Advances in CELL CULTURE

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CONTRIBUTORS TO THIS VOLUME

Antony W. Burgess

Peter Carlson

J. Gressel

Leonard Hayflick

Paul K. Horan

Stefan T. Jaronski

Timothy J. Kurtti

Mark S. Lesney

Katharine A. Muirhead

Ulrike G. Munderloh

Harry Murakishi

Advances in CELL CULTURE

Edited by

KARL MARAMOROSCH

Robert L. Starkey Professor of Microbiology
Waksman Institute of Microbiology
Rutgers University
New Brunswick, New Jersey

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LEONARD HAYFLICK

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CONTRIBUTORS TO VOLUME 3

Numbers in parentheses indicate the pages on which the authors' contributions begin.

ANTONY W. BURGESS, *Tumour Biology Branch, Ludwig Institute for Cancer Research, Melbourne 3050, Australia (231)*

PETER CARLSON, *Crop Genetics International, Dorsey, Maryland 21076 (1)*

J. GRESSEL, *Department of Plant Genetics, The Weizmann Institute of Science, Rehovot 76100, Israel (93)*

LEONARD HAYFLICK, *Center for Gerontological Studies, University of Florida, Gainesville, Florida 32611 (303)*

PAUL K. HORAN, *Department of Immunology, Smith, Kline & French Laboratories, Philadelphia, Pennsylvania 19101 (57)*

STEFAN T. JARONSKI, *Department of Entomology, North Carolina State University, Raleigh, North Carolina 27650 (183)*

TIMOTHY J. KURTTI, *Waksman Institute of Microbiology, Rutgers—The State University of New Jersey, Piscataway, New Jersey 08854 (259)*

MARK S. LESNEY, *Department of Horticulture, Michigan State University, East Lansing, Michigan 48824 (1)*

KATHARINE A. MUIRHEAD, *Department of Immunology, Smith, Kline & French Laboratories, Philadelphia, Pennsylvania 19101 (57)*

ULRIKE G. MUNDERLOH, *Waksman Institute of Microbiology, Rutgers—The State University of New Jersey, Piscataway, New Jersey 08854 (259)*

HARRY MURAKISHI, *Department of Botany and Plant Pathology, Michigan State University, East Lansing, Michigan 48824 (1)*

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PREFACE

Volume 3 of *Advances in Cell Culture* appears approximately eighty years after plant cell cultivation was first attempted by Haberlandt. His tradition of excellence has been maintained by those who have expanded cell cultivation and its numerous applications.

This volume continues the wide coverage, initiated in Volume 1, of *in vitro* culture with seven contributions on vertebrate, invertebrate, and plant cell cultivation. The important practical applications of cell culture in this rapidly developing field are stressed by several authors. This emphasis will be helpful in broadening the basic knowledge of practicing tissue culturists. Readers will find stimulating ideas as well as new concepts and fresh approaches to problems in the articles included. Facets of cell culture that may have immediate as well as long-range economic potential are presented. The contributions reflect the thinking and accomplishments of those who are in the forefront of the broad field of cell culture today. The depth and sophistication of the articles indicate current strength in the diverse areas of *in vitro* research.

In this volume, a biographical sketch has been devoted to Philip R. White, remembered not only for his pioneering plant tissue culture work but also for his contributions to animal cell culture and to *in vitro* research as a whole.

KARL MARAMOROSCH

Caroline White and Philip R. White

PHILIP RODNEY WHITE

July 25, 1901–March 25, 1968

In the first volume of *Advances in Cell Culture* a biographical sketch was devoted to Ross G. Harrison, the undisputed pioneer of vertebrate cell culture. In the second volume there was a sketch of Richard B. Goldschmidt, who started invertebrate cell culture. If now I were to feature the actual creator of plant cell culture, the biography of Haberlandt would have been proper since at the turn of the century he pioneered the cultivation of plant cells and succeeded in the isolation and in the aseptic growth of cells from various plants in a solution containing salts and sugar (Haberlandt, 1902). Haberlandt's admirable vision led him to postulate the concept that plant cells in culture possessed the capacity to develop into complete plants and that plant hormones, unknown at that time, are required for cell division, growth, and differentiation. His vision proved correct but, ironically, he himself did not succeed in attaining plant cell division *in vitro*. This breakthrough was achieved in 1939 in France when, independently, Gautheret (1930) and Nobecourt (1939), adding the auxin indole-3-acetic acid, were able to obtain unlimited growth of carrot roots *in vitro*. In the same year Philip R. White visited both laboratories in France, thus gaining firsthand knowledge of the role of auxin. Upon returning to the United States, White succeeded in obtaining a continuous callus culture of the tobacco hybrid *Nicotiana glauca* × *Nicotiana langsdorfii* (White, 1939). Only much later did it become apparent that the fortunate choice of this hybrid tissue that synthesized adequate levels of hormones was responsible for White's success (Murashige, 1979). In 1941 Van Overbeek *et al.* noticed that the liquid endosperm of coconut (coconut water or "milk") stimulated the development of *Datura* embryos *in vitro*. The "coconut milk factor" was identified by Miller *et al.* (1956) as kinetin. The discovery of other cytokinetins soon followed, and their addition to culture media permitted the cultivation of cells from diverse species of monocots and dicots. In 1957 Skoog and Miller revealed that the formation of shoots and roots depended on the balance of auxin and cytokinin in the culture medium. The following year the first somatic embryogenesis was achieved by Steward *et al.* (1958) and by Reinert (1958). Thus the prediction of Haberlandt that plant cells are totipotential was proved correct. During the next two decades practical applications of plant cell culture in agriculture and forestry followed in rapid succession, stimulated by Morel's (1960) demonstration that the shoot apex culture of orchids could be utilized

