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HISTORY OF MEDICINE / ИСТОРИЈА МЕДИЦИНЕ

Historical retrospective of the Neurophysiological Laboratory of the Faculty of Medicine in Novi Sad

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SUMMARY
Over the course of several decades at the Faculty of Medicine in Novi Sad, fundamental studies in the field of neurosciences were of great importance and were continually kept up-to-date with global scientific achievements. These studies were applied by using the stereotactic method, single-unit recording, and electroencephalography. The Laboratory of Neurophysiology was established in 1965 and since 1978 microelectrode and microiontophoretic techniques important for the registration and analysis of the activity of individual neurons were fully developed. Under the great influence of Russian neurophysiological school (P.K. Anokhin, K.V. Sudakov), the emphasis was on the study of Anokhin's theory of functional systems. Recently, epilepsy, brain ischemia and the influence of different medications, auxiliary medicinal products, and physical agents (electromagnetic radiation) on the central nervous system, behaviour, learning and investigated pathological conditions have been studied. Scientific collaboration with renowned institutions in the country and abroad has been established, numerous scientific projects have been carried out, expert international meetings have been organized, and numerous significant studies have been published. These results have often been the basis for further clinical investigations and the improvement of preventive or curative treatment of patients. Researchers from the Laboratory participated in the education of new generations of neurophysiologists, encouraging their scientific curiosity and love for fascinating mechanisms of the nervous system.
Keywords: history, 20th century; physiology, history; schools, medical/history; Serbia

INTRODUCTION

The purpose of this study was to present the evolution of neurophysiology at the end of the 20th and the beginning of the 21st century at the Faculty of Medicine in Novi Sad. There was a tendency for neurophysiological research to keep pace with contemporary attitudes within the Russian school of neurophysiology, but at the same time respecting the achievements and trends of Western physiological thought.

ESTABLISHING NEUROPHYSIOLOGY

The beginning of educational and scientific work in the Kingdom of Serbs, Croats and Slovenes in the field of physiology is related to a famous Austrian physician and professor from the Faculty of Medicine in Leipzig, Dr. Richard Burian (1871–1954). He was invited to organize the educational process. In 1920, at the Faculty of Medicine in Belgrade, Dr. Burian became a professor and contributed enormously to establishing the Institute of Physiology, which even today bears his name [1]. The University of Novi Sad and the Faculty of Medicine were established on the same day, on May 18, 1960. In order to organize lectures, many prominent domestic and foreign professors were invited. In 1961, the Department of Physiology was founded. The first director of the Department and the first professor of Physiology was Prof. Aleksandar Sabovljev (1907–1963) from the Faculty of Medicine in Sarajevo, a former student of Prof. Richard Burian. He founded the basic concept of work and development of Physiology in Novi Sad as an interdisciplinary branch of medicine [2].

In October 1963, Prof. Radmilo Anastasijević (1915–1975), also Prof. Richard Burian's student, became the director of the Department of Physiology. With the support of state institutions, the initial problems were solved by the construction of new buildings and by providing financial resources for purchasing specific equipment in scientific and educational purposes.

The founder of Neurophysiology at the Faculty of Medicine and the Laboratory of Neurophysiology (established in 1965) was Prof. Mihailo Bajić. From 1961 to 1967, Prof. Bajić, as a scholar of the USSR, stayed at the Department of Normal Physiology at the Sechenov First Medical Institute in Moscow. He worked under the guidance of a brilliant follower of I. P. Pavlov, academician Pyotr Kuzmich Anokhin (1898–1974) himself, author of the theory of functional systems (1968) [3]. Functional systems are extremely dynamic, autonomous central–peripheral organizations, united in cyclic organization in order to provide the ultimate

