
AT THE BEGINNING OF THE WAY

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In those remote days I studied at the biological faculty of the Kyiv National University and used to see Platon Grygorovych in both the chair and the institute. He produced an impression of an arrogant, self-satisfied person due to his confident imposing walk. Later, when I was a fourth-year student and future physiologist, he conducted practical classes for our group, and I was very surprised to notice that in reality there was no arrogance in his behavior, and my former *notion* of him seemed to be impossible. His attitude to us became at once friendly and informal. We heard that he was working hard on his Doctoral, dedicated to the processes of excitement and inhibition in two neuron reflector arcs of the spinal cord. I regret that our group did not use to attend his lectures which were highly appreciated by the following graduations. But even in short introductory part of lecture he, as a teacher, demonstrated broad and profound knowledge, ability to explain simply and understandably those complicated tasks we were faced with. Platon Grygorovych was a brilliant lector. My under-graduation and graduation works were in physiology of digestion. But it was Platon Grygorovych that made me turn sharply towards electrophysiology. Once while we were talking about our future work after graduation from the University, Platon Grygorovych said that physiology is, first of all, experimental science, and one can learn the most advanced methods of exploration at the department of general physiology headed by Prof. Danylo Semenovych Vorontsov. Soon I realized that the department was the leading electrophysiological center in the country, and it was the place where I started working as a laboratory assistant. My first tutor was Danylo Semenovych

Vorontsov, who had become an academician by that time. He was a scientist of the highest professionalism. When Platon Grygorovych, a professor at that time, was invited to the Bogomoletz Institute of Physiology of the Academy of Sciences to get the position of the head of the department, he became my second tutor.

I remember the very beginning of my working activity at the department. It was a huge room on the ground floor of the red building of the University. One third of it was occupied with two screening chambers with electrophysiological equipment. It was a place where Danylo Semenovych, Platon Grygorovych together with S.D. Kovtun performed their experiments. There were two laboratory assistants, Lida Savoskina and me, and a technician Vira Danylivna whose duty was to keep our overalls snow-white and well-starched, and the parquet polished until it shined. Her participation in experiments was to keep ether anaesthesia in cats operated on, and a cat quite often fell asleep together with Vira Danylivna. The students of the University were making their undergraduate and graduation works there. We, laboratory assistants, used a small dark nook adapted for works with photo documents of the results of experiments to stay alone and whisper. We recorded nervous activity from the screen of an oscillograph with a help of an ordinary camera. Copying technique was not available that time, so we had to take photos of articles and antique scientific editions. I had a personal place at one of the working tables and during the whole month I was making miniature electrodes for stimulation of the cat's brain. First of all, I had to become good at soldering the thinnest wires. That work turned out to be a sort of checking my skill and capacity for work. After I had passed it, I was allowed to participate in experiments with Danylo Semenovych. A month later Danylo Semenovych heard that I worked free of charge, without any salary. Shocked and angry, he went to the administration part of the University, and only after that I was taken on the staff.

I should notice that I was not the only one to start working with Danylo Semenovych in such a way. Platon Grygorovych passed more serious training: in addition to soldering, he studied the principles of electro- and radioengineering and assembling equipment. Native industry didn't produce equipment to provide electrophysiological investigations, and we lacked money to buy it abroad, so we were forced to create a lot of things by ourselves.

Formation of Platon Grygorovych as a brilliant experimenter and a perceptive, deeply interested in Science scientist took place under the guidance of Danylo Semenovych. That was Danyla, as he was named by his pupils, who inspired Platon Grygorovych in his devoted love to physiology which he had preferred to medicine. As you know, Platon Grygorovych graduated from both the biological faculty of the University and the therapeutic faculty of the medical institute. He also studied at the conservatoire and dreamed to become an outstanding pianist.

Work in foreign laboratories was of great importance for his further achievements. At first, he worked in Shanghai with Prof. Chang who had come from USA

to socialist China. After that he worked in Canberra, Australia in the laboratory of famous neurophysiologist J. Eccles. I must notice that that time, in 1959, trips abroad were limited for the soviet citizens and only very promising scientists managed to get them with great difficulties.

The time Platon Grygorovych got the position of the head of department at University's institute was unforgettable. It was accompanied with great changes in the building interior. Of course, we did not have money to acquire new furniture, so we moved about "pre-revolutionary" one which was available in the department. And after that all staff together with the head scoured the cupboards, tables and covered them with colourless varnish. The boxes with alkaline accumulators were painted in white. Indeed, it became more pleasant to work in a renovated room.

