Trends in Pharmacological Sciences

April 1983

# Otto Krayer 1899–1982

Otto Krayer arrived in Berlin in 1927 when Professor Paul Trendelenburg accepted the Chair of Pharmacology and brought Krayer, his first assistant, with him from Freiburg. Krayer was from Kondringen, a village just north of Freiburg, where the Schwarzwald and the Rhine valley meet. He talked little about his early life in English, though he would do so in German, interestingly reverting to a pronounced Baden dialect that was difficult to understand even for other Germans. He was a happy member of the village community. He was a keen and good skier, enjoying the long run from the heights of the Schwarzwald to the outskirts of town. He obviously loved the countryside and the foundation of his life-long interest in botany must have been laid at this time.

Krayer had completed his medical studies in Freiburg but had spent periods elsewhere, in Munchen and Berlin. He had been obliged to return to Freiburg, near home, when inflation made the transmission of money across the country impossible. On receiving his MD in 1926, Krayer went directly into pharmacology. Within a short time he was on the staff in Freiburg, Germany. Garry, later Professor of Physiology in Glasgow, and Anichkov of Leningrad worked in this laboratory and he became their life-long friends.

Shortly after his arrival in Berlin, Trendelenburg started a losing battle with tuberculosis. Over the next few years, Krayer found himself increasingly in charge of the Department and when Paul Trendelenburg died in 1931, Krayer was already the Acting Head. To appreciate the events of the next few years, it is important to understand certain unwritten, but immutable, rules of academic preferment in Germany. First, the top Chair, which Berlin certainly was, will be filled by someone who is already the incumbent of another Chair. Second, if any Chair is offered to a non-incumbent, it must be accepted or the hope of a Chair forever foresworn. No second offer of a Chair will be made to someone who has refused a first offer.

Heubner came from Gottingen to Berlin as Chairman. Krayer became Professor Extraordinarius in 1932. He expected to be offered a Chair soon and had brilliant prospects in Germany. He was ambitious to start building the best department of pharmacology ever. In 1933 he was offered the Chair of Pharmacology in Dusseldorf. The Professor of Pharmacology in Dusseldorf had been Ellinger but he was dismissed because he was said by the Nazis to be Jewish. Krayer refused the Chair. He had never met Ellinger and knew little of him; but he did not want a Chair that had been made vacant for such a reason. The Ministry of Education pondered the refusal for a while, then ordered, rather than invited, him to Dusseldorf. Again he refused. The Ministry replied by dismissing him from his post and even forbidding him access to any university in Germany. Krayer was completing Volume 2 of Paul Trendelenburg's *Die Hormone*, left unfinished at Trendelenburg's death. Friends brought books and journals from the library for him to complete the work. On 31 December 1933 he gave the proofs to Springer-Verlag and left Germany. He did not even visit Germany again until 1948.

The main themes of Krayer's scientific interests were evident even before he left Germany. Although from the first he was interested in molecular mechanisms of drug actions, he was not willing himself to leave integrated physiological systems for the sake of simple *in-vitro* systems. Rather, he encouraged the work on simpler systems and then sought to establish the applicability of molecular findings to integrated physiological systems. He well understood that establishing the quantitative contribution of postulated molecular mechanisms of a drug to a known effect of the drug in a whole animal requires as much, often much more, work than the generation of the hypothetical mechanism itself. He was always pained when effects of ridiculous concentrations of drugs in test tubes were assumed to immediately explain all effects in intact subjects. Such casual assumptions became increasingly frequent although one dares to hope that they have been decreasing in more recent years.