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result, useful for an organism. According to Anokhin, operations and architectonics of functional systems include the following: afferent synthesis, decision making, forming the action result acceptor, efferent synthesis, the action itself, and recurrent afferent information about the result. Prof. Bajić conducted examinations of bioelectrical activity of the brain of experimental animals (electroencephalography) in the process of formation of conditioned reflexes after the application of strychnine, which facilitates synaptic transmission and the effect of acetylcholine. He followed the dynamics of formation of conditioned reflexes and registered the reduction in the number of involved brain structures over the time of its consolidation. The results are integrated in the candidate dissertation, which was successfully defended in 1966 in front of the Council of The Russian Academy of Sciences, acquiring the scientific degree of the Candidate of Medical Sciences of the USSR. At that time, in the Socialist Federal Republic of Yugoslavia there were only 21 doctoral theses defended in the field of medical sciences [4]. Prof. Bajić also defended his doctoral dissertation in Novi Sad in June of 1969 [5]. In the dissertation, he highlighted the significance of reticular formation in the processes of conditioning, learning, and excitation of higher parts of the nervous system.

In the neurophysiological laboratory in Novi Sad, Prof. Bajić introduced stereotactic method for implanting microelectrodes, which enabled the registration of a single-unit activity in various brain structures in different conditions and during the application of biologically active substances. Prof. Bajić was a visionary, a wide-range scientist, but also an excellent manager. At the time he was the dean of the Faculty (1973–1977) and the director of the Institute of Preclinical Disciplines (1975–1990), Prof. Bajić significantly improved the activities, especially of the neurophysiological laboratory, thanks to the acquisition of modern equipment, by developing cooperation with other institutes in the country and abroad, and by generating quality staff for dealing with the demanding and futuristic research in the field of neurosciences. For merits in the development of neurophysiology and international cooperation, Prof. Bajić was awarded I.M. Sechenov Gold Medals in 1978 and 2005 in Moscow.

THE BEGINNINGS OF NEUROPHYSIOLOGICAL RESEARCH

Since the Department of Physiology did not yet possess an EEG machine, research in the field of neurophysiology actually began at the Provincial Children's Hospital. The first EEG machine at the Department of Physiology (a 14-channel EEG of the Galileo brand – model E 14b), was purchased in 1971 through the funds of the Community of Medical Scientific Institutes of the Republic of Serbia (Figure 1). In the same year, Pavlovian chamber was constructed for EEG studies. It consisted of three parts: detection of EEG in humans, in animals, and for sleep studies. It was a rarity at the time and unique in former Yugoslavia. One of the first works in the field of neurophysiology that emerged from this laboratory was the master thesis

Figure 1. EEG analysis in neurophysiological laboratory on the Galileo EEG in June 1974 (left to right: professors: Bajić, Sudakov, and Lažetić) (photo from the archive of the Department of Physiology)

"Neurophysiological aspects of the effects of neuroleptics and anti-Parkinsonian drugs" by Prof. Zvonimir Lević (1936–2009), a famous neuropsychiatrist, later also the director of the Belgrade Clinic for Neurology [6].

Research of the nervous system was slow since the tracking of activity of the central nervous system is more complexed than tracking any other system in organism. Thus, plenty of researchers of the neurophysiological laboratory and other segments of the Faculty used EEG,the stereotactic method and the technique of registering single-unit activities in examinations of various brain structures. Special focus was on the changes of spontaneous bioelectric activity of different brain structures in terms of the formation of functional systems of behaviour.

Prof. Bogosav Lažetić made a series of studies on registering EEG activity of rabbits during repeated light stimulation and during feeding behaviour (Figure 2), starting his experiments in Moscow and finishing them in Novi Sad. He examined the structure of functional systems that are the basis for orienting response [7]. He defended his doctoral dissertation in 1979.

Prof. Danka Filipović studied alimentary and motivational excitation of *nucleus caudatus* and sensorimotor cortex and their changes under electro-cutaneous and light stimulation. She registered EEG and neuronal single-unit activity during electro-cutaneous and light stimulation [8]. Registered changes in neuronal activity of *nc. caudatus*

