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LEONOR MICHAELIS

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A Biographical Memoir by

LEONOR MICHAELIS, D. A. MACINNES AND S. GRANICK

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AN AUTOBIOGRAPHY WITH ADDITIONS BY
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LEONOR MICHAELIS was certainly the most influential scientist, during the past half century, in the introduction of the methods of physical chemistry into biology and medicine. In this role he served both as investigator and, especially through his publications, as teacher. At the request of the director of the Rockefeller Institute, of which he was a Member, he prepared a brief account of his most unusual career. As it does not appear that this account can be improved upon, it is given below, with a few necessary alterations. To this has been appended a description of Dr. Michaelis as he appeared to his friends and coworkers. The autobiography follows:

Leonor Michaelis was born January 16, 1875, in Berlin, Germany, in an environment which was far removed from science. He entered a "Gymnasium," from which he graduated in 1893, passing the "Abiturienten Examen," which entitled the bearer of its diploma to acceptance as a student at any German university. This particular Gymnasium (Koellnisches Gymnasium), although decidedly of the "humanistic" type (in contrast to what was called "Real-gymnasium," with emphasis on science and mathematics and no Greek) happened to have, by some development in its earlier history, a creditable chemical and physical laboratory which was not much used in general but, due to the personal interest of two professors, was open to a selected, voluntary group of students. Here he learned enough chemistry so that later, after entering the university, he was

able to start with organic chemistry. As regards physics, a vivid recollection was the measurement of ohmic resistances with the Wheatstone bridge on Sunday mornings. On clear December nights a fairly good telescope permitted observations of Mars, Venus, Jupiter and Saturn, especially the eclipses of Jupiter's moons. Although he was more inclined to science than to humanistic trends, he had no difficulty with Latin or Greek. His professor, noticing his efficiency in Latin, even took the trouble to warn him against studying classical philology, because of the poor rewards, as evidenced by his, the professor's, status. However, this warning was unheeded, as Michaelis selected science when the time was ripe. With no one to advise him, and no idea of how pure science could provide a living, he chose the study of medicine as the best approach to science. He entered Berlin University in 1893.

Among his teachers may be mentioined: Waldeyer in anatomy, Oskar Hertwig in histology and embryology, DuBois-Reymond in physiology, Emil Fischer in chemistry. In his first semester, which was a short summer semester, he took a "public" (i.e. non-paid for) course in counterpoint with Professor Bellermann, an interesting fellow, of a very old school. Music ended for him with Mozart. According to him, Beethoven made mistakes, or took impermissible liberties in the manner in which he "answered" the theme of a fugue: instead of answering a fifth with a fourth, by contraction, he answered a fifth with a fifth, which forced him to modulate to the dominant at much too early a stage of the game. However, very soon, there was not time left for such diversions. In 1895 Michaelis passed the pre-medical examination ("Physicum"), with DuBois-Reymond as the Dean of the faculty, who proclaimed his passing of the examination with "A" in all disciplines. Such success was not achieved by him at the final examination in 1897, where the really medical specialties were involved, especially surgery and gynecology. However, he did pass these too. The last semester, including the final examination, was in Freiburg i. B. Even before going to Freiburg he passed his doctor's examination (which was at that time separated

from the examination for the State license for medical practice) in Berlin, in 1896.

While still a student at the university, he spent his free time in the laboratory with Professor Oskar Hertwig, and published there a paper on the histology of milk secretion, which was rewarded with a prize from the medical faculty. He published another paper on the cytology of the fertilization of the ovum of the amphibian, *Triton taoniatus,* and wrote his doctor's thesis on the determination of the direction of the first cleavage in the frog's egg. This work with embryology induced him, a year later, to write a short textbook on embryology for medical students, which in rapid succession went into nine editions, but finally his interests were so far removed from embryology that he would no longer take the responsibility of bringing it up to date for a tenth edition.

