Department of Pathology
University of Veterinary Medicine

Veterinary digital pathology

Literature review

Maria Slapgaard
N2OTFJ

Supervisor:
Dr. Horváth Dávid Géza

Hungary, Budapest
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1. ABSTRACT

Digital pathology means the display and interpretation of digitalized histopathological sections on a computer monitor. The technology has many advantages in diagnostics, education, and also in research activities. Whole slide scanners performing the digitization are capable of scanning routinely (hematoxylin and eosin) stained sections, as well as slides labeled with special procedures, immunohistochemistry (IHC) or in situ hybridization (ISH) in high resolution. The resulting whole slide images (WSIs) can be examined with digital image analysis softwares, capable of supracellular, cellular and subcellular structure detection and differentiation in an objective and reproducible manner. The diagnostic workflow could be less time consuming, and the consultation possibilities could also expand. In research settings it has an increasingly important aspect, as tissue evaluation of morphological and molecular procedures is subjective and often difficult to repeat. In addition to objectivity and reproducibility, numerical data obtained with digital image analysis is easier to handle. The educational activity is more interactive, the instructor can clearly mark certain lesions, even several special staining procedures on a single monitor. There is no need to replace worn out or damaged slides and the system is also suitable for examinations. Despite the numerous benefits, the methodology has several limitations. The different scanners create their own file format that could interfere with interinstitutional cooperation. The introduction of the routine use of digital pathology can entail significant costs and depending on the performance of the diagnostic laboratory, terabytes or petabytes of data creates a significant burden on storage. Within artificial intelligence, the most effective deep learning systems are criticized, as it is not yet possible to explain how they get to their decisions. These problems remain to and probably will be solved in the near future.

2. INTRODUCTION

Digital pathology or virtual microscopy is a relatively new branch of pathology including the gathering, organization, interpretation and sharing of pathological samples on glass slides converted into high-quality digital images, the whole slide images (WSIs). These images can easily be stored, visualized, and analyzed on electronic devices like a computer, and also shared across countries, clinics, and hospitals. It is an important tool of artificial intelligence (AI) which has many application possibilities in research, education and diagnostics. Digital pathology has proven to be beneficial for teaching in universities and creating more flexibility for pathologists working from home [1, 2]. The digitization of glass slides and the use of whole-slide imaging are fast-evolving technologies that are transforming veterinary pathology. It has great potential to increase workflow efficiencies for veterinary pathologists, as well as in the diagnosis and patient care [2].

Histomorphology and its evaluation, have served as a major tool for scientists and pathologists for over 125 years. Expansions and new techniques were developed every decade, and there has been a major focus on the increasing quality and amount of information from histological slides, and their use for diagnostic purposes [3].

The traditional methods of digital pathology involved the transmission of static images taken with microscope-mounted cameras between remote sites. However, over the years, it has evolved into robotic telepathology, where a pathologist can control a robotic stage from a distance and view a specimen in real-time. Currently, there is a diverse range of imaging devices for digital pathology that can be purchased. These devices include microscope smartphone mounts, affordable software solutions that enable the manual utilization of a light microscope as a substitute scanner, as well as independent high-capacity whole-slide scanners that have obtained acceptance from the FDA (class II medical devices) [4]. Nowadays, digital pathology relies mostly on whole slide imaging (WSI) where scanners are used to create high-quality digital images of entire histological sections. After digitization, the images can be evaluated through a computer monitor that imitates a light microscope, allowing pathologists to view the images in a more convenient and efficient manner [3].

In the past digital pathology was time-consuming and had great expenses, especially for storing, and the advantages were not sufficient to validate the costs. With the major digital evolution that has been occurring over the past 10-20 years, these statements are no longer true. Rapid and more efficient scanning is possible, and the costs of production and storage have been reduced making digital pathology a more beneficial tool in diagnostics, education, and research [5].