That was the time of intensive and fruitful development of general physiology abroad. The introduction of cardinal new method of exploration, such as extrathin glass microelectrodes served the stimulus and the beginning of revolutionary changes. Those microelectrodes were micropipettes with the tip size less than $1\ \mu\text{m}$, filled with electrolyte as a current conductor. The electrodes penetrated through protoplasmatic membranes easily and allowed direct study of physiological processes in single cells of a small size. The English scientists Alan Hodgkin and Andrew Huxley, who together with J. Eccles became Nobel Prize winners, played an enormous role as the founders of the membranous theory to explain physicochemical events underlying nervous processes. It became quite obvious that the surface membrane is an amazing protein lipid structure with absolutely special physicochemical properties. We must add by right the name of Danylo Semenovich Vorontsov to these famous names. As far back as 1946, while studying electric responses of the spinal cord neurons in a frog, Danylo Semenovich came to a conclusion that they were of a membranous origin, and he made a perceptive suggestion that excitement and inhibition were different physiological processes. Later, it was proved experimentally by Platon Grygorovych.

Science in the USSR was surviving a hard transition period following persecutions which it had endured during the period of the fight against formal genetics and so-called anti-Pavlov distortions in physiology. In the country where such famous geneticists as N.I. Vavilov, N.K. Koltsov, and S.S. Chetverikov had just recently demonstrated wonders, we, the university graduates of 1955, knew nothing. That fundamental biological discipline was banned and it was contemptibly named either pseudoscience or mendelism-morganism. Gaps in knowledge we had to fill in on our own. Our university mate V.A. Kordium became later the founder of modern genetics in Ukraine. In physiology, the ideas of the cytophysiologist from Leningrad academician D.N. Nasonov about paranecrosis predominated. Together with the cytologist V.J. Aleksandrov he offered a protein theory of damage and excitement. That theory did not give any role to the surface membrane in cell vital functions, and excitement was regarded as the result of impairment in the cytoplasm protein structure and its sorption property.

We should consider the great merit of Platon Grygorovych that he used a new approach to study cell electric phenomenon and promoted its development in the USSR. To do that, it was necessary to possess not only scientific clarity of vision but fearlessness, because he was likely to be caught in servility towards the West. The microelectrode method turned out to be very fruitful in neurophysiological investigations. As a matter of fact, we had to start our work from zero overcoming all sort of difficulties. To begin with, we needed new equipment. Platon Grygorovych gave his private money to an institute engineer who was an experienced developer of radio measuring technology to fabricate it. Perhaps, you do not know that the salaries of the higher school lecturers and researchers in the USSR were very poor, really symbolic. We needed an amplifier with particular input characteristics joined to a cathode oscillograph and "machine", as we called an apparatus to stretch glass micropipettes. Platon Grygorovych made a lot of things himself which was quite usual in those difficult post-war years. A micro-manipulator to help administration of a microelectrode into a single cell was not available that time, so Platon Grygorovych used an old microscope with gliding desk instead of it. Only a few years later his first postgraduate Ju.P. Limansky developed an oil micromanipulator. Microelectrodes were made of refractory glass. Glass-blowers who worked only at the chemical faculty of the University supplied us with capillary components to them for separate pay. Further we faced with another problem, dealing with heated platinum loop we used in "machine". Although platinum was considered non-burnable, in fact, it burned out. As only wire was available that time, so it needed rolling. How to roll the wire? It was a difficult question. It was my idea to offer Platon Grygorovych to put the wire under a tram. A tram track was near the University, and Platon Grygorovych agreed with me willingly. He decided to ensure my safety in case of unforeseen circumstances, so he was keeping an eye on my work out of the window. Fortunately, everything ended successfully. Absolute methyl spirit was an intermediate filler for microelectrodes. We could not boil those microelectrodes inside an exhaust-hood because this just was not available. Shortly after, Platon Grygorovych published very useful and informative textbook "Microelectrode technique", which became a handbook for a lot of researchers.

Thus, we started recording state potentials. The first object to research was the tailor's muscle of a frog. Platon Grygorovych was easy to work with in spite of the difference between our official status and regalia. There was no formalism, but only friendliness and approachability. I got an independent field of work. I will never forget how at first we were helpers-assistants to each other. While I was administrating a microelectrode into the muscle, Platon Grygorovych worked with a camera. Soon I so well mastered with experiments that I could work on my own. In 1957, at the conference in Leningrad when I reported the results of our study, I "got through a battle" with the supporters of sorption theory and managed to prove that a state potential really pre-existed. That discussion prompted

the representatives of Leningrad's school of physiologists not only to change their mind but also to turn to the microelectrode studies. Similar conferences became traditional and they were often held in Kyiv. The scientific results were summarized, and strategic trends in exploration of the cells in excitable tissues were discussed at the conferences. Nowadays, scientists from all over the world attend our conferences in Kyiv. Weekly seminars with tea-drinking are also traditional. The seminars were initiated at the Institute of Physiology at the University. The initiator of them was Danylo Semenovych who, in his turn, was taught in Saint-Petersburg by the famous physiologist M.Ye. Vvedensky. The research workers from other institutes also visited the seminars. That time a lot of people got hooked on microelectrodes.

