either of the senior investigators was in the acknowledgements. This was particularly true of a long series of papers, many listed below, related to the growth, development, and phenotypic properties of sympathetic, parasympathetic, sensory, enteric, and hippocampal neurons in tissue culture.

Clockwise from bottom right: Furshpan, Ed Kravitz, David Hubel, Torsten Wiesel, Dave Potter.

This series of experiments was initiated by a critical methodological step made by Dennis Bray, Richard Mains, Linda Chun, and Paul Patterson in the Furshpan-Potter laboratory. They established conditions in which sympathetic neurons from newborn rodents could be grown in culture at low density in the absence of glial cells. If ascorbic acid, required for one of the synthetic enzymes, was present, the cultured neurons increased their content of catecholamines over several months, matching the production per neuron seen during development in vivo. Surprisingly, when glial and supporting cells were also present in the cultures, the neurons began to express acetylcholine. Working with

Paul O'Lague, Kunihiko Obata, Phillipa Claude, and Peter MacLeish, Furshpan and Potter established that, under these conditions, the cultured neurons formed cholinergic synapses among themselves.

To determine whether observed cholinergic and adrenergic properties represented different populations of neurons, or were both expressed in individual cells, Furshpan and Potter developed an elegant microculture system in which a single neuron was cultured with a dozen or so cardiomyocytes. In these cultures, the synaptic properties of single neurons could be followed over time. With Steven Matsumoto and Story Landis, they found that, in the microcultures, individual neurons formed cholinergic synapses with themselves (autapses) and either adrenergic, cholinergic, or mixed synaptic connections with the cardiomyocytes. By following individual neurons over time, they found that a single neuron could change from a purely adrenergic phenotype to a mixed adrenergic/cholinergic phenotype to a purely cholinergic phenotype. Further analysis by Matsumoto and Dinah Sah revealed that individual cells could also release other neurotransmitters, including adenosine, serotonin, and a long-acting excitatory substance, possibly neuropeptide Y, which is abundant in sympathetic neurons.

Collectively, these observations clearly demonstrated the plasticity of the neuronal transmitter phenotype and, importantly, that a single neuron could release multiple chemical transmitters. These findings were initially met with great skepticism among neuroscientists, but like rectifying electrical synapses, electrical inhibition, and electrical coupling, they are now described in neuroscience textbooks.

Others in the Furshpan-Potter laboratory extended these initial studies in a variety of ways. Colin Nurse and Peter MacLeish found that sympathetic neurons develop cholinergic properties when cultured with skeletal myocytes as well as cardiomyocytes. Direct contact between the cells was not required, suggesting that transmitter phenotype was influenced by secreted factors; the addition of several known cytokines could induce a change in phenotype. Rae Nishi and Alan Willard found that secreted factors could affect the transmitter properties of enteric neurons, and Paola Baccaglini and Ellis Cooper showed that such factors could influence transmitter expression in nodose sensory neurons. Landis and her collaborators demonstrated an adrenergic-to-cholinergic transition in vivo by sympathetic neurons innervating sweat gland targets. Importantly, Paul Patterson found that a soluble factor mediated the conversion and later, as an independent investigator at Caltech, established its identity as Leukemia Inhibitory Factor, a multifunctional cytokine.

Thus, the pioneering studies by Furshpan, Potter, and their collaborators opened the door to a new understanding of the complexity of synaptic transmission at individual synapses and to a rich body of subsequent investigations into the developmental mechanisms that determine transmitter phenotype.

## Epilepsy

In a final phase of experimental science, Furshpan began a study of cortical seizures, mainly using cultured hippocampal neurons. If grown in the presence of agents blocking neurotransmission, these neurons exhibited epileptiform activity when the blockade was removed. The studies grew out of long-standing observations that cortical neural networks can generate vigorous spontaneous activity. This capability is normally repressed, but when the suppression is overcome, the neurons explode into synchronous firing. Cultured hippocampal neurons presented a convenient experimental model for the very large, sustained depolarizations that are characteristic of spreading depression propagating across the cortex. Interestingly, the intense seizure-like activity in the cultures led to extensive neuronal death within the first day after removal of the agents blocking neurotransmission.