One significant step towards firmly establishing digital pathology in both veterinary and human medicine was its acceptance for primary diagnosis. In 2014, large veterinary diagnostic firms like Antech and IDEXX, transitioned to a 100% digital workflow for their anatomic pathology services. In contrast, the Food and Drug Association approved the first digital pathology system in 2017 for use in primary diagnostic readings of human specimens, but wide adoption was still lacking [4].

3. IMPORTANCE IN VETERINARY MEDICINE

Digital pathology has become increasingly important in veterinary pathology. It was used in veterinary routine diagnostics prior to its similar human implementation, mainly because of the stricter law regulations concerning human tissue samples [4]. Digital pathology has the potential to change veterinary pathology by reducing the level of labor intensity and increasing access to remote working pathologists. It also offers a better balance between daily life and the work of pathologists due to the flexible working hours [2].

The implementation and use of digital pathology differ among several academic institutions and clinics. Some veterinarians and pathologists incorporated these recourses into their daily practice and curriculum over a decade ago, while others may still not have access to these tools, because of their price, and the lack of storage capacity and trained personnel. Whole slide images are often used as equipment to enhance education and for the purpose of interinstitutional collaborations, and many facilities now teach histology and histopathology with digital slides [4, 6].

The veterinary field that first started properly using digital pathology and incorporated it into daily work, was toxicologic pathology. It was found to be very beneficial because of the possibility of instantly sharing the samples online with experts in the field, without the need for transportation that could lead to significant information loss and material damage. Also, image analysis can potentially capture details the human eye is not able to see even with a microscope, such as mild biomarker staining or disease progression, making digital pathology and whole slide imaging highly important for more in-depth investigations [4].

4. HARDWARE

1. Scanning

Numerous types of slide scanners offered by various suppliers and vendors operate with some differences but with the same main goal, to produce the highest quality digital images [7]. Most digital imaging providers use a tiling scheme, while others may use somewhat different technology to create these digital images. These systems will either create tiles from individual high-resolution images or make linear scans of tissue areas using a linear scanning arrangement. To produce a single digital image of the histologic section, both approaches require the tiles and line scans to be stitched together and smoothed [3]. The lines or tiles need to be saved and stored at different resolutions in order to create numerous images of the same slide, which, when combined, form a picture pyramid (Figure 1). By doing this, you can examine the whole-slide image on a display without experiencing image latency while zooming in and out. This is not the same as "z-stacking", which creates multiple scans along a slide's z-axis in order to enable a digital fine focus [4].