Krayer worked, for example, on the cardiovascular system. He was interested in the cardiovascular effects of drugs not thought of primarily as cardiovascular drugs. (He did, however, work intermittently on digitalis: a German pharmacologist once told me that all German pharmacologists were required to 'do time' on cardiac glycosides in expiation for Withering not having been German!) He worked on the cardiovascular effects of neosalvarsan and of tissue extracts (the latter before Gaddum and von Euler). The work on neosalvarsan was occasioned by clinical reports of cardiovascular effects after intravenous injection of neosalvarsan in the treatment of syphilis. There were several other occasions when a line of Krayer's research was started by clinical findings. The work on neosalvarsan began a life-long interest in the complex reflexes arising from physiological receptors in and around the heart, as exemplified by the Bezold-Jarisch reflex. In a different

*Otto Krayer*

vein, this time the coronary sinus, and exploiting the exquisite sensitivity of the eserinized leech muscle to acetylcholine, he and Feldberg demonstrated the appearance of acetylcholine (or, rather, as they said, a substance with acetylcholine-like activity) in the blood of the coronary sinus of the dog when the vagus was stimulated. This was the first demonstration in a mammalian system of the effect Otto Loewi had shown in the frog.

On leaving Germany, Krayer went to London. H. H. Dale, A. V. Hill and others, with help from the Rockefeller Foundation, were preparing assistance for medical scientists leaving Germany. Krayer joined E. B. Verney's Laboratory at University College, London and was always grateful to Verney for his gracious reception. Although he was only in London for a short time, he became proficient in the heart–lung preparation of Starling and realized its potential in analysing the cardiovascular effects of many of the agents that interested him most.

In 1934 Krayer accepted the position of Visiting Professor and Head of the Department of Pharmacology at the American University of Beirut. He personally taught the whole course of pharmacology, just as he had done in Berlin, following the German tradition. His 3 years in Beirut were the least productive of research in his career but he inspired his students – some even into research careers. He also thoroughly enjoyed his life in Beirut: the hot sunshine, the forested hills and the antiquities. He also felt a great affinity for the Arab people.

While in Beirut, Krayer made his only visit to the USSR, attending the 15th Physiological Congress in Moscow in 1935 and then taking a trip alone, as he supposed, to Georgia. After an exciting ride through the Caucasus in an open bus over a new, but dusty, military road to Tblisi, he found himself doubting what should be his next move. When he started to make enquiries, as best he could, he immediately found a helpful individual who, not only spoke German, but seemed to know him and what he wanted to do. He gratefully accepted the help though he always wondered how 'alone' his trip had been. The story has a counterpart. Many years later, Anichkov arrived in Boston, with a party from the Soviet Union. He bounced into Krayer's office and gave him a bear-hug, then said he must bring in the man from the State Department. When Krayer asked what State Department man, Anichkov replied: 'The spy, but such a nice spy.'

Harvard celebrated its tercentenary in 1936 and Krayer made his first visit to the USA as the Representative of the American University of Beirut. It was felicitous that the American University should have chosen its Visiting Professor of Pharmacology as the representative to the celebration, because Harvard happened to be seeking a pharmacologist to lead its department. The great Reid Hunt had reached the end of his career. Krayer was able to meet luminaries of the Medical School and it was even found possible to make him a temporary Lecturer so that he could give some lectures on pharmacology to the medical students. The outcome was that, although he returned to Lebanon for a time, 1937 saw him installed as Associate Professor of Pharmacology at Harvard.

Once again, he was teaching pharmacology more or less single-handed. The only teaching member remaining from Reid Hunt's department had become mainly an administrator in the Dean's office.

In 1938, H. B. van Dyke left the Chair of Pharmacology at the Peiping Union Medical College for Columbia University and Krayer was invited to succeed him in China. He was greatly attracted to the position and, I think, inclined to go. One of the factors in his decision, however, was a petition by the Harvard medical students to the administration asking that he be persuaded to stay. Petitions by medical students in those days were a great rarity, and Krayer was clearly moved by the appreciation. He also probably looked at the departments of Walter B. Cannon and of Baird Hastings and saw that his ambition to build a great department of pharmacology would not be served by going to China. He settled in Boston and married Ruth Philipp, a physician whom he had known from Germany, and bought a large, dignified house in West Newton, a suburb of Boston. Krayer was a man of enormous natural dignity. He was not in the least pompous, and was very approachable. Small children sought him out in a crowd and climbed in his lap. He was awesome on first encounter but any hint of fear rapidly dissipated (except in the laboratory).