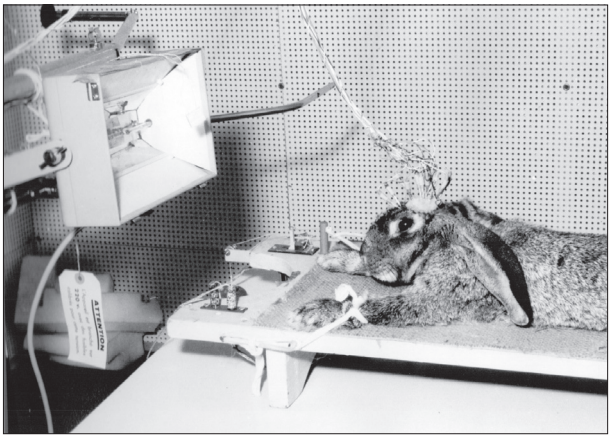

Figure 2. Experimental environment during the making of one of the doctoral dissertations in the Laboratory [7]

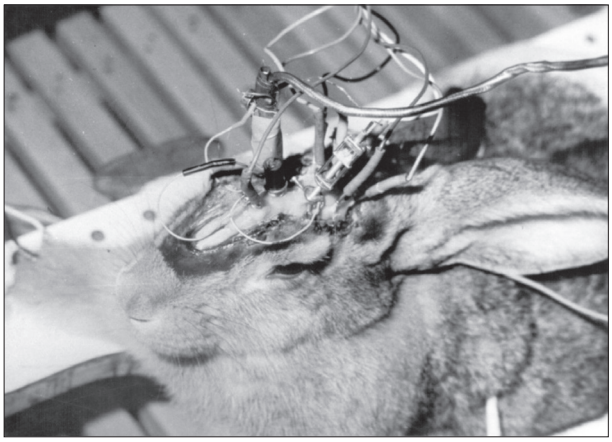

Figure 3. The figure shows simultaneous registration of EEG activity and activity of single neurons using microelectrodes implanted in deep subcortical brain structures on an experimental animal (rabbit) [8]

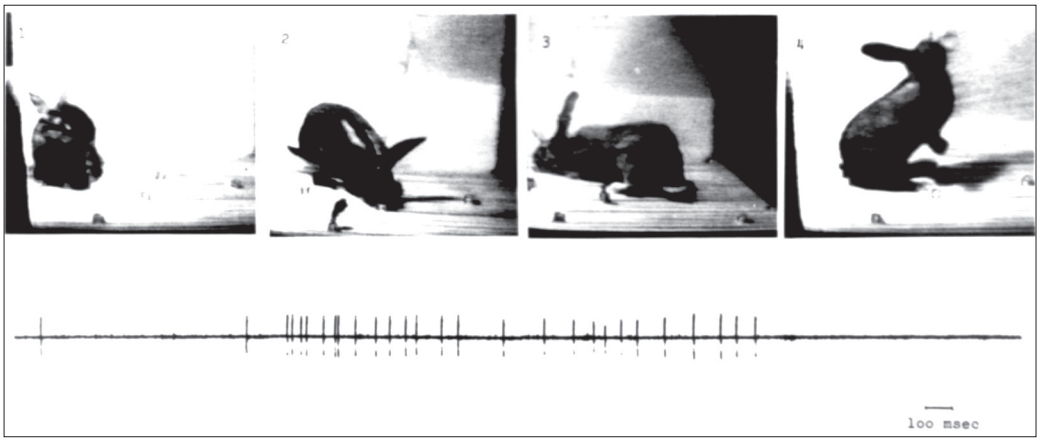

Figure 4. Orienting and searching reaction is a fundamental biological reaction; the figure shows a rabbit with implanted microelectrodes in *nucleus caudatus*; the neuron was active during the search for food and orienting in a newly introduced space (the frames are cut out from a video made in the Neurophysiological Laboratory [8]

and sensorimotor cortex in the act of alimentary behaviour have indicated that examined structures participate in the realization of coordinated motor activity, learning and emotional states related to alimentation as dominating motivation. It was also found that specific components of studied structures were involved in certain stages of registered forms of behaviour (Figure 3) [9, 10].