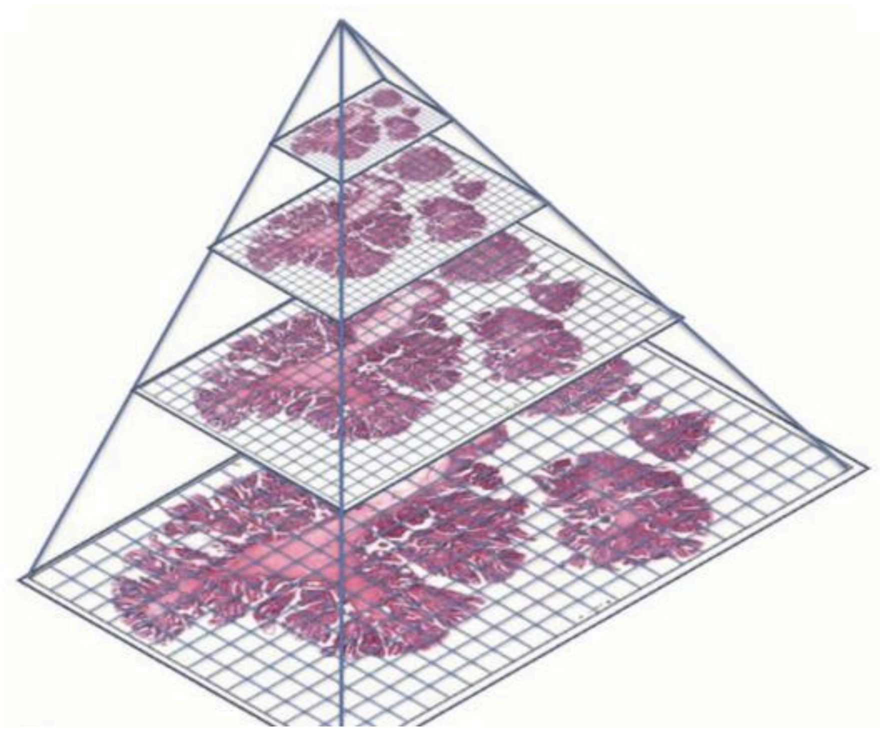

Figure 1: A pyramid-shaped digital slide storage system with several magnifications. In this instance, the layers of the pyramid store four distinct digital slide magnifications [4].

When facilities and laboratories purchase scanners, their decisions are based on their intended use, storage capacity, and budget. Although the selections of scanners vary among institutions, it will also change in time parallelly to the increasing user needs. There are four types of whole slide scanners: integrated, high throughput, low throughput, and real-time hybrid/robotic scanners, whereas most of the institutions with an average to high level of workload will need high-throughput scanners. Despite some variations in the technical setup, all scanners have four main constituents that are roughly equivalent to those of conventional light microscopes. These are the light source, robotics for moving the slide holder (slide stage), one or more objectives, and an associated camera to record the images. The process of digitizing histologic sections into digital pictures is the major component and difference of digital pathology in comparison to the conventional approach (Figure 2) [6][7].

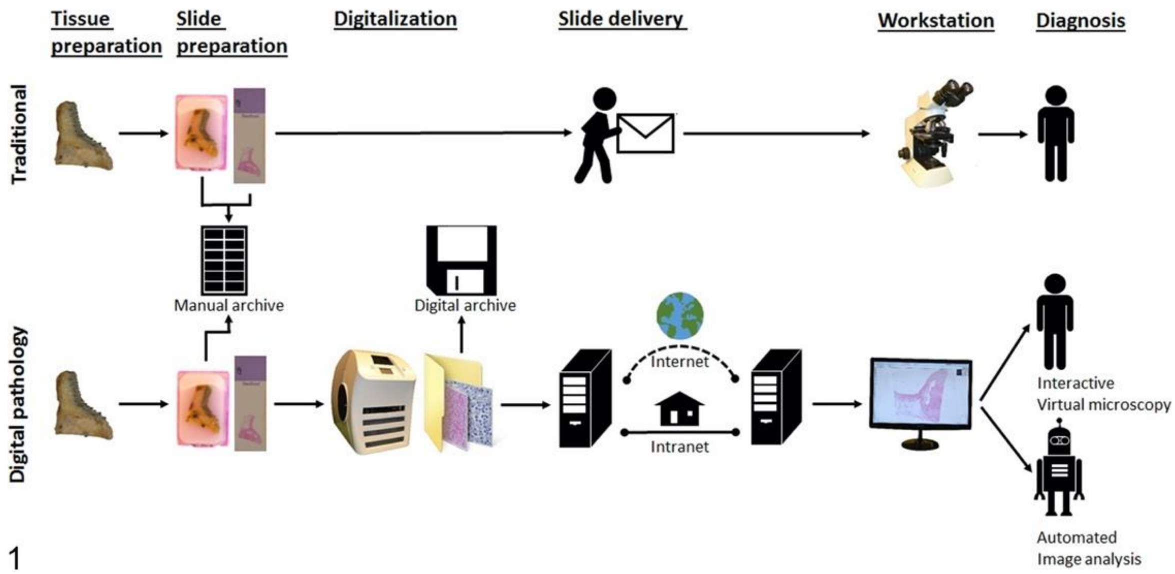

Figure 2: A comparison between traditional workflow and digital pathology. The primary process that separates digital pathology from classical pathology is the scanning of histologic sections using a slide scanner to create whole-slide images. These images can then be analyzed by automated image analysis or managed and viewed by a pathologist at a computerized workstation [7].