In the laboratory, Krayer was an artist who naturally expected everyone to achieve his standards. Whenever he entered the laboratory of anyone doing an experiment which yielded graphic recordings in real time his attention was immediately attracted to features of the recordings that detracted from their aesthetic perfection. Adjustments would have to be made. In the earlier years, the recorders were kymographs and Krayer brought kymography to its highest level as an art. He was sad that the polygraphs that succeeded kymographs were less able to show the most salient results in elegant graphical form, clear at a glance. I did not work with polygraphs so was spared the anguish of trying to make the tracings beautiful, but he took the same artistic pleasure in making figures for publication and that I could not escape. The departmental drawing board was stationed beside a door to his office and a botched line would miraculously summon him from his office for an over-the-shoulder look. He never, ever raised his voice to anyone, under any circumstances, in all the years I knew him. But he could make a botcher very, very uncomfortable. In encountering tech-

nical difficulties in preparing a heart–lung experiment, he was known occasionally to address the dog in German, but no one around understood spoken German very well, especially Krayer's dialect, so it was assumed to be a harmless incant, though perhaps it wasn't. His dedication to a good experiment was passionate and it was hard for him to contain his exasperation when things did not go perfectly. When he was young, he did not always succeed, but as he grew older he became calmer, at least outwardly.

The war came and major building of the department was postponed. Despite being relatively recent arrivals from Germany, the Krayers seemed to have suffered little trouble during the war years. They lived quietly, as they did all their lives, and Krayer's activities centered on the department. He introduced laboratory experiments into the pharmacology course for medical students, something that had never been possible in Germany. He continued to give demonstrations during the course of lecture. His main helper was Mr George whom he inherited from Reid Hunt.

Krayer is well-remembered by his students as a clear and helpful lecturer, with animation and emphasis but without histrionics. He taught them more than they thought they needed to know about materia medica and toxicology, but he also taught them well about general medical pharmacology which they appreciated. As to the materia medica, he held that samples of all drugs mentioned in a lecture should be exhibited and he assembled a large collection of samples for this purpose. His interest was undoubtedly related to his interest in botany. As time passed, more of the drugs mentioned became undistinguished white powders so, as Krayer gradually relinquished the actual teaching, exhibiting died out. Krayer complained, but did not make it an issue. Toxicology has always been taken seriously in Germany. Krayer was appalled at the general state of academic toxicology in the USA and ensured that the medical students heard at least a little about heavy metals, arsenic, etc. If he had been heeded more perhaps we would not have the sorry state today where pharmacology has little input into toxicology and essentially pharmacological judgements in toxicology are made by people with little appreciation of pharmacology.

One of Krayer's early co-workers in Boston was Rafael Mendez, who had left a career in pharmacology in Spain to become a member of the government of the Republic. With the coming of Franco, Mendez left Spain hurriedly and reverted to pharmacology. It gave Krayer much satisfaction to help a refugee, especially one who was such a good pharmacologist. Mendez left for a distinguished career in Mexico City.

During the remainder of Krayer's incumbency there was a steady stream of talented people coming to the laboratory and working for a few years before moving on to good positions elsewhere. Some of these people were, like Krayer, physiological pharmacologists such as Moe, Acheson and Farah. Krayer was, however, interested from the first in mechanisms and encouraged the biochemical inclinations of Ellis and, later of Weiner. He was also interested in pharmacokinetics and encouraged Goldstein's early studies. He regarded endocrinology as part of pharmacology and worked with Astwood, and recruited Riggs, Munson and Kenny. He was even tolerant of the possibility of behavioral pharmacology and encouraged a man with primarily psychological training, W H. Morse, to join the department.