After returning from Freiburg to Berlin, he continued his research in Hertwig's laboratory, when, by some strange coincidence, he became acquainted with Professor Paul Ehrlich, who was preparing a review article on blood cytology. Michaelis had observed that during the colostrum period in the guinea pig, eosinophilic cells accumulate in the lactic gland. Ehrlich asked him to show him the histological preparations and offered him, spontaneously, a position as his private assistant in the State's Institute for serological testing and research, where Michaelis was to continue studies in staining, which Ehrlich himself no longer could pursue after entering his research in immunology. A result of these studies was the vital staining of what was later called "mitochondria," with Janus green, published in 1900. This method was introduced into this country by Cowdry.

The agreement with Professor Ehrlich was that after one year's research in his laboratory Michaelis should study clinical medicine, according to Ehrlich's theory that only a man of sufficient wealth should stay permanently in fundamental scientific research. So Michaelis became assistant to Professor M. Litten in a department of one of the municipal hospitals in Berlin from 1900 to 1904. It was a women's ward, with cases not only in various branches of internal

medicine and infectious diseases, but also, in part, gynecology. Here, in addition to his clinical duties, he established, as well as circumstances allowed, a chemical laboratory.

In 1904, a paper by Michaelis on some iron-containing inclusions in tumors of the urinary bladder aroused the interest of Professor Ernst von Leyden, who engaged him as research assistant for the newly created Institute for Cancer Research at the university clinic in the "Charite" hospital in Berlin. He was sent to Copenhagen to bring Jensen's mouse carcinoma to Berlin. During these studies he found that various races of mice had different susceptibilities for cancer transplantations: Mice from Copenhagen "took" transplantations in a higher percentage than did mice from Berlin. On another occasion he was sent to Jena to secure the first ultramicroscope, constructed by Siedenhopf and Zsigmondy, and built in the Zeiss optical works. Although Michaelis did not expect to discover the agent of cancer with it, he had an opportunity to study colloid chemistry and physical chemistry on this occasion. In 1903 he acquired the position as Privatdocent at the University of Berlin, with a thesis concerned with immunological protein precipitins. From these studies the following result may be mentioned: In serum albumin, after very short digestion with pepsin, at a time when apparently only a part of the protein has been altered by the enzyme, the immunological properties are profoundly changed. From his immunological studies of a somewhat later period, based on the experience gained thereby, the following may be listed: He was the first to observe that aqueous extracts of normal livers could be used, instead of the livers of syphilitic fetuses, for the Wassermann test, the results being in the same direction, although, for practical purposes quantitatively inferior to the latter. (Shortly therafter Landsteiner discovered that it was the alcohol extract which contained the active principle both in syphilitic and in normal organs.) Furthermore, he was the first to show that the complement fixation test in the Wassermann reaction for syphilis could be replaced by a direct precipitation test, on using extracts of syphilitic fetus of especially high efficiency.

In 1905, Michaelis received the title of "Professor," but realized

that the race for any advancement in a truly academic career was hopeless. So he accepted in 1905 the new created position of bacteriologist in the municipal hospital "am Urban" in Berlin, whose medical director was Professor Albert Fraenkel, the discoverer of pneumococcus, and who also had been a pupil of Von Leyden, but of an older generation. Here he stayed until 1922, during which time to a great extent jointly with his friend, Peter Rona, the chemist of the same hospital, in addition to his routine work in bacteriology, he became interested in physico-chemical problems. Together with his friend, he built up, in a most primitive form and on a most restricted scale, a laboratory in which several problems were investigated, among which the following may be enumerated:

(1) Problems concerning blood sugar were studied. A series of investigations was undertaken in connection with which methods for deproteination of blood serum by adsorption, especially with kaolin and ferric hydroxide, were developed. It was shown by "compensation dialysis" that the glucose of the blood exists in the free state, not combined with a protein, and that the blood sugar content of the erythrocytes is not equal to that of the plasma.