A maximum capacity of slides exists for each scanner, it ranges between 1 and 400. It is an essential component that needs to match the institution's digital burden. The large slide holders are necessary for large diagnostic laboratories with a huge workload in order to prevent time-consuming manual assistance of the holder [7].

The vast majority of slide scanners include a bright field light source, and some of them even include an inbuilt source of light for the activation of fluorochromes, which enables them to digitize slides that contain fluorescent stains. It creates the possibility of examining the fluorescence signals at any time without fading and quality decrease [7].

With whole-slide imaging, the concept of magnification as it is generally understood in light microscopy is not applicable because the images are viewed on screens of varying sizes, which have the ability to magnify or reduce the initial magnification. In whole-slide imaging, resolution is expressed in micrometers per pixel and is determined by the image sensor and the objective that is used to scan the slide [3].

The different scanners use 20× to 100× objective lenses, allowing for maximum screen magnifications of 200× to 1000×, similarly to a light microscope. Scanners contain one or more objectives, with the ability to add two additional lenses to increase the magnification. Although the vast majority of scanners have objectives with a magnification of 20× and/or 40×, only a few types, such as the Leica Aperio CS0, are able to scan at 1000× magnification while maintaining oil immersion [7].

The majority of digital scanners can only produce 2D planar images, in comparison to conventional pathology in which the pathologists often evaluate several focus planes. 3D whole slide scanners should be available on the market soon, which will increase the pressure to create more sophisticated and effective methods for examining 3D stacks of digital slides [1, 8].

Some vendors also produce scanners using oil immersion lenses or that are capable of fluorescent scanning and depending on the goal and cultured tissue/cells, florescent scanners might need a much higher resolution (40×, 60×, or 100× magnification), which again increases scanning time. A 100× objective usually requires z-stacking, and the majority of fluorescent scanners scan separately the channels of the fluorophores [3]. The specific scanners capable of “z-stacking” and acquiring several planes can be useful for more in-depth diagnosis, including confirming the presence of mitotic figures. However, due to the corresponding increase in acquisition times and data, z-stacking has primarily been used in research, and not primary diagnostics [1, 8].

The quality of the tissue and slide preparation, including uneven tissue slices, tissue artifacts, and the staining or immunolabeling quality, are some of the factors that can limit the capabilities of digital slide scanners [3]. Other tissue artifacts are air bubbles, tissue folding, tissue sectioned at an excessive thickness, tearing, compression, over and under-staining, and stain intensity variability can all have an impact on the final digital image and result in poor identification [4]. Because of this, excellent specimen preparation is the first step towards making good-quality digital images, moreover, quality control systems must be adapted so that scanned images are of the best possible standard. Furthermore, it is important to note that the scanner itself also can have a detrimental impact on quality. This can occur through out-of-focus scanning or uneven illumination of the slide. There are currently tools that have the capability to find these histological artifacts and eliminate them from our analysis, but quality control prior to scanning should be carefully considered to obtain consistent, high-quality scans [3, 4]. Before scanning, slides should be cleared of dust, cleaned, and checked for deviations to ensure the sharpness and focus of the images [3].

2. Viewing digital slides

To observe digital pathology specimens, scientists utilize computer interfaces that mimic the features of a light microscope (Figure 4). Slide viewers work by accessing and showing the tissue area. A slide viewer has basic features like controls that let the user change the magnifications, rotate and zoom in on a tissue section to look more closely and take photomicrographs of digital images that have not been changed or annotated. These can all be used in reports and publications [3].

Figure 4: Screenshot of Aperio's ImageScope digital slide viewer. The digital slide viewer mimics a microscope with some extra features, including (A) a filmstrip showing slides available to be seen, (B) a zoom slider in order to alter magnification, (C) slide label, (D) a small portion of the slide displaying the current place within the slide. (E) a magnifier to view at smaller portions of the slide at a higher magnification [3].

