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Henryk Kadyi's preparatory techniques — exploring anatomical traditions of Vienna, Krakow and Lviv

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Abstract: Henryk Kadyi's (1851–1912) work in anatomical research is closely related to researching fundamental problems of preserving anatomic specimens for science and teaching. The goal of this study is to describe the professional life of Henryk Kadyi, to highlight his achievements for modern anatomy and to use his story to demonstrate the importance of mentoring, international scientific communication and cross border cooperations for scientific success.

The development of novel conservation methods gained him enormous respect of the scientific community and secured him a place in the history of medicine. Kadyi was educated in Vienna and shaped by Hyrtl and Langer. He then started residency in Krakow, where he was supervised by Teichmann, a disciple of Hyrtl. There, he received a call for professorship in Lviv. At each location, he was influence by local conditions, which improved his anatomical skills. This is a good example of the interconnection of knowledge, skills, scholarship, and scientific tradition between academic centers — in this case, Vienna, Krakow, and Lviv in the second half of the 19th century. Our paper focuses on Kadyi's preparatory techniques with a particular emphasis on the influence that the education and skills acquired in Vienna and Krakow had on them and how they were shaped when Kadyi was already an independent researcher in Lviv.

Keywords: anatomy, anatomical preparations, history of medicine, Henryk Kadyi, history, 19th century, Lviv, Krakow, Vienna.

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Materials and Methods

The paper uses original works and studies devoted to the history of preparatory methods in anatomy. It conducts a comparative analysis following historical methodology.



Discussion

Historical analysis

Henryk Kadyi (1851–1912), professor of anatomy at Lviv University, was regarded as one of the most eminent researchers in preparatory techniques. He studied medicine in Krakow and Vienna (1870–1875), then worked at the Department of Anatomy at the Jagiellonian University in Krakow under Professor Ludwik Karol Teichmann (1823–1895). Teichmann is recognized as a master of preparatory techniques, which enabled him to achieve several important discoveries, especially in the anatomical structure of the lymphatic system [1]. Kadyi was co-working with Teichmann for several years, having the opportunity to learn and practice the art of preparation, especially when it comes to injection masses and corrosive techniques. In Krakow, he began his scientific project to fully describe the spinal cord's vascular system. In agreement with Teichmann, exploring several materials and techniques, Kadyi was willing to start a close scientific collaboration with Professor Albert Adamkiewicz (1850–1921), whose name is to this day associated with the so-called Adamkiewicz artery (arteria radicularis magna). Adamkiewicz, from 1879 head of the Department of General and Experimental Pathology at the Jagiellonian University, was interested in the same research field. Yet, the planned cooperation never came to be for clearly non-scientific reasons.

There is little doubt that when Kadyi moved to Lviv, he had already mastered the art of anatomical preparation. It should be noted that Kadyi, during his time in Krakow, had been a pupil of Teichmann, who in turn had been introduced to the most modern techniques by Joseph Hyrtl (1810–1894), one of Vienna's leading anatomists. Remarkably, this means that both student and teacher (Kadyi and Teichmann) had received anatomical training from Hyrtl during their respective early careers.

It is important to note the connections between Joseph Hyrtl, Karl Langer, Ludwik Karol Teichmann, and Henryk Kadyi as an example of the transmission of knowledge between academic centers — Vienna, Krakow, and Lviv, which, in some respect, can be described as scientists' relay. By analyzing the characteristics of the methods they used in preparatory techniques, their personal attitude to research, and their intellectual background, we strive to display these interdependencies.

We point out that Hyrtl, Langer, Teichmann, and Kadyi shared the same attention to the vascular system, working on the best injection masses and corrosive methods to achieve their goals. The deep historical research on those subjects, going back to the works of Frederick Ruysch (1638–1731) and Jan Swammerdam (1637–1680), was shared by them to check proper pathways for the new solutions to the problems they faced. We underline that attitude towards perfection of methods in preparatory practice, with the thought that "it can always be better" as a rule constantly present. It successfully led Henryk Kadyi to his intensive works on masses intended for microscopic and macroscopic examination of anatomical preparations, developing new substances for staining the gray matter of the brain or research on gutta-percha injections, which was one of Teichmann's fields of inquiry, too.