(2) The method of the hydrogen electrode was developed in order to measure the hydrogen ion concentration. It was shown that the effect of an enzyme such as invertase, trypsin, etc. depends on the concentration of the hydrogen ions, and not on the titration acidity. Just when this work was coming to a conclusion, the paper by Sorensen on the same subject was published. However, being familiar with the method, Michaelis, although deprived of the priority of this finding, extended these studies by showing that the dependence of enzyme activity on pH was of the same nature as the dependence of the dissociation of a weak acid on pH. The theory of buffers (under the name of "hydrogen ion regulators") was developed.

(3) The theory of the dissociation of amphoteric electrolytes, especially of the isoelectric point of amino acids and proteins was developed in a quantitative way, after Hardy had introduced the

concept of isoelectric point purely qualitatively. It was shown that the isoelectric point was a turning point not only for the sign of the charge, but also a maximum or minimum for other properties such as solubility, viscosity, and precipitability of certain proteins in the native state (globulins), or in the denatured state (albumins). A method for the determination of the isoelectric point was based on this principle. The criterion for denaturation of an albumin was defined as the precipitability in a buffer solution at the pH of the isoelectric point, provided pH is varied under such conditions as to keep the total salt concentration low and constant (today one would say, "on working at low and constant ionic strength").

(4) The method of electrophoresis was developed in such a way as to keep the pH constant during the flow of current and the isoelectric points were determined with this method for casein, hemoglobin, serum albumin, serum globulin, native and denatured globin, gelatine, and edestin. It was shown that the results of this method agreed with those of the method mentioned above. In contrast to previous statements by other authors it was shown that the charge of a colloid, enzyme, or even of whole bacteria, was not a property inherent in the substance alone but was dependent upon pH and that there is an isoelectric point characteristic of the substance.

(5) The theory of enzymatic activity was studied by comparing the enzymatic activity at constant pH and varied concentrations of the substrate. The concept of affinity constant, concerned with the strength at which the enzyme combines with the substrate, was developed and experimentally studied, first with invertase, then with various other enzymes. This affinity constant has been termed by British authors the "Michaelis-Menten constant."

(6) The nature of enzyme inhibitors was investigated. Two classes of inhibitors were distinguished: those which act by competition with the substrate, for the combination with the enzyme, and those which influence the rate at which the enzyme-substrate compound disintegrates into the split products and free enzyme.

(7) Research was carried out on the acid agglutination of certain

bacteria, each at a characteristic pH, such as typhoid bacillus, and several types of paratyphus bacilli; the absence of acid agglutination in *Bact. Coli.* and some intermediary properties of the various dysentery bacilli were observed.

(8) The process of adsorption was studied. As regards the absorption of electrolytes including dyestuffs, ion-exchange adsorption and on the other hand "equivalent" adsorption, i.e., adsorption of the electrolyte as a whole without exchange, were distinguished. The experiments were carried out, to a large extent, with dyestuffs as adsorbates.

(9) The theory, practice, and calibrations of the "uni-colored" pH indicators, especially the various nitrophenols, was investigated.

It is hard to imagine now how the difficulties with respect to the extremely restricted laboratory facilities as to space and equipment were overcome. In addition there was the problem of the assignment of proper working places for the many postgraduate students, including some who came from abroad, who wished to study with him, and last, but not least, the difficulties due to the fact that the city administration strongly discouraged any kind of research in the municipal hospitals.

During the war Michaelis was engaged in hospital work, in part in hospitals for tuberculosis, then on gastrointestinal diseases, and finally on infectious diseases. After the war he returned to his position as a bacteriologist in the "staedtisches Krankenhaus am Urban." In 1921, the new government bestowed upon him the position of a professor ("professor extraordinarius") for physical chemistry applied to medicine and biology at the University of Berlin, but without salary, without budget, and without a laboratory. So he did not hesitate to accept, in 1921, the offer of an industrial firm of a position in which he was given laboratory facilities for himself and his students, and where he served as a consultant for the making and cataloguing of scientific apparatus.