## Teaching

At HMS, Furshpan and Potter were renowned for their dedication and skill as teachers. They developed one of the first courses for medical students that integrated neurophysiology and neuroanatomy. Most importantly, they developed a revelatory method of using the movement of ionic currents to teach axonal and synaptic electrophysiology, topics that were at the time largely opaque to those without an engineering background and often misrepresented in textbooks. Their lectures, which were widely adopted by others, were carefully honed anew each year during a week at the MBL when they rehearsed the lectures with each other. Their approach to teaching was also personal. Each year David memorized the names of the hundred or so first-year medical students in the neuroscience class. The best demonstration of their popularity with students is that they were invariably lampooned in the annual medical student musical spoof, most memorably one year as “Bedpan and Potty.” Anecdotally, it was said that their course was responsible for a significant increase in the number of HMS graduates who chose to specialize in neurology. Later, Ed was a major architect of the New Pathway for Medical Education, based partly on his observation of classes at the Harvard Business School.

## Educational Access

Throughout his career, Furshpan was devoted to issues of racial equity in education. In combination with Potter and Kravitz, Furshpan played a major role in significantly increasing the number of Black medical students at Harvard (from about an average of one per year or none at all to twelve or more). Their efforts were recognized at the May 2019 HMS Division of Medical Sciences Hooding Ceremony, at which Ed and Potter shared the first annual presentation of the Career Service Award. "Furshpan and Potter represent two really amazing faculty pioneers who worked together as close partners to transform HMS curriculum and diversity over many years," said David Van Vactor, director of the division and professor of cell biology in the Blavatnik Institute at HMS. Joan Reede, HMS Dean for Diversity and Community Partnership, worked with Furshpan and Potter for three decades and was amazed at their ability to get students excited in their creative case-based approach to learning. Boston area high school students and Harvard graduate and medical students all benefitted from this innovative approach, always with an emphasis on "now, what is really fundamental in this case?" Reede thought the approach was very "humble and remarkable," coming from such eminent scientists. One of the first cases, called "Mary's Mystery," was a case of intermittent seizures, including electroencephalograms in the diagnosis even for the high school students, with the emphasis on "getting the science right for the patient."

Kravitz has noted, "In addition, they were well known for working on a national scale to 'enlarge the pipeline' for students from underrepresented groups, particularly Native American communities, to pursue successful careers in medicine and research, starting in the 1960s." Their signal achievement was to organize a special summer program for high school students from the Hopi and Fort Peck reservations in which a group of students and their teachers came to Harvard for a three-week course organized around medical and scientific issues particularly relevant to Native American health. Kenneth Smoker, head of the Wellness Program at the Fort Peck Hopi reservation, said that the Native American Program at Harvard "didn't just change lives; it saved lives…. [Furshpan and Potter] listened to students and motivated them to dream big, to begin to think like doctors and to focus on health outcomes." Later in their careers, Furshpan and Potter taught a neuroscience course every year at Morehouse College that was open to students from Spelman College, Morehouse College and its School of Medicine, and Clark Atlanta University. Furshpan's concern for others was expressed during the last four years of his life by his work on the manuscript for his book *Empathy and the Human Condition*, which aimed to explore some of the biological aspects of "man's inhumanity to man" that he sought so fervently to change throughout his career.

## ACKNOWLEDGMENTS

We thank John Hildebrand, Peter MacLeish, Shelagh Foreman, Joan Reede, Kenny Smoker, and Jim Wohl for help with facts and perspectives throughout this project.

## SELECTED BIBLIOGRAPHY


1950 With E. G. Boettiger. Observations on the flight motor of Diptera. *Biol. Bull.* 99:346–347.

1952 With E. G. Boettiger. The recording of flight movements in insects. *Science* 116:60–61.

1954 With A. C. Giese. Ultraviolet excitation of a stretch receptor. *J. Cell Comp. Physiol.* 44:191–201.

1956 The effects of osmotic pressure changes on the spontaneous activity at motor nerve endings. *J. Physiol.* 134:689–697.