5. ADVANTAGES

1. Increased productivity

One noteworthy benefit associated with the adoption of digital pathology is the improved ability of the staff to gain a comprehensive understanding of their own work. This is possible because of the organization and quick availability of digital pictures on a computer platform. Comparing digital and traditional slides, the digital slides are all located on a digital device and accessible within 10 seconds. Traditional glass slides may need significant time to find in archives and might be placed in different rooms and used by other colleges. Additionally, it could take a remarkable time to consult with a colleague. This time can be spared, and possible interruptions minimized. Colleagues can instead view the specimen when it suits them without being disturbed or wasting valuable time [2, 9, 10].

Pathology is experiencing a growing personnel issue in the context of an increasing diagnostic burden. It is estimated that between 2010 and 2030, there will be 3.7 practicing pathologists for every 100,000 persons in the United States, down from 5.7 in 2010. The Bureau of Health Professions projects a decrease of 30% in pathologists in the United States. Over the course of the next five to 10 years, the UK will experience a shortage of consultant pathologists in all subspecialties. 615 pathologists, or 32% of the field, are over 55 years old and are anticipated to retire over the next five years according to data from the Royal College of Pathologists. Therefore, it is promising to implement DP in order to speed up the process of diagnosis for less negative impact of the loss in pathologists, and better patient care [11, 12].

The utilization of AI technology has the potential to support the comprehensive reporting system, expedite the process of reporting, and provide a more objective assessment of morpho-biological characteristics. The utilization of AI technology in the reporting of specific traits or lesions might provide valuable assistance to pathologists in effectively prioritizing complex cases and effectively managing the growing demands of their job. The purpose of implementing digital pathology is not to replace human resources, such as pathologists and laboratory workers, but rather to improve existing capabilities. The primary objective of this system is to provide aid, support, and enhanced efficacy in the field of diagnostic procedures and overall performance. In addition, the integration facilitates enhanced resource allocation, better cost-effectiveness, and improved consistency in pathology reviews (figure 5) [12].

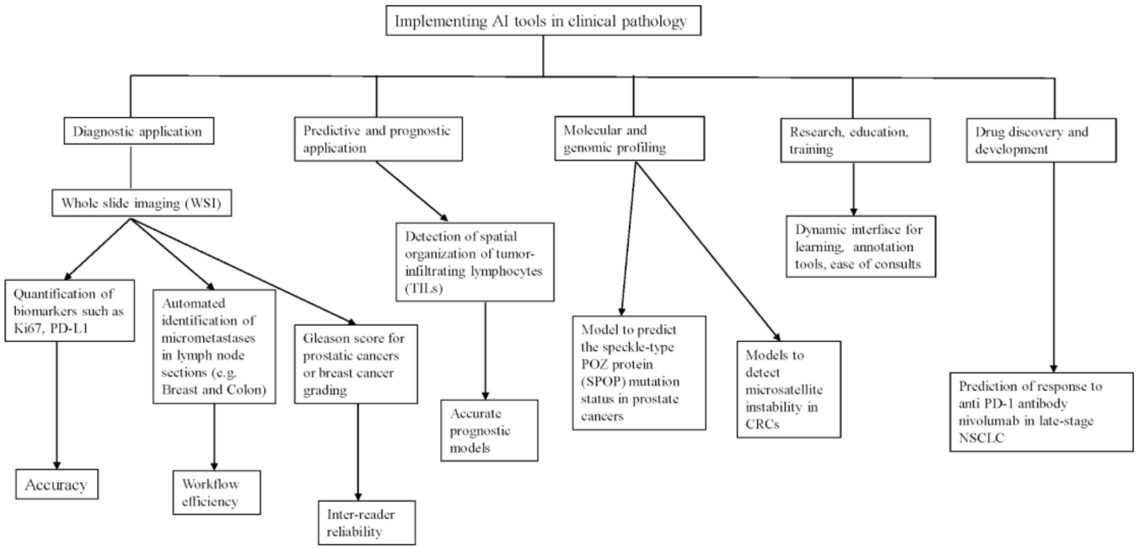

Figure 5: Milestones in DP with representation of three "revolutions" in the field [12].