for commercial orchid cloning. Soon Morel's work became widely adopted by orchid growers the world over, and within a short time florists began to propagate various flowers and ornamentals using *in vitro* techniques. Food and fiber plants and several species of forest trees were mass produced and widely propagated. Plant pathologists obtained pathogen-free clones by *in vitro* cultivation methods, and new horizons opened through *in vitro* hybridization and plant breeding. What but a few years earlier was in the realm of science fiction is now rapidly becoming reality as described in several chapters in this and in the two earlier volumes of *Advances in Cell Culture.*

While most plant tissue workers had concentrated on the use of plant material for the improvement of plant varieties or for the study of cell and tissue growth and differentiation, White's creative imagination led him to include vertebrate cell culture as well. His influence on the development of cell culture was, therefore, more profound, and I felt that it would be proper to include his biographical sketch in this volume even though, as mentioned earlier, the beginnings of plant cell culture not only preceded him, but also the first vertebrate and invertebrate cell and tissue cultivation. White's demonstration in 1934 of the unlimited growth of excised tomato roots is usually cited as the first successful case of plant tissue culture (Murashige, 1979). This culture had been maintained by White for thirty years. According to Waymouth (1981), White carefully transferred week by week pieces of roots 2 cm in length to fresh culture medium in small Erlenmeyer flasks. "By week's end, the roots had grown to several centimeters and were ready for subculture. Meticulous records were kept, assuring that the roots were indeed the progeny of the original isolates."

There was also a personal reason for my selecting White at this time rather than one of the other important contributors to the development of plant cell culture. In 1947, while working as a graduate student at Columbia University and the Brooklyn Botanic Garden, I had the good fortune to travel with my major professor, Lindsay M. Black, to Storrs, Connecticut to attend the symposium of the Society for the Study of Growth and Development (presently the Developmental Biology Society). It was the first meeting that I ever attended, and, upon arrival, Black introduced me to his friends Philip and Caroline White. Needless to say, I was as greatly impressed by them as I was by Harrison whom I met at the same symposium. In the following years, at subsequent Growth Symposia, I realized that Mrs. White was the driving force that enhanced the tremendous ambition and ego of her husband. Although she did not contribute to plant cell cultivation directly, her impact on her husband's work and career was profound. In various,

often unconventional ways, she influenced his actions as those who knew them both will certainly recall. Thus, in a way, both never ceased to serve the cause of tissue culture which Philip nurtured through a lifetime of evolution and growth (Maramorosch, 1968). I shall try to present a picture of White as the world knew him and as history will record him, realizing fully my inadequacy to do justice to such a task.

Philip Rodney White and his twin brother Omar were born in Chicago, the sons of Henry K. White and Mary J. Pattee. His youth was spent in rural Montana. He received his bachelor's degree at the University of Montana, where he was an assistant in botany from 1920 to 1921. His early career was varied and involved numerous positions. He moved from Montana to the University of Washington as a graduate assistant from 1922 to 1923. From 1923 to 1924 he was at the Ecole Normale d'Instituteurs in Valence, France, where he taught English and became acquainted with the French language, the people, and the country. His friendship and understanding of the French people lasted his entire life. From 1925 to 1926 he worked as a microscopy technician at the U. S. Department of Agriculture. At the same time he started his graduate work at The Johns Hopkins University in Baltimore, where he obtained his doctorate. In 1926 he participated in a Johns Hopkins Tropical Expedition to Jamaica, and this first contact with the tropics was followed by a tour of duty with the United Fruit Company, from 1926 to 1928, to Jamaica, Panama, and Costa Rica. In 1928 he was appointed Assistant Professor of Botany and Plant Physiology at the University of Missouri, where he first became exposed to plant cell culture, witnessing the pioneering work of William J. Robbins, who, in 1922, succeeded in maintaining excised tomato roots in artificial media (Robbins, 1922).

From 1929 to 1930 White held a National Research Council Fellowship at the Boyce Thompson Institute for Plant Research in Yonkers, New York. At that time he met Caroline, who fell in love with him, divorced her first husband, and soon married Philip. The following year, as a Rockefeller Fellow, White went to the Institute for Plant Physiology at the University of Berlin. On his return to the Boyce Thompson Institute, he was invited by Luis O. Kunkel to join his group. At that time Kunkel was leaving the Boyce Thompson Institute to become the head of the newly created Plant Pathology Division at the Rockefeller Institute's branch in Princeton, New Jersey. During the following fourteen years in Princeton, White advanced from Fellow to Assistant to Associate of the Rockefeller Institute, and during this fruitful period he developed the techniques for plant tissue culture and the media that have since been used in laboratories all over the world.