1957 With D. D. Potter. Mechanism of nerve-impulse transmission at a crayfish synapse. *Nature* 180:342–343.

1959 With D. D. Potter. Transmission at the giant motor synapses of the crayfish. *J. Physiol.* 145:289–325.

With D. D. Potter. Slow post-synaptic potentials recorded from the giant motor fibre of the crayfish. *J. Physiol.* 145:326–335.

1963 With T. Furukawa. Intracellular and extracellular responses of the several regions of the Mauthner cell of the goldfish. *J. Neurophysiol.* 25:732–771.

With T. Furukawa. Two inhibitory mechanisms in the Mauthner neurons of goldfish. *J. Neurophysiol.* 26:140–176.

1964 "Electrical transmission" at an excitatory synapse in a vertebrate brain. *Science* 144:878–880.

1966 With D. D. Potter and E. S. Lennox. Connections between cells of the developing squid as revealed by electrophysiological methods. *Proc. Natl. Acad. Sci. U.S.A.* 55:328–336.

1968 With D. D. Potter. Low-resistance junctions between cells in embryos and tissue culture. *Curr. Top. Dev. Biol.* 3:95–127.

1974 With P. H. O'Lague, K. Obata, P. Claude, and D. D. Potter. Evidence for cholinergic synapses between dissociated rat sympathetic neurons in cell culture. *Proc. Natl. Acad. Sci. U.S.A.* 71:3602–3606.

1976 With P. H. O'Lague, P. R. MacLeish, C. A. Nurse, P. Claude, and D. D. Potter. Physiological and morphological studies on developing sympathetic neurons in dissociated cell culture. *Cold Spring Harb. Symp. Quant. Biol*. 40:399–407.

Chemical transmission between rat sympathetic neurons and cardiac myocytes developing in microcultures: Evidence for cholinergic, adrenergic, and dual-function neurons. *Proc. Natl. Acad. Sci. U.S.A*. 73:4225–4229.

1978 With P. H. Patterson and D. D. Potter. The chemical differentiation of nerve cells. *Sci. Am*. 239:50–59.

With P. H. O'Lague and D. D. Potter. Studies on rat sympathetic neurons developing in cell culture. I. Growth characteristics and electrophysiological properties. *Dev. Biol*. 67:384–403.

With P. H. O'Lague and D. D. Potter. Studies on rat sympathetic neurons developing in cell culture. II. Synaptic mechanisms. *Dev. Biol*. 67:404–423.

With P. H. O'Lague and D. D. Potter. Studies on rat sympathetic neurons developing in cell culture. III. Cholinergic transmission. *Dev. Biol*. 67:424–443.

1980 With D. D. Potter and S. C. Landis. Dual function during development of rat sympathetic neurones in culture. *J. Exp. Biol*. 89:57–71.

1980–81 With D. D. Potter and S. C. Landis. On the transmitter repertoire of sympathetic neurons in culture. *Harvey Lectures* 76:149–191.

1981 With D. D. Potter and S. C. Landis. Adrenergic-cholinergic dual function in cultured sympathetic neurons of the rat. *Ciba Found. Symp*. 83:123–138.

With D. D. Potter and S. C. Landis. Chemical differentiation of sympathetic neurons. *Adv. Biochem. Psychopharmacol*. 28:275–285.

1983 With D. D. Potter and S. C. Landis. Transmitter status in cultured rat sympathetic neurons: Plasticity and multiple function. *Fed. Proc*. 42:1626–1632.

1986 With D. D. Potter, S. G. Matsumoto, S. C. Landis, and D. W. Sah. Transmitter status in cultured sympathetic principal neurons: Plasticity, graded expression and diversity. *Prog. Brain Res*. 68:103–120.

With S. C. Landis, S. G. Matsumoto, and D. D. Potter. Synaptic functions in rat sympathetic neurons in microcultures. I. Secretion of norepinephrine and acetylcholine. *J. Neurosci*. 6:1061–1079.

With D. D. Potter, S. C. Landis, and S. G. Matsumoto. Synaptic functions in rat sympathetic neurons in microcultures. II. Adrenergic/cholinergic dual status and plasticity. *J. Neurosci*. 6:1080–1098.