Less than one year later, he received a call from the medical school in Nagoya, Japan, to a professorship in biochemistry. It was an un-

usual event, since no foreigner had been engaged in a full professorship in Japan for a long time. The reason was that this medical school had just been raised from a lower grade to university rank, and the dean wished that at least one of the new professorships should be initiated, although not permanently kept, by a European. Michaelis accepted the call and never regretted it. The original offer, to remain for one year, was extended by mutual consent to three years. He had ample opportunities and many graduate students for research, in addition to his teaching duties.

One result of the studies in Nagoya was the finding of a membrane specifically permeable for cations only, the "dried collodium membrane." Other papers of that time were concerned with an extension of the theory and practice of potentiometric measurements, and the extension of the theory of pH.

During his stay in Japan Michaelis received, quite unexpectedly, a letter from Professor Jacques Loeb asking whether he would be willing to undertake a lecture tour in the U.S.A. on his way back to Europe. The result of this correspondence was that he made a lecture tour, not on a return trip to Europe, but during the summer vacation of 1924, after which he returned to Japan until the expiration of his contract a year later. On the day of his departure from Japan in 1924, he received news of the sudden death of Jacques Loeb. However, the arrangements for the lecture tour had been well prepared by Loeb and everything proceeded according to schedule. At this time Michaelis received an offer from Johns Hopkins University to be a resident lecturer for three years, to begin in the spring of 1926, after the termination of his contract in Japan. He accepted. At the expiration of his contract with Johns Hopkins he received a call to the Rockefeller Institute in New York, where he was a Member from 1929 to 1941, and subsequently a Member Emeritus.

His research in America was essentially in the fields of oxidation-reduction processes: the oxidation potentials of reversible systems; the role of heavy metals as catalysts for oxidation; and especially the role of semiquinone radicals in reversible oxidation systems. The

existence of such free radicals, existing in equilibrium with their "parent substances," even in aqueous solution, appeared to be unbelievable to almost all chemists. The paper was rejected by American journals and he had it published abroad. It was a depressing experience. Today his claims are generally accepted in every textbook. For many years, every manuscript concerned with this subject was sent by the editor of the *Journal of the American Chemical Society* to Michaelis as a referee, and never again was a paper of his on this subject rejected by any journal. The conversion of the majority of organic chemists was essentially brought about by the fact that Michaelis demonstrated the paramagnetism of the compounds in question, which left no doubt about their nature as free radicals or "odd molecules." A summary of the diversified studies along these lines is in the *Annals of the New York Academy of Science,* 1940, and a semipopular review of the problems and all their implications as regards thermodynamics and kinetics of oxidation and respiration is given in the National Sigma Xi Lecture of 1945 (*American Scientist,* 1946).

Among other subjects studied was the conversion of keratin (hair, feathers, etc.) by the reduction of the S-S bridges to SH groups, into soluble proteins, digestible by pepsin and trypsin, and reoxidation to S-S compounds with random structure, without restoration of the original form. Thioglycolic acid was used as a specific reducing agent. Michaelis later learned that this publication was the cornerstone of big industrial enterprises in the field of cosmetics. The principle was used for the "dry permanent wave," and the industry now manufactures the necessary ingredients on an enormous scale.

Another topic studied in his laboratory, in which, however, the greater credit should be given to his associate, S. Granick, is iron metabolism, including studies on ferritin and the new specific protein apoferritin.

Michaelis was elected a member of the National Academy of Sciences in 1943, a Fellow of the American Association for the Advancement of Science in 1929, and was a Fellow of the New York Acad-

emy of Sciences. He was Harvey Lecturer in 1924, and National Sigma Xi Lecturer in 1946. He received the honorary degree of LL.D. from the University of California, Los Angeles, in 1945.

Among his coworkers, some whose share in the works published jointly must be estimated at not less than 50 percent, or even more, are: Peter Rona, Maxwell P. Schubert, David R. Goddard, Carl V. Smythe, and Sam Granick.

At this point the autobiographical sketch ends.

Submitted on September 15, 1948, it covers most of Michaelis's career. However, he kept up active research until his death on October 8, 1949.