2. Collaboration

A major benefit of utilizing digital pathology is its capacity to facilitate communication and promote cooperation among individuals and institutions spanning multiple nations and continents. Samples can be shared immediately and discussed for a second opinion in a very short period without the risk of important material being lost [1, 4, 9, 10, 13]. Content-based image retrieval (CBIR), is an AI tool for pathologists, that allows them to effectively search for images similar to the one under examination within a comprehensive histopathology database. This holds significant importance in aiding pathologists in the diagnosis of complex and infrequent situations [12].

3. Education

Another advantage of whole slide imaging is the better education possibilities. It is advantageous nowadays because it is a more appealing modern way of educating the younger generations, increasing their motivation and individual studying outside of the universities [1]. Additionally, all students review the same presentations and specimens and have constant access to the material online from any location [4].

4. Cost-effectiveness

Many factors should be taken into consideration before implementing digital pathology in practical settings. The slide scanners have an upfront cost of a minimum of 100,000 dollars, with additional yearly maintenance and service costs, and maybe even a full-time image analyst. A close relationship and cooperation between the IT company and the staff of the clinic is necessary to establish and maintain a high-quality understanding and usage of the tools, and the complexity of the IT infrastructure completely depends on the goals, slide volume, and user volume of the system. It is crucial to consider these matters prior to implementing digital pathology in order to reach the set goals and optimize performance [3].

No real-life cost-efficiency analysis for a complete digital pathology implantation with more than five years of follow-up has been reported, but there are cases in which companies that have implemented digital pathology into their unit have gained economically over the years. This is due to the decreasing expenditures for labor workers, transport fees, and expensive, fragile glass slides that can break and decrease in quality over the years [1].

In 2019, an article described the implementation of DP in an American pathology department at Memorial Sloan Kettering Cancer Center. They began to switch from research to diagnostic whole slide imaging in 2015 on a small number of cases. The academic center produced about 125,000 slides every month in 2017. WSIs increased to 23,000 digital slides per month by mid-2017. The application reduced glass slide demands by 93%. Digital pathology also reduced ancillary immunohistochemistry from 52% in 2014 to 21% in 2017 after it was implemented. In addition, there was a yearly immunohistochemistry savings of 114 000 dollars, and a total cost saving estimate of 267 000 dollars per year, due to the digitization [6] [1].

5. Accuracy

Digital images allow easier and more extensive annotation than traditional histopathological slides, and also for more accurate measurements. This increases the traceability of deviations and transparency of the diagnostic procedures. According to a previous research, diagnostic methods using digital scans of cell and tissue samples are just as precise as using a microscope to examine traditional slides. It offers the most compelling proof to date that digital pathology can eventually replace traditional microscopy. It was also established that the diagnosis made by digital pathology and microscopy nearly always agreed (greater than 98% agreement) [13].

6. Image analysis

There are several approaches for digital pathology image analysis, and depending on the process, different factors must be considered during the creation, performance evaluation, and application. A single or a combined method could be used, the institution needs to determine the most suitable one for their tasks and its planned purpose before beginning to design an automated image analysis system. Methods can be chosen based on the availability of information, model flexibility, accessible computing power, acceptable processing time, capacity to process results, and algorithmic resilience, in addition to attaining the best test performance and result possible [14].

The application of digital image analysis primarily serves to extract quantitative information from tissues, which cannot be evaluated to the same extent or in the same manner through manual assessment. The evaluation categories that are most frequently utilized include area-based, object-based, and cell-based (figure 3). Regardless of the particular technique used for image analysis, the pathologist plays a crucial role in the workflow in order to guarantee the production and interpretation of high-quality data [4].