With D. D. Potter and S. G. Matsumoto. Synaptic functions in rat sympathetic neurons in microcultures. III. A purinergic effect on cardiac myocytes. *J. Neurosci*. 6:1099–1107.

1987 With S. G. Matsumoto, D. W. Sah, and D. D. Potter. Synaptic functions in rat sympathetic neurons in microcultures. IV. Nonadrenergic excitation of cardiac myocytes and the variety of multiple-transmitter states. *J. Neurosci*. 7:380–390.

1989 With D. D. Potter. Seizure-like activity and cellular damage in rat hippocampal neurons in cell culture. *Neuron* 3:199–207.

A cell-culture approach to the study of seizure activity. *Q. J. Exp. Physiol*. 74:975–985.

1990 With W. J. Koroshetz. Seizure-like activity and glutamate receptors in hippocampal neurons in culture. *Neurosci. Res. Suppl*. 13:S65–S74.

With M. M. Segal. Epileptiform activity in microcultures containing small numbers of hippocampal neurons. *J. Neurophysiol.* 64:1390–1399.

1991 Seizure-like activity in cell culture. *Epilepsy Res*. 10:24–32.

1997 With T. H. Glick, E. G. Armstrong, S. E. Hyman, and E. M. Hundert. Neurologic education for the future: A decade of curricular reform at Harvard Medical School. *Eur. J. Neurol.* 4:102–106.

1998 With B. Murray, A. Alessandrini, A. J. Cole, and A. G. Yee. Inhibition of the p44/42 MAP kinase pathway protects hippocampal neurons in a cell-culture model of seizure activity. *Proc. Natl. Acad. Sci. U.S.A.* 95:11975–11980.

## SELECTED PAPERS BY E. J. FURSHPAN TRAINEES


1970 D. Bray. Surface movements during the growth of single explanted neurons. *Proc. Natl. Acad. Sci. U.S.A.* 65(4):905–910.

1973 D. Bray. Branching patterns of individual sympathetic neurons in culture. *J. Cell Biol.* 56(3):702–712.

R. E. Mains and P. H. Patterson. Primary cultures of dissociated sympathetic neurons. III. Changes in metabolism with age in culture. *J. Cell Biol.* 59:361–366.

R. E. Mains and P. H. Patterson. Primary cultures of dissociated sympathetic neurons. II. Initial studies on catecholamine metabolism. *J. Cell Biol.* 59:346–360.

R. E. Mains and P. H. Patterson. Primary cultures of dissociated sympathetic neurons. I. Establishment of long-term growth in culture and studies of differentiated properties. *J. Cell Biol.* 59:329–345.

1974 K. Obata. Transmitter sensitivities of some nerve and muscle cells in culture. *Brain Res.* 73(1):71–88.

P. H. Patterson and L. L. Chun, The influence of non-neuronal cells on catecholamine and acetylcholine synthesis and accumulation in cultures of dissociated sympathetic neurons. *Proc. Natl. Acad. Sci. U.S.A.* 71(9):3607–3610.

1975 C. A. Nurse and P. H. O'Lague. Formation of cholinergic synapses between dissociated sympathetic neurons and skeletal myotubes of the rat in cell culture. *Proc. Natl. Acad. Sci. U.S.A.* 72(5):1955–1959.

1976 S. C. Landis. Rat sympathetic neurons and cardiac myocytes developing in microcultures: Correlation of the fine structure of endings with neurotransmitter function in single neurons. *Proc. Natl. Acad. Sci. U.S.A.* 73(11):4220–4224.

P. H. Patterson, L. F. Reichardt, and L. L. Chun, Biochemical studies on the development of primary sympathetic neurons in cell culture. *Cold Spring Harb. Symp. Quant. Biol.* 40:389–397.

1977 L. L. Chun and P. H. Patterson. Role of nerve growth factor in the development of rat sympathetic neurons in vitro. III. Effect on acetylcholine production. *J. Cell Biol.* 75(3):712–718.

L. L. Chun and P. H. Patterson. Role of nerve growth factor in the development of rat sympathetic neurons in vitro. II. Developmental studies. *J. Cell Biol.* 75(3):705–711.