Masters and pupils

In 1869, Henryk Kadyi (1851–1912) enrolled in the Medical Faculty at the Jagiellonian University (after graduation from Lviv Gymnasium), attending classes for two semesters, including lectures on human anatomy given by Prof. Ludwik Karol Teichmann. Then, he moved to Vienna to

continue his studies. Already deeply interested in anatomical studies, he came under the influence of Professor Karl Langer (1819–1887), who was a pupil and successor of Joseph Hyrtl (1810–1894). Langer, one of the most talented and authoritative researchers of topographical anatomy, paid much attention to the vascular system, working, among other subjects, on the vascular system of the tubular bones [2]. It should be noted that the first paper delivered by Kadyi focused on the variations in the arrangement and course of the arm's veins. The author shows and explains that they can be reduced only to one common type, although with three topological variations [3].

Having obtained his doctorate in Vienna in 1875, Kadyi returned to Krakow, where, in 1876, he obtained the position of assistant to Professor Teichmann at the Department of Descriptive Anatomy at the Jagiellonian University. It was during that time when he perfected his preparatory techniques. Teichmann is already known as a leading figure in the field and a world-known authority on the anatomy of the lymphatic system [4]. He was able and willing to share his knowledge and art with his young assistant. Kadyi became a close coworker of Teichmann, especially on setting masses, including the later famous *Teichmann putty mass*, as evidenced by the references made by its inventor [5]. However, Kadyi, contrary to his master, became increasingly interested in comparative and developmental anatomy, in which he differed from the "classical" descriptive anatomy. Kadyi's *O oku kreta pospolitego z punktu widzenia porównawczego i anatomicznego* [A Comparative and Anatomical Study of the Eye of the Common Mole] published in 1878, in which he gave a detailed description, both histological and anatomical, of the organ of vision in this mammal should be considered as a good illustration of his above-mentioned attitude towards comparative anatomical problems. This work was published contemporaneously to his habilitation thesis.

Realm of preparatory techniques — early period

In 1878 Kadyi became interested in preparatory techniques when he discussed and evaluated the glycerin-carbolic acid method Zygmunt Laskowski (1841–1928) developed for fixing soft tissue [6]. Kadyi's remarks on Laskowski work bring up some important information for our topic about the early introduction of Kadyi to glycerin and carbolic acid when he was still in Vienna. He recollects:

"[...] being an anatomical demonstrator in Vienna, I had the opportunity to experience the advantages of glycerin and carbolic acid in anatomical technique. In the years 1873 and 1874, the Department of Anatomy at the Josephinum, under the direction of Prof. Langer, suffered from a lack of materials because of the extraordinary influx of students, which prompted Prof. Langer to introduce the injection of carbolic acid. The liquid we used there had a slightly different composition than that recommended by Prof. Laskowski: we mixed 5 parts of glycerin with 2 parts of crystalline carbolic acid solution in an equal or double amount of high-proof alcohol. Two or three liters of this liquid injected into adult corpses were enough to protect them from putrefactive processes for 3 or 4 weeks."

Then, Kadyi writes: "I must mention here the preservative fluid, tested in the years 1872 and 1873 by Dr. Jurie under the leadership of Prof. Langer, which contains 4 parts of ordinary sugar and 2 parts of saltpeter to 28 parts of glycerin. The use and effectiveness of this solution are the same as Prof. Laskowski's carbolic glycerin." Finally, it concludes: "It follows that the main factor in these concomitant preservations is the action of glycerin. Carbolic acid and similar additives only stop the putrefactive process that has already begun" [6].

Then, Kadyi informed his readers that glycerin had been used for years and was still present in Professor Teichman's department. He also gave us a short but instructive description of his experiments in preparing soft tissues.

"I have long since tried to store such preparations in water containing about 5% carbolic acid, with fairly successful results. The muscles at first turn pale as the blood pigment is drawn out of them, but then, after a few weeks, they again take on a pink color, which gradually darkens to a chestnut shade, while the connective tissue remains white, and the tendons retain a silvery sheen" [6].

At about the same time, Kadyi showed a growing interest in microscopic studies in morphological anatomy. This coincided with a long-planned scientific trip that took him to research centers in Vienna, Prague, Berlin, Hamburg, Jena, and Leipzig. In the zoological department in Leipzig which was headed by Professor Rudolf Leuckart (1822–1898), in 1879, Kadyi published his work concerning the problem of preparatory techniques [7]. Developing an original mass consisting of soap dissolved in high-proof alcohol, he could embed preparations intended for microscopic examination, which was considered as cheap, simple, and giving good practical results [8].