While there, he became associated with Wendell M. Stanley, John H. Northrup, Francis O. Holmes, Armin C. Braun, George L. McNew, Max A. Lauffer, Lindsay M. Black, H. S. N. Green, William Trager, Theobald Smith, Carl TenBroek, M. Kunitz, John B. Nelson, and many others. The years at the Rockefeller Institute were perhaps not only the most stimulating for White but for all who came in direct contact with him and his brilliant work. The pace, as well as the outstanding quality of his research, did not diminish in the years that followed. In 1945 he went to the Institute for Cancer Research in Philadelphia, where he worked for five years as a member and director of the Division of General Psychology. In 1951 he joined the staff of the Roscoe B. Jackson Memorial Laboratory in Bar Harbor, Maine and began the last, and longest continuous experience of his life. While there, he continued his comparative research on tumors of plants and animals and on the development of chemically defined media for animal and plant cultures.

Upon his retirement in 1968, his interest in Theophrastus led him to study Greek and to travel to Greece to write a book in which he planned to emphasize the merits of Theophrastus' botanical treatises. Unfortunately, he was unable to complete this book. From Greece, on a National Science Foundation assignment, he traveled to India for an extended lecture tour at many universities and research institutes. When he reached Bombay, where he became ill with hepatitis, he continued to work until the last moment, recording from his hospital bed the introduction to a lecture scheduled for delivery before the Indian Council of Medical Research. His last words were heard by the audience, stunned by the sad awareness that the lecturer, whose voice so clearly came through the loudspeaker, had just departed forever. Caroline calmly delivered the lecture after she told the audience that her husband had just passed away.

Throughout his active career, White was engaged in research and was amazingly productive. In addition to numerous publications in scientific journals, he was the author of "A Handbook of Plant Tissue Culture" (1943) and of the comprehensive work "The Cultivation of Animal and Plant Cells," first published in 1954 and revised in 1963. White's influence on the development of tissue culture and on his contemporaries and students was profound. This influence was one of his great achievements, as was his ability to organize conferences and meetings dealing with cell and tissue culture. Among them were the Decennial Review Conference on Tissue Culture at Woodstock, Vermont in 1956 and the 1964 International Conference on Plant Tissue Culture at Pennsylvania State University. I was fortunate in being

able to attend both and to have the opportunity to admire White's organizational abilities and his painstaking attention to the smallest of details, as well as to his masterful handling of the financial problems involved in these international conferences.

White was also active in politics, especially in the early 1960's when his independence and fierce love of social injustice were expressed at anti-war, anti-nuclear, and civil rights meetings. He also emphasized the crucially important interactions necessary between scientists in diverse disciplines, particularly botanists and zoologists, to advance biological knowledge (White, 1955). Those who knew White personally in the period when he worked at Bar Harbor remember him as a warm human being, a brilliant experimenter, a great scholar, and a man whose inspiring example set the goal of activities for many younger followers. During this period, he gave unselfishly of his talent and was particularly devoted to helping young investigators.

Among the many honors received by White was the AAAS Prize in 1937 for his account of the ascent of water in trees. He served as president of the Tissue Culture Association from 1958 to 1959. From 1947 to 1953 he was in charge of the widely known summer tissue culture program at Mt. Desert Island Biological Laboratory.

The spirit of Philip R. White will continue to pervade the tissue culture world, and the foundation which he laid will continue to grow and strengthen in that spirit. Regrettably, he did not live to see the applications of plant cloning to agriculture, work which he pioneered. At the time of his death, many regarded him as the greatest of plant tissue culture experts.

KARL MARAMOROSCH

REFERENCES

Gautheret, R. J. (1939). *C. R. Acad. Sci. Paris* **208,** 118–120.
Haberlandt, G. (1902). *Sitz-Ber. Mat.-Nat. Kl. Kais. Acad. Wiss. Wein.* **111,** 69–92.
Maramorosch, K. (1968). *Indian Phytopathol.* **21,** 155–158.
Miller, C. O., Skoog, F., Okumura, F. S., von Saltza, M. H., and Strong, F. M. (1956). *J. Am. Chem. Soc.* **78,** 1375–1380.
Morel, G. M. (1960). *Am. Orchid Soc. Bull.* **29,** 495–497.
Murashige, T. (1979). *In* "Practical Tissue Culture Applications" (K. Maramorosch and H. Hirumi, eds.), pp. 27–44. Academic Press, New York.
Nobecourt, P. (1939). *C. R. Soc. Biol. Paris* **130,** 1270–1271.
Reinert, J. (1958). *Ber. Deut. Botan. Ges.* **71,** 15.
Robbins, W. J. (1922). *Bot. Gaz.* **73,** 376–390.
Skoog, F., and Miller, C. O. (1957). *Symp. Soc. Exp. Biol.* **11,** 118–130.