Prof. Vesna Ivetić also registered EEG and single-unit activity. Her field of research was low- and high-frequency electrostimulation of hypothalamus, hippocampus, and reticular formation and evoked effects on bioelectric activity of the cortex in hungry and satiated animals. Simultaneously, the influence of these stimulations on the breeding cycle and behaviour of experimental animals was also registered. It had already been discovered that hippocampus was associated with motivation, emotions, learning, memory, and goal-directed activity, but Prof. Ivetić studied the way of activating the hippocampus in the realization of some individual forms of behaviour. She elaborated and explained the activity of neurons of the hippocampus (in its areas CA-1 and CA-3) in alimentary behaviour [9, 11]. Her dissertation confirmed O'Keefs findings of "place cells" in hippocampus and showed that there are neuronal ensembles (so-called "goal cells") in the hippocampus, whose neuronal activity correlates with the development of the goal-directed alimentary behaviour [12]. Professors Ivetić and Filipović were among the first in neurophysiological examinations in the world who monitored neurophysiological functions on laboratory animals during free and feeding behaviour recording it on video tape (Figure 4).

Later on, a microiontophoretic method was also applied in investigations in the Laboratory. Prof. Đorđe Sterio simultaneously examined EEG and single-unit activity during the microiontophoretic application of primary neurotransmitters – acetylcholine and noradrenalin. The focus of his doctoral dissertation was the influence of cycloheximide, an inhibitor of protein biosynthesis, for which he discovered it slowed down the process of learning. He also concluded that arousal in motivational hypothalamic structures highly specific for orienting response leads to the synthesis of proteins in sensorimotor cortex [13]. Electrophysiological and neurochemical mechanisms of learning that were later in the focus of examinations conducted in the Laboratory aimed to unravel the complexity of this mechanism [14, 15]. It became obvious that not only the whole brain has an integrative function, but also a single neuron itself.

Guided by experience and knowledge of older colleagues from neurophysiological laboratory, Prof. Nada Naumović's first field of research was microiontophoretic application of cycloheximide, actinomycin D, and acetylcholine, and their effect on the learning process and activity of neurons and on cortico–subcortical relations. These experiments were aimed at revealing how neurochemical processes regulate the electrogenesis in neurons and which new properties occur in the electrogenesis in the course of learning [16]. As a young scientist, eager for new knowledge, she participated in a European training program in neural correlates of cognitive processes with presenting, as well as in her own scientific research. The essence of interest of Prof. Naumović during her investigation on the doctoral thesis was the examination of experimental atherosclerosis, cerebral ischemia, and influence of a specific calcium channel blocker (nimodipine) on their development and it's neuroprotective or neuromodulatory effect on brain activity [17, 18]. Based on the findings obtained on experimental animals (rabbits), clinical studies were performed in patients following an ischemic type of stroke. The results showed that nimodipine, in addition to known effects, exhibits anti-atherogenic and neuroprotective effects [19].

INTERCONNECTION BETWEEN BASIC RESEARCH AND CLINICAL WORK

Since 1993, Prof. Sterio has continued his successful career at the Department of Neurosurgery at the NYU School of Medicine, where he used the microelectrode technique for the treatment of Parkinson's disease in humans [20].

Prof. Filipović and Prof. Ivetić completed their specialization in neuropsychiatry and subspecialization in neurophysiological diagnostics, and Prof. Naumović in physical medicine with medical rehabilitation. Studies on experimental animals have gained new value because they became the basis for close cooperation with clinics and for the improvement of their work. The outcome of this developmental course was targeted research, which was carefully planned in close collaboration with clinics, and results that improved the concepts of prevention and provided a better outcome of therapeutic treatment of patients with neurological disorders [21]. Certainly, from this kind of cooperation, a series of multidisciplinary scientific publications emerged.

SCIENTIFIC COLLABORATION

The cooperation between Yugoslav scientists and Russian physiological school began in the middle of the 20th century. In 1956, Prof. Ljubisav Rakić from Belgrade (1931–) started the international cooperation with institutions in Moscow, USSR. In the early 1960s, Prof. Bajić maintained and strengthened the cooperation during his stay in Moscow. Two years after he became the dean of the Faculty, he established the Institute of Preclinical Disciplines in 1975, which created a new level of cooperation with the Moscow Institute.