The theory of potentiometric titrations of free radicals which he had developed was applied to an extension of the pH scale in strongly acid solutions where customary pH measurements are difficult to interpret. His continued interest in the forces between molecules in aqueous solution led to studies of the dimeric resonance bond, quinhydrone type interactions, metachromatic staining, and the interaction of basic dyes with acidic polymers like heparin or nucleic acid. One of his last experimental studies resulted in the discovery of the free radical of tocopherol at liquid air temperature, which may have significance in the understanding of the mechanism of function of this antioxidant vitamin. In a review paper he discussed a general theory of step reactions and its application not only to protons and electrons but also to groups of atoms.

Michaelis could have made substantial contributions to nearly any branch of human endeavor that he chose to enter. And in so doing he would have brushed away obstacles that would have completely discouraged a man of ordinary capabilities.

One outlet for his abundant energies was music. His early interest in the theory of that subject was mentioned above, and music was an avocation throughout his lifetime. His natural talent was shown when, to entertain friends, he would create on a suggested melody improvisations in the style of any of the better-known composers.

Another field in which he could have made his mark was philology. He had a phenomenal memory and acquired languages readily. In his three years in Japan he learned to speak the difficult language of that country.

Had he chosen mathematics for his career it seems most likely that he would have made important contributions to that field. He was able to use mathematics effectively as a tool in his researches, and was influential in bringing the methods of that science into biology through his papers and books.

Through his wide reading he was conversant with concepts of mathematical physics such as the various aspects of the quantum theory and of wave mechanics, and he made use of the electronic theories of the structure of atoms and molecules in his researches.

Michaelis was under average height, wore thick lenses in his eye glasses, and was of stocky build. He was not impressive in appearance when one first met him, but his face lit up with a vivacious expression when conversations turned to matters of intellectual interest. Not always tactful with his colleagues, he was invariably patient and kindly in his dealings with younger workers.

He is survived by his wife, Hedwig Philipsthal Michaelis, and his daughters, Ilse Wolman and Eva M. Jacoby.

KEY TO ABBREVIATIONS

Advance. Enzymol.=Advances in Enzymology, New York
Advance. Protein Chem.=Advances in Protein Chemistry, New York
Aichi J. exp. M.=Aichi Journal of Experimental Medicine, Nagoya
Ann. N. Y. Acad. Sci.=Annals of the New York Academy of Sciences
Annual Rev. Biochem.=Annual Review of Biochemistry
Arch. f. Anat. u. Physiol.=Archiv fur Anatomie und Physiologie—Physiologische Abteilung. Supplement
Arch. Biochem.=Archives of Biochemistry
Arch. ges. Physiol.=Archiv (Pflugers) für die gesamte Physiologie des Menschen und der Tiere, Berlin
Arch. Hyg.=Archiv für Hygiene und Bakteriologie, Munich
Arch. mikr. Anat. Entwick.=Archiv für mikroskopische Anatomie und Entwicklungsgeschichte
Beitr. chem. Phys. Path.=Beitrage zur chemischen Physiologie und Pathologie
Ber. Deut. chem. Ges.=Berichte Deutsche chemische Gesellschaft, Berlin
Ber. ges. Physiol.=Berichte über die gesamte Physiologie und experimentelle Pharmakologie, Berlin
Berl. klin. Wschr.=Berliner klinische Wochenschrift
Biochem. Centr.=Biochemisches Centralblatt, Berlin
Biochem. Zschr.=Biochemische Zeitschrift, Berlin
Biochim. biophys. Acta=Biochimica et biophysica Acta, Amsterdam
Biol. Bull.=Biological Bulletin, Lancaster
Bull. Johns Hopkins Hosp.=Bulletin of the Johns Hopkins Hospital, Baltimore
C. rend. Laborat. Carlsberg=Comptes rendus des travaux du Laboratoire Carlsberg, Copenhagen
Chem. Rev.=Chemical Reviews, Baltimore
Chem. Ztg.=Chemicker-Zeitung
Chim. et Ind.=Chimie et industrie
Deut. Aerzte-Ztg.=Deutsche Aerzte-Zeitung, Berlin
Deut. Essigind.=Deutsche Essig-Industrie
Deut. med. Wschr.=Deutsche medizinische Wochenschrift, Leipzig (includes Vereins-Beilage)
Fed. Proc.=Federation Proceedings (Federation of American Societies for Experimental Biology), Baltimore
Fol. Serol.=Folia Serologica
Fortsch. Med.—Fortschritte der Medizin
Indust. Engin. Chem.=Industrial and Engineering Chemistry, Easton, Pa.
J. Am. Chem. Soc.=Journal of the American Chemical Society
J. Am. Leather Chemists Assoc.=Journal of the American Leather Chemists Association
J. Biol. Chem.=Journal of Biological Chemistry
J. Chem. Educ.=Journal of Chemical Education