As the study of molecular pharmacology became more of a possibility Krayer sought for people with relevant interests. Lubin came with a Ph.D. in biophysics from Massachusetts Institute of Technology to work on the biophysics of muscle and of excitation–contraction coupling. Briggs came to work on the molecular chemistry of muscle-contraction. When it appeared that nuclear magnetic resonance techniques might contribute to understanding the binding of drugs to receptors, Jardetzky was recruited from Pauling's laboratory and installed with his machine in a basement. By this time, Goldstein had left for Stanford but his legacy proved much stronger than he may have expected when he departed

During this period, Krayer was involved in a continuing series of studies on veratrum. He was interested in all aspects of this substance. He was interested in the botany and grew *Veratrum vivide* in his garden (and the gardens of his colleagues). He developed the medicinal chemistry by bringing a superb chemist, Uhle, into the department to work with him and he collaborated with Kupchan in the Department of Chemistry. He was interested, of course, in the cardiovascular pharmacology of the alkaloids and clarified differences between the effects of ester and of amine alkaloids The complex reflex effects the former elicited from receptors in and around the heart tied in with his earliest interests. He identified the anti-acceleratory effects of the latter on the heart. Finally, he was interested in the clinical applications of the agents and worked on these first with Meilman and then with Flacke. Many of the visitors to the department worked with him on veratrum or related problems and Wood (Mayo), Reiter (München), Kosterlitz (Aberdeen), Matallana (Cali), Kottegoda (Colombo), Innes (Winnepeg) and Paasonen (Helsinki) are among the 25 or so co-authors of the papers on veratrum.

One of the main tools used by Krayer in much of his research in Boston was the dog heart–lung preparation, after Starling. He used it as others used a Warburg apparatus, or isolated guinea-pig ileum or catheterized human heart: as a means of obtaining a certain kind of physiological and pharmacological information on a variety of different agents. He collaborated with Aub, Nathanson and Zamecnik in studying the influence of antitoxin on the action of *Clostridium* toxin on the heart; with Wollenberger on direct cardiac actions of barbiturates and local anesthetics and Astwood and Alper on effects of corticotrophin and $\alpha$-intermedin He brought the techniques of preparation, conduct of the experiments and recording of the results to a high state of art. He used the preparation as a dramatic and informative teaching demonstration for many generations of students. Weiner filmed such a demonstration, with Krayer being assisted by his long-time helper, W. Mosimann. The work was characterized by elegance and soundness and, in at least one instance, by leading to a 'break-through'.

In 1955 Krayer read a letter published in the *New England Journal of Medicine* reporting instances of heart failure in patients on reserpine, a drug recently introduced for the treatment of hypertension. Ever alert for pharmacological contributions to the solution of clinical problems, he added reserpine to the blood of the heart–lung preparation to see whether reserpine had direct, negative inotropic effects. To his considerable surprise, the predominant effect was an increase in heart-rate. The heart-rate of a heart–lung preparation is increased by catecholamines and Paasonen was making catecholamine assays in the laboratory at the time. He assayed the blood from heart–lung preparations and showed that the addition of reserpine led to a large increase in norepinephrine concentrations in the perfusing blood. So reserpine liberated norepinephrine from stores in the heart. This was the first demonstration of such an effect of reserpine.

Krayer realized the importance of the discovery, but made no attempt to exploit it. He told his finding freely to visitors to the laboratory. Although the finding was published in abstract in 1957, definitive papers did not appear until 1958. In 1955 it was found and reported by others that reserpine depleted tissues of their 5-HT and strong advocacy positions were taken as to whether the effects of reserpine, notably on the CNS, were mediated through norepinephrine or 5-HT. The evidence was inadequate so the battles were fought instead with intimidating words even though, to an interested outsider, the best guess seemed that both would be important in their own way. Krayer viewed the scene with disdain and did not stay in the field. Later, the issues were further complicated when it was found that reserpine also released dopamine and other amines.