Returning to Krakow, he mainly worked on techniques for visualising the vascular systems, which remained one of his main research fields. In 1881, Kadyi moved to Lviv: first as a professor of anatomy and pathological anatomy at the Veterinary School. Then, from 1894 until 1912, when he died, he was professor for descriptive and topographical anatomy at the Medical Faculty at Lviv University [9]. In Lviv, he extensively worked on preparations that involved creating collections for museum purposes. He used and constantly modified old and reliable techniques, especially those he learned when working with Teichmann. He was also experimenting with new chemical solutions.

Paraffin

In 1891 [10] and 1895 [11], Kadyi published two papers entitled "O stosowaniu parafiny do sporządzania trwałych preparatów anatomicznych" [Utilizing Paraffin for the Preparation of Permanent Anatomical Specimens]. They essentially summarize lectures delivered at congresses, and describe a paraffin-based method for conserving parenchymal organs and topographic sections in a dry state. Although paraffin had been already used for microscopic preparations in 1867 by Edwin Klebs (1834–1913) and in 1868 by Wilhelm His (1831–1904), Kadyi was now expanding its use to a "macroscopic" scale. He designed a three-step procedure: In a first step, the specimens were saturated with high-proof alcohol; in a second step the alcohol was replaced by gasoline and in the third, the specimens were perfused and then transferred to and bathed in liquid paraffin at 50 to 60 degrees Celsius. The method allows preserving a large range of specimens, including corrosion preparations fixed with celloidin. Paraffin prevents their structures from shrinking and prevents them from drying out or losing volume and shape. As it was described and noticed:

"The lungs, by filling the alveoli with paraffin, take on, and after drying, permanently retain the form and volume they have during inhalation. The injection of paraffin through the trachea is carried out in water heated slightly above the melting point of paraffin (40–60°C). Kadyi shows a preparation of human lungs made in this way, on which not only the shapes and arrangement of the lobes are perfectly visible, but also the free spaces occupied previously by the heart and other organs between the lungs" [10].

Brain and spinal cord

In 1900 a paper entitled "O barwieniu ośrodków nerwowych za pomocą zaprawy (barwienia) metalami ciężkimi" [The Application of Heavy Metal Mordants in Staining Central Nervous organs] was published by Kadyi [12, 13]. In general, it presents a technique for staining the brain's

gray matter with the help of an injectable substance which comprises copper, uranium, and lead acetates. As he informs: "Already more than three years ago (that is in 1897), using staining with borax carmine or lithic carmine and then washing for several days in strong alcohol (60%) acidified with hydrochloric acid, I succeeded in obtaining larger pieces of the brain stained through the whole structure, in which the difference between the grey and white matter was accentuated much to my satisfaction" [12].

In the following years Kadyi's principal approach changed. Now macroanatomy was the starting point for further work on methods that can be used in microanatomical research. Kadyi concluded that different protocols are required to stain various morphological components of nervous tissue with carmine. This was due to differences in the physical structure as well as the chemical level of reactions that are induced in the tissue under the influence of this dye. One of the most important discoveries made during this research was that the staining potential of the dye depends not so much on the properties of the dye but on the previous specimen processing steps. Consequently, he began designing different specimen processing protocols. Special attention was paid to introducing various salts [14]. It quickly became obvious that acetates are promising; with copper and uranium acetate producing the best results. Brain or spinal cord sections exposed to them easily stained in sodium carmine. During these experiments, Kadyi noted that the gray matter stains first, while stain uptake by the white matter requires significantly longer exposure to the dying solution. Therefore, he proposed interrupting the staining process at the "right time" to achieve exclusive staining of gray matter.

Furthermore, Kadyi identified external factors (temperature), structural factors (physical properties of the tissue), and factors related to the acetate concentration and the dye itself as influencing the outcome of the staining process. He extrapolated that a skilled anatomist may specifically stain tissues, by simply modifying these parameters [12]. As a consequence, he defined and published four different procedures for staining different components of the brain and spinal cord.