J. Gen. Physiol. = Journal of General Physiology, Baltimore
J. Phys. Colloid Chem. = Journal of Physical and Colloid Chemistry, Baltimore
Jahrber. ges. Physiol. = Jahresberichte über die gesamte Physiologie
Jahrkurs. ärztl. Fortbild. = Jahreskurse für ärztliche Fortbildung
Klin. Wschr. = Klinische Wochenschrift, Berlin
Kolloid Zschr. = Kolloid-Zeitschrift
Med. Klin. = Medizinische Klinik, Berlin
Med. Woch. = Medizinische Woche
Münch. med. Wschr. = Münchener medizinische Wochenschrift
Proc. Soc. Exp. Biol. = Proceedings of the Society for Experimental Biology and Medicine, New York
Sc. Month. = Scientific Monthly, Lancaster, Pa.
Sitzber. Berl. haematol. Ges. = Sitzungsberichte Berliner haematologische Gesellschaft
Ther. Gegenwart = Therapie der Gegenwart
Therap. Monatsh. = Therapeutische Monatshefte
Trans. Electrochem. Soc. = Transactions of the Electrochemical Society
Ver. Beil. = Vereins-Beilage (see Deutsche Medizinische Wochenschrift)
Ver. Berl. med. Ges. = Verhandlungen Berliner medizinische Gesellschaft
Verh. Deut. Kongr. inn. Med. = Verhandlungen des Deutschen Kongresses für innere Medizin
Verh. Kongress inn. Med. = Verhandlungen des Kongresses für innere Medizin
Verh. physiol. Ges. Berl. = Verhandlungen physiologische Gesellschaft, Berlin
Verh. Ver. inn. Med. = Verhandlungen des Vereins für innere Medizin in Berlin
Virchows Arch. = Virchows Archiv für pathologische Anatomie und Physiologie und für klinische Medizin
Wschr. Brauerei = Wochenschrift für Brauerei
Zbl. Bakt. = Zentralblatt für Bakterologie
Zschr. Bal. = Zeitschrift für Balneologie, Klimatologie und Kurort-Hygiene, Berlin
Zschr. Chem. u. Ind. Kolloide = Zeitschrift für Chemie und Industrie der Kolloide
Zschr. Elektrochem. = Zeitschrift für Elektrochemie and angewandte physikalische Chemie
Zschr. exp. Path. = Zeitschrift für experimentelle Pathologie und Therapie
Zschr. ges. exp. Med. = Zeitschrift für die gesamte experimentelle Medizin
Zschr. Immunforsch. = Zeitschrift für Immunitätsforschung und experimentelle Therapie, Jena
Zschr. klin. Med. = Zeitschrift für klinische Medizin
Zschr. Krebsforsch. = Zeitschrift für Krebsforschung, Berlin
Zschr. phys. Chem. = Zeitschrift für physikalische Chemie, Leipzig

Zschr. phys. diät. Ther.=Zeitschrift für Physikalische und diätetische Therapie, Leipzig

Zschr. physiol. Chem.=Zeitschrift für physiologische Chemie (Hoppe-Seyler)

Zschr. Untersuch. Nahrungsmitt.=Zeitschrift für Untersuchung der Nahrungs- und Genussmittel, Berlin

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