During the late 1950s conditions at last became propitious for Krayer to build his department in numbers. New and much expanded quarters were being planned and prepared for the department, under Krayer's constant and vigilant eye. Support for research by the National Institutes of Health was reaching its zenith. In a few years the number of active workers in the department grew from half a dozen or so to 30 or more. Blinks, Kelleher, Koch-Weser, Marshall, Ryser and Waud joined the department, as did a whole group of neurophysiologists under Kuffler. The number of graduate students increased and the small numbers from earlier years, which had included van Maanen, Maling, Root and Harris, became much larger as, among others, Brimijoin, Fleming, Langer, Muskus, Routledge and Smith joined them.

Krayer had been seeking a neuropharmacologist for the department for some years. He followed with admiration the work of Kuffler at Johns Hopkins and tried to recruit one of Kuffler's students, but two or three slipped through his fingers. It then appeared that Kuffler himself might be moved, and Krayer set about mobilizing support in the school to make this possible. Enthusiasm was easy to generate, but real-estate less so, and it became clear that all the space for neurophysiology would have to come out of the new quarters for pharmacology. Krayer had qualms, but he did not waver, and the department was augmented by Kuffler himself, Hubel, Wiesel, Dudel, Potter and Furshpan. Dudel returned to Germany but shortly thereafter Kravitz and Nicolls joined. It was an outstanding accession for the School, but too much to be assimilated by the Department of Pharmacology. Although relations were excellent between many of the people in the Laboratory of Neurophysiology and Neuropharmacology and many of the people in the rest of pharmacology,

the laboratory remained a clearly defined entity. Krayer had an intense loyalty to pharmacology: not just to pharmacological science of high quality done under any guise, but to pharmacological science done in a Department of Pharmacology. He worried that by fostering neurophysiology he may have foreclosed some future possibilities for the Department of Pharmacology. He undoubtedly had, though an appraiser of contributions to the School as a whole is likely to forgive him.

U. Trendelenburg arrived from Oxford in 1957 and quickly established himself as a focal figure in the department, both in its research and teaching. U. Trendelenburg was the son of Krayer's first mentor, P. Trendelenburg. U. Trendelenburg had become considerably anglicised by his sojourn in Oxford, while Krayer had become thoroughly (though not quite completely) Americanized, but they shared almost coincidental ideals and goals. Typically, this led Krayer to worry about showing favoritism, so that poor young Trendelenburg had to prove himself academically more than if his name had not been Trendelenburg.

Krayer retired from the University in 1966, and although he remained in Newton for some years, had little further to do with the Department of Pharmacology. The department he had built disastrously lost many of its members after his retirement and he agonized over this decline. Despite his thorough Americanization, he never fully accepted that an American department, no matter how good the research, careful the coverage of the field and excellent the teaching, is not an Institut; the organization rarely survives its creator.

One of Krayer's great strengths was his absolute honesty and trustworthiness. When he advised a course of action, one could be quite sure that it represented his judgement of what was best for the individuals involved, untinged by what was best for Krayer. As Goldstein said in his Sollmann Oration, when Krayer advised him to go to Edinburgh for a period, he went without question: 'If he had told me to go to Tierra del Fuego, I would have gone'. The complete loyalty to his colleagues and helpers engendered a reciprocal loyalty to himself. He was dedicated to the pursuit of excellence but did not believe this pursuit required a lack of consideration of others or ruthlessness. He confidently expected people to rise to excellence. While on a few occasions this caused some difficulty, as with one or two graduate students who stubbornly refused to rise to his expectations, in the great majority of instances people did respond by doing better than they had thought they could. He never discarded a human being. Anyone who had ever been associated with the department had a permanent claim to his concern. His was a remarkable combination of paternalism and *laissez-faire*. He would take everyone's personal problems to heart, but would not interfere, or even comment, unless asked for help. He treated the staff of the Department as scientific colleagues and would not direct the lines of their work. He was reluctant to give scientific advice, unless asked, even when he thought one of his staff was following a relatively unprofitable line. It is a privilege to have known him.