Shortly later, Platon Grygorovych headed the laboratory of general physiology (later department) which was organized specially for him at A. Bogomoletz Institute of physiology of the Academy of Sciences of the USSR. He took L.A. Savoskina as a laboratory assistant and me as a research worker to that laboratory. The other researcher was the postgraduate student of Danylo Semenovych Vania Semeniutin. At first, we worked in the same room. Then the postgraduate student Jura Limansky appeared at the department. In the beginning, that heavily-build and imposing ex-head doctor of big tuberculosis sanatorium was sometimes mistaken for Platon Grygorovych. The second postgraduate student was the physicist Vladimir Mayskiy who was stoically doing operations on frogs. Then, the first woman, the postgraduate student Iryna Vladymyrova joined us. All of them are still working at the Institute. Platon Grygorovych showed himself not only as a brilliant experimenter but also as an excellent organizer. Within a short time, he created a high-quality scientific team. His department was becoming something like Mecca for those scientists who decided to study microelectrode techniques. That scientific direction started intensively developing at various institutes of the former Soviet Union. Numerous works by Platon Grygorovych and his colleagues have become classic. He gave the Science a pleiad of outstanding scientists and initiated biophysics and molecular physiology. Today his followers are recognized by the international scientific community as a school of Academician P.G. Kostyuk.

Although my contribution to this school is quite modest, I am proud of being his first follower. Soon after the beginning of my work with Platon Grygorovych he offered me the choice of two subjects for independent research: either state potentials in different cells or ion mechanisms of state potential formation. I chose the latter which seemed to be much more difficult than the first one but it was more interesting. The peculiarity of Platon Grygorovych as a scientific supervisor is that he offered one a wide sphere for activity, in every possible way supporting the initiative of the students to do something new and interesting and directing them to solving the main problems. He is rather an active observer. He regularly taught us to look for the main idea as unforgettable Danyla used to say and as he did. He was accurate in everything up to pedantry. Only a few people

know that during the first years of our work together Platon Grygorovych organized regular inspections of our desks and when he managed to find disorder in any of them its content was thrown away on the floor. His other habit was never to be late. He never agreed to be a co-author in the supervised works if he had not taken a direct part in them.

We had to start at the very beginning to clear up that state potential was really created by trans-membranous gradients of main inorganic ions. It aroused the necessity to measure their intracellular concentrations. We needed a flame photometer to do that but it was not available in either the Institute or University. Fortunately, we succeeded to get it in the Institute of Geology. Their photometer was home-made, and only several years later our institute could buy a photometer of industrial production.

I remember an unpleasant situation with one of my articles sent to Leningrad which was reviewed in biased, savage way. It was the first and the last time when I saw that Platon Grygorovych could be indignant. He dictated me answers to the reviewer in which he blamed him in a sharp and ruthless way for inability to understand the work matter. Subsequently, one of the physiologists from Leningrad confessed to Platon Grygorovych that he had been the author of that review and he had liked the answer.

Determining the content of ATP and creatinine phosphate in the muscles was one of my initiatives. The high energy of phosphorus links in their molecules can be used for different cellular processes. My work was carried out with help of unique research instruments such as inhibitors of metabolism. I would like to mention that we revealed uneven distributions of ions between different intracellular structures and organoids. The final accord of that study was making selective microelectrodes which allowed us to record rather activities than concentrations of potassium and sodium ions in the cytoplasm of muscular and neuronal cells. Microelectrodes made of glass AS-1 became a principal new method to measure pH in the cytoplasm.

Over half a century my life has been linked with the Institute of Physiology. The time has flown and a lot of things have changed. For example, the modern high block of our Institute was built thanks to Platon Grygorovych's efforts. *But he is still the same*, he is my teacher. I am an elderly person now, but his erudition, devoted love to Science, energy multiplied by untiring inquisition, his deep honesty is still a stimulation factor for me.

I would like to complete my reminiscences with Shakespeare's words: "Where the thought is strong, there the action is full of force".