In 1971, a protocol for Scientific-Technical Cooperation between Yugoslavia and the USSR was signed in Belgrade. A visit of a Russian academician Konstantin Viktorovich Sudakov (1932–2013) to Novi Sad in 1974 (Figure 1) institutionalized the scientific cooperation. It was actively maintained for decades, up to 2005. The main directions of research were general mechanisms of behaviour and motivations and integrative activity of neurons. On the basis of this cooperation, several interstate scientific projects were convened and even a few symposia with monographs as the topic were organized. After the last one, organized in 1998 in Novi Sad, on the 100th anniversary of Anokhin's birth, an extensive monography titled "Basic and clinical aspects of the theory of functional systems" was published [22].

Many associates of the Institute of Normal Physiology from Moscow worked in Novi Sad as well at the international station at the Institute of Marine Biology in Kotor. This institute was opened in 1961 (at that time it was known as the Bureau for Marine Biology). Because of its foreign policy interests, American Government partially financed these investigations through the Public Law 480. The research at the Institute was also observed by the Central Intelligence Agency and its report from 1973 (declassified in 2009) emphasized significant progress in the field of neurophysiology in Yugoslavia among all other medical fields [4]. At the Institute's the Laboratory for Brain Research were investigated the characteristics of single-unit activities of abdominal ganglia of the sea slug *Aplysia depilans*, which is analogous to biological object of *Aplysia californica* (used by Eric Kandel, the Nobel Prize winner in 2000, in his decades-long research). By the application of well-established microelectrode and microiontophoretic techniques on this model, the researchers from the Laboratory of Neurophysiology from Novi Sad gave significant contribution to these studies [23]. The characteristics of some neurons of ganglia were examined as well as the influence of different substances on their activity and mechanisms of learning.

Successful cooperation was also maintained with the laboratories of medical faculties in Belgrade, Niš, Kosovska Mitrovica, Banjaluka, and Sarajevo as well as with the Faculty of Sciences in Novi Sad. Together with the Military Technical Institute in Belgrade, the influence of various nerve agents on the activity of neurons and the possibility of epileptiform discharges due to the applications of diagnostic ultrasound were studied. By the last decade of the previous century, a joint research was realized with collaborators at the University of Rostov-on-Don, especially with the A.B. Kogan Research Institute for Neurocybernetics. Many publications came out from this collaboration [24].

Researchers from the Institute of Preclinical Disciplines actively participated in international meetings of the International Brain Research Organization (IBRO), the European Brain and Behaviour Society (EBBS), and the European Neuroscience Association (ENA). Fruitful cooperation and validity of scientific results of the Institute has been recognized in scientific community. In August 1987, the Institute was bestowed with the honor of organizing the 19th Annual

Meeting of the European Brain and Behaviour Society in the city of Novi Sad. Prominent and distinguished physiologists from Japan, with whom Prof. M. Bajić and associates had established correspondence and an exchange of scientific thoughts, participated in the conference. Scientific articles from this congress were printed in entirety by an Oxford-based publishing house – Pergamon Press [9].

MODERN TENDENCIES

By virtue of Prof. Bajić's undertakings, the first computer was purchased and a new position for a master in electrical engineering was created in 1981. As a result, all subsequent research at the Institute of Preclinical Disciplines had computerized data processing that was targeted and specially designed for each type of research carried out. The purchase of DELTA 340/10 computer system (Iskra-Delta, Kranj, Yugoslavia) with 128 KB of internal memory was a huge financial investment, but it enabled the analysis of recorded bioelectric potentials in real time, which was a capability available to only a few centers in the world at the time.

During the 20th century, a very important issue of influence of artificial electromagnetic fields on the human organism appeared. Thorough examination of this influence has been conducted at the Department of Physiology. The Laboratory for Magnetobiological Research, formed in the 1980s, used different devices, which were the basis of numerous studies, to expose experimental animals to magnetic radiation [25]. Specially designed appliances were used for exposing animals to constant and variable electromagnetic fields and morphofunctional changes in central nervous system were documented. It was shown that the main link to the damages in the central nervous system is in fact the vascular system, which consequently leads to secondary changes in the nervous tissue. Many of these studies found their place in monographies dedicated to the topic of magnetobiology [26].