Sodium carmine staining produced quite good, but not ideal results. During the next years, Kadyi therefore continued evaluating other substances and, seven years later, he was able to present other tissue preparation and staining protocols for contrasting the brain and spinal cord tissues [14, 15]. Using the uranium acetate solution as the "bath" for future tissue preparation, Kadyi placed the tissues in a weak alkaline carmine dye, i.e., the potassium carmine solution, and then diluted it with water or a very weak solution of potassium hydroxide. That method produces sections of the brain or spinal cord with the grey matter strongly colored and the white matter completely colorless.

Vascular and lymphatic vessels

Kadyi and his mentors observed that substances, such as resins, waxes or Teichmann's putty, when injected into the blood or lymphatic system, do not enter the smallest vessels. Therefore, Kadyi experimented with barium putty tinted with carmine or Berlin blue and carmine oil [14]. Yet, even these solutions were not able to fill all capillaries. Hence, in 1907, Kadyi and his pupil Józef Markowski (1874–1947), continuing their research in Lviv, developed a new injection method for visualizing small vessels of the spinal cord [16]. The technique was dedicated to lymphatic vessels and blood capillaries and was based on gutta-percha mass, which already Teichmann had used extensively. Teichmann's method required dissolving gutta-percha in carbon disulfide (CS₂) before injecting it into the blood vessels. However, this method required keeping the specimen at

a stable temperature of 50 to 60 degrees Celsius to keep the injection mass liquid. Kadyi and Markowski managed to modify Teichman's method to allow the whole procedure to be carried out at room temperature. They followed Teichmann's protocol for preparing gutta-percha but removed the disulfide from the preparation using alcohol before heating it in a thermostat. This measure allowed smooth penetration of the injected solution into the capillary bed [16]. Kadyi also proposed staining the gutta-percha solution with a carmine precipitate and he continued experimenting with gutta-percha solutions by mixing them with different dyes and chemical compounds (e.g., chloroform) until he died in 1912.

Results

The literature search portrays Kadyi as a researcher with a passion for exploring and developing preparatory techniques — and as someone who was willing to share his knowledge with other anatomists. His passion was stirred while he was still a student in Vienna who had the opportunity to work closely with Karl Langer. Here, he gained his fundamental knowledge of various, then, cutting edge anatomical preparatory methods. This paid off when he discussed Zygmunt Laskowski's work a few years later and during his professional life. After being appointed assistant to Teichmann in Krakow, he was able to continue his Viennese education as Teichmann, before being called as professor to the Jagiellonian University, worked with Joseph Hyrtl, who in turn was Langer's teacher and predecessor. In the six years Kadyi was associated with Krakow University he had the opportunity to perfect his skills under Teichmann's supervision. He added injection and corrosive techniques to his portfolio and became interested in preparing specimens for microscopy, even inventing new embedding media.

After rotating to Lviv, Kadyi continued experimenting with preparation techniques and immediately started to pass his knowledge on to his disciple and successor, Józef Markowski. Together, they created and equipped anatomical collections; first at the Veterinary School and then at the Faculty of Medicine. During his training as student and assistant, Kadyi modified methods established by Teichmann. However, in the early 1890s he finally was ready to come out with genuine original proposals, which resulted in three, highly important milestones of anatomical preparation: Firstly, the introduction of paraffin wax as an embedding medium for microanatomy; secondly, in the development of a technique for staining brain and spinal cord specimens and thirdly, in the modification of the gutta-percha method to perfuse blood vessel and lymph capillaries. These achievements impressively demonstrate the mastery and deep anatomic knowledge Kadyi managed to gain, while working on building his position as an internationally acknowledged researcher. In fact, it was the long-term emanation of the anatomical tradition in Vienna (Hyrtl, Langer), which was further shaped and refined in Krakow (Teichmann), and which finally contributed to the discovery and establishment of novel anatomic methods in Lviv (Kadyi).

Conclusion

The art and science of anatomical preparation is an excellent example for demonstrating the importance of knowledge transfer between scientists and universities. The laws of physics and chemistry are identical in all institutions worldwide. Basic principles of conservation and perfusion are common knowledge among anatomists in all countries. But it needs individuals like Kadyi, Teichmann and Langer, who are willing to mentor young scientists, delighted to share their results with

colleagues and eager to transfer knowledge across borders and cultures. As nicely shown in this study, such conditions generate scientific progress, trigger new concepts and techniques, and solve long standing problems. Henry Kadyi's scientific life is a splendid embodiment of these principles.

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