**Acknowledgement**

I would like to thank Professor U. Trendelenburg for his help in preparing this appreciation.

P. B. DEWS

*Laboratory of Psychobiology, Harvard Medical School, 25 Shattuck Street, Boston, MA 02115, U.S.A.*

# Mosaic

## Risk assessment of drug carcinogenicity

*The proceedings of a workshop held in Skokloster, Sweden, 19–21 April 1982.*

Carcinogenicity is one of the most rapidly-developing areas in toxicological research at present, and there is no shortage of opportunities to learn about the new developments at meetings, symposia and workshops. This workshop, which was organized by The Association of the Swedish Pharmaceutical Industry (LIF), and to which some 60 scientists from Sweden and several other countries were invited, had set itself a rather specific goal: the discussion of a few 'case histories' of drugs which had, because of certain positive or ambiguous results in mutagenicity or carcinogenicity tests, caused difficult problems with regard to risk-assessment in man.

In his opening address Zbinden (Zurich) reviewed the strengths and weaknesses of the rodent life-time bioassay, and the many pitfalls such as poor management and performance at every level, sampling errors, false diagnoses, genetic and nutritional influences, stress factors and spontaneous diseases, that often affect the outcome of these ambitious experiments. These pitfalls are responsible for the fact that a considerable percentage of these studies raise more questions than they provide answers. Another reason why the rodent life-time bioassay has lost its former predominance as the decisive carcinogenicity experiment is that several mechanisms are now known to underlie tumor development in such experiments. Williams (Valhalla, N.Y.) described their subdivision into 'genotoxic' and 'epigenetic' mechanisms, and reviewed the many new *in-vitro* and *in-vivo* techniques which are used to assess DNA damage resulting in DNA repair, chromosomal aberrations, sister chromatid exchange, specific locus mutations and malignant cell transformation.

The observation that phenobarbital, which is not a human carcinogen, caused liver tumors in chronically treated mice and rats has led to the recognition that tumor promotion (a concept developed for mice skin-tumors) is also possible in the liver. Tumor promotors belong to the group of epigenetic carcinogens whose hazards must be assessed differently from those of the genotoxic carcinogens.

The true hazards of the genotoxic carcinogens were underlined by Schmähl (Heidelberg) who demonstrated the cancer-producing properties of alkylating drugs in animal experiments and in man. He pointed out that for such agents the *in-vivo* bioassay was still an excellent tool which might even provide important clues with regard to the organ affinity of potent carcinogenic drugs.

The problem of organ affinity of genotoxic carcinogens was also discussed by Neumann (Würzburg) who used dimethylaminostilbene as an example of a strong electrophilic carcinogen. Covalent binding of such compounds to DNA is considered the primary lesion. This is a particularly important concept since it permits the study of dose–effect relationships at very low doses. Such data are important if one wishes to extrapolate from animal experiments conducted with high doses to the low level exposure of man.

A very difficult problem which has plagued toxicologists for many years is the interpretation of the many tumors induced in experimental animals by chronic treatment with steroid hormones; this was discussed by El Etreby (Berlin). As exemplified by the estrogens, which cause renal carcinoma in male hamsters, leiomyomas in guinea-pigs, lymphoid tumors in mice, ovarian carcinomas in dogs and adrenal cortical carcinomas in rats, these substances are notorious for their species differences. In the case of the mammary tumors induced in beagle bitches by certain contraceptive progestogens, it was possible to demonstrate that this effect was also due to a species-specific sensitivity resulting from a high affinity for cytoplasmic progestogen receptors. Moreover, these hormones markedly increased growth hormone levels in the dog. The so-called 'clean progestogens', which did not produce mammary tumors in dogs when given at 25× the