Since the early 1980s, a significant part of the research in neurophysiological laboratory has been dedicated to the study of hyperexcitability of the nervous system and epilepsies [27]. Electroclinical characteristics of a wide spectrum of substances on experimentally induced epilepsies in laboratory animals have been studied [28]. These studies found their clinical application in 1995, when the Cabinet for Clinical Neurophysiology was established. It was dedicated not only to routine clinical work, but also to research activities. EEG characteristics after frequent use of cellular phones was investigated [29]. In the end, by the acquisition of modern appliances, the registration and analysis of cognitive evoked potentials (P300) are performed [30]. Beneficial effects of intermediate physical activities on cognitive functions have also been documented [31].

CONCLUSION

The Laboratory of Neurophysiology at the Faculty of Medicine was developed at a very particular time. Although it has always been a part of the University of Novi Sad, at the same time it changed four countries and at least two forms of economic and social organization. Researchers from the Laboratory succeeded in preserving scientific autonomy, and with great scientific zeal and devotion managed to develop it. Professors who led the Laboratory endeavored to keep up with modern standpoints as well as to be competitive with the achievements on all levels of international scientific thought. This became possible by the openness for cooperation and appreciation of all the achievements and trends of scientific research conducted in the USSR, as well as throughout Western Europe, USA, Japan, and other parts of the world.

In the Laboratory of Neurophysiology, the stereotactic method was applied in electroencephalographic, microelectrode, and microiontophoretic examinations to monitor brain functions during different activities of organisms. At that point in time, Novi Sad was one of a handful of centers in the world in which neuronal activity was registered during free behaviour of animals and during their goal-directed behaviour. Monitoring of bioelectrical manifestations in the central nervous system in different functional states of experimental animals and in different forms of their behaviour gave significant contribution to the understanding of neurophysiological mechanisms, as well as to defining certain structures involved, in particular the functional system and forms of behaviour of an individual.

The results obtained in the Laboratory are published in numerous scientific publications in prestigious international journals. International multidisciplinary scientific meetings have been organized in Novi Sad, and researchers have presented their work and gave lectures by invitation at international expert meetings.

The outcomes of basic studies have found their meaning in clinical application. In this sense, the Laboratory was directly involved in improving the prevention strategy for epilepsy and stroke, as well as in improving the therapeutic outcome of patients with neurological disorders.

Professors and researchers of the Neurophysiological Laboratory have been unselfishly and devotedly transferring their knowledge and experience to new generations of researchers. Based on this work and research, hundreds of student, graduate, master, doctoral, and scientific expert papers have been written. This extensive educational work created the new staff of the Faculty of Medicine in Novi Sad, who distinguished themselves at many departments and significantly contributed to the existence, development, and credibility of the Faculty of Medicine, University of Novi Sad.

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Историјска ретроспектива неурофизиолошке лабораторије на Медицинском факултету у Новом Саду

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САЖЕТАК

На Медицинском факултету у Новом Саду су током више деценија велики значај имала фундаментална истраживања у области неуронаука, која су константно ишла у корак са глобалним научним достигнућима, а спровођена су применом метода стереотаксије, микроелектродне регистрације активности појединачних неурона и електроенцефалографије. Лабораторија за неурофизиолошка истраживања је основана 1965. године, а од 1978. године у потпуности је разрађена микроелектродна и микројонтофоретска техника, значајне за регистрацију и анализу активности појединачних неурона. Под великим утицајем руске неурофизиолошке школе (П. К. Анохин, К. В. Судаков) акценат је био на изучавању теорије функционалних система Анохина. У новије време су истраживани епилепсија, исхемија мозга и утицај различитих медикамената, помоћних лековитих средстава и физикалних агенаса (електромагнетних зрачења) на централни нервни систем, понашање, учење и истраживана патолошка стања. Установљена је научна сарадња са истакнутим институцијама у земљи и иностранству, успешно су реализовани научни пројекти, организовани стручни међународни скупови и објављиване бројне значајне научне публикације. Ови резултати су често били основа за даља клиничка истраживања и унапређење клиничког и превентивног рада. Истраживачи ове лабораторије су учествовали у едукацији нових нараштаја неурофизиолога, подстицали њихову истраживачку радозналост и љубав за фасцинантне механизме функционисања нервног система.

Кључне речи: историја, 20. век; физиологија, историја; медицински факултет, историја; Србија