L. L. Chun and P. H. Patterson. Role of nerve growth factor in the development of rat sympathetic neurons in vitro. I. Survival, growth, and differentiation of catecholamine production. *J. Cell Biol.* 75(3):694–704.

L. L. Chun and P. H. Patterson. The induction of acetylcholine synthesis in primary cultures of dissociated rat sympathetic neurons. II. Developmental aspects. *Dev. Biol.* 60(2):473–481.

L. L. Chun and P. H. Patterson. The induction of acetylcholine synthesis in primary cultures of dissociated rat sympathetic neurons. I. Effects of conditioned medium. *Dev. Biol.* 56(2):263–280.

L. F. Reichardt and P. H. Patterson. Neurotransmitter synthesis and uptake by isolated sympathetic neurones in microcultures. *Nature* 270(5633):147–151.

P. A. Walicke, R. B. Campenot, and P. H. Patterson. Determination of transmitter function by neuronal activity. *Proc. Natl. Acad. Sci. U.S.A.* 74(12):5767–5771.

1978 S. C. Landis. Growth cones of cultured sympathetic neurons contain adrenergic vesicles. *J. Cell Biol.* 78:R8–R14.

1980 E. Hawrot. Cultured sympathetic neurons: Effects of cell-derived and synthetic substrata on survival and development. *Dev. Biol.* 74(1):136–151.

S. C. Landis. Developmental changes in the neurotransmitter properties of dissociated sympathetic neurons: A cytochemical study of the effects of medium. *Dev. Biol.* 77(2):349–361.

1981 C. A. Nurse. Interactions between dissociated rat sympathetic neurons and skeletal muscle cells developing in cell culture. II. Synaptic mechanisms. *Dev. Biol*. 88(1):71–79.

C. A. Nurse. Interactions between dissociated rat sympathetic neurons and skeletal muscle cells developing in cell culture. I. Cholinergic transmission. *Dev. Biol*. 88(1):55–70.

P. A. Walicke and P. H. Patterson. On the role of cyclic nucleotides in the transmitter choice made by cultured sympathetic neurons. *J. Neurosci*. 1(4):333–342.

P. A. Walicke and P. H. Patterson. On the role of Ca2+ in the transmitter choice made by cultured sympathetic neurons. *J. Neurosci*. 1(4):343–350.

1982 P. I. Baccaglini and E. Cooper, Influences on the expression of acetylcholine receptors on rat nodose neurones in cell culture. *J. Physiol*. 324:441–451.

P. I. Baccaglini and E. Cooper. Electrophysiological studies of new-born rat nodose neurones in cell culture. *J. Physiol*. 324:429–439.

P. Claude, E. Hawrot, D. A. Dunis, and R. B. Campenot. Binding, internalization, and retrograde transport of 125I-nerve growth factor in cultured rat sympathetic neurons. *J. Neurosci*. 2(4):431–442.

E. Hawrot. Characteristics of the association of nerve growth factor with primary cultures of rat sympathetic neurons. *J. Neurosci. Res.* 8(2–3):213–224.

1983 P. I. Baccaglini and P. G. Hogan. Some rat sensory neurons in culture express characteristics of differentiated pain sensory cells. *Proc. Natl. Acad. Sci. U.S.A.* 80(2):594–598.

1985 A. J. Doupe, S. C. Landis, and P. H. Patterson. Environmental influences in the development of neural crest derivatives: Glucocorticoids, growth factors, and chromaffin cell plasticity. *J. Neurosci*. 5(8):2119–2142.

A. J. Doupe, P. H. Patterson, and S. C. Landis. Small intensely fluorescent cells in culture: Role of glucocorticoids and growth factors in their development and interconversions with other neural crest derivatives. *J. Neurosci*. 5(8):2143–60.

A. L. Willard and R. Nishi. Neurons dissociated from rat myenteric plexus retain differentiated properties when grown in cell culture. II. Electrophysiological properties and responses to neurotransmitter candidates. *Neuroscience* 16(1):201–211.

1988 R. Nishi and A. L. Willard. Conditioned medium alters electrophysiological and transmitter-related properties expressed by rat enteric neurons in cell culture. *Neuroscience* 25(3):759–769.