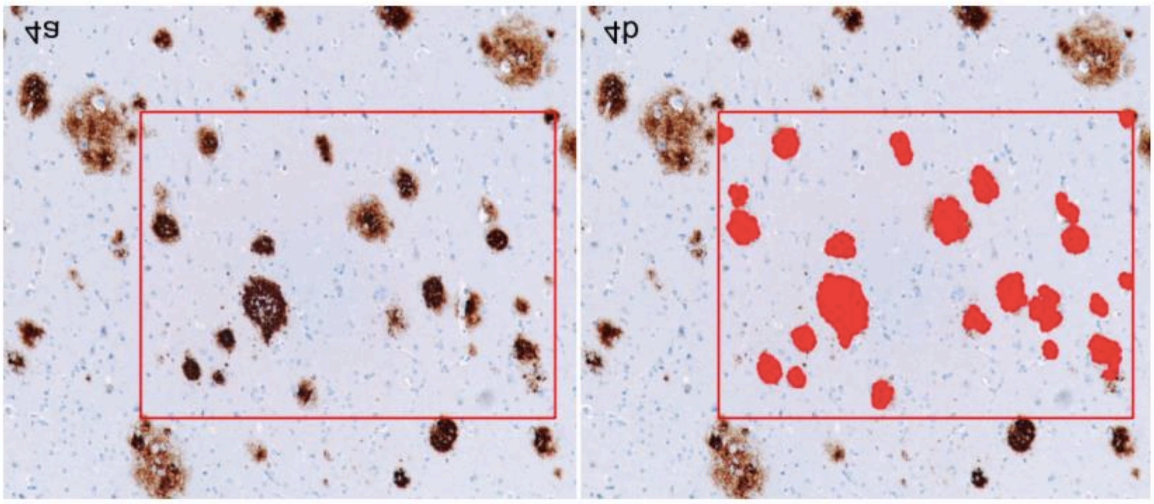

Figure 3: Senile plaque found in the human brain. An illustration of an object-based measurement that does not rely on the detection of individual cells. (A) Brown plaques have successfully been recognized using immunohistochemical labeling for beta-amyloid. The location of the area of interest is shown by the red square. (B) Algorithm markup (in red) of areas containing beta-amyloid plaques [4].

6. DISADVANTAGES/CHALLENGES

1. Expensive equipment

Digital pathology clearly has many advantages making it highly valuable, but it is also important to consider the disadvantages before installing digital pathology equipment into practice. The justification for DP implementation usually focuses on efficiency improvements. The equipment could be extremely expensive, and only a part-time digital pathology implementation would not be economically profitable. The primary digital diagnosis workflow in pathology must eventually be completely digital in order to become cost-effective, in the meantime, glass slides must still be accessible upon request [3, 4].

2. Unstable technology

One of the primary issues encountered with digital pathology is unstable technology. The implementation of digital pathology requires the availability of high-speed internet connectivity and the use of high-magnification scanners, both of which are susceptible to network, software, and hardware malfunctions. Additionally, there is a lack of a universal format for virtual slide data, which can lead to challenges in storage and interinstitutional cooperation [15].

Technically, a major challenge in DP is the color variations of the tissue caused by differences in slide preparation, staining, and even entire slide scanners. It is evident that decision support systems designed to operate on digital pathology pictures must be able to manage these variations [8].

The accurate identification of the specimen during the scanning procedure, can be accomplished either by manual means or by utilizing a barcoding system. It is important to note that WSI systems exhibit optimal performance when operating under optimal optical conditions. In order to secure this outcome, it is imperative to establish quality standards throughout the laboratory sectioning and staining procedures. The objective of these standards is to optimize the available scanning area and enable the equipment to precisely focus on the specific tissue slice [9].

The 20× is the most commonly used objective for histopathologic evaluations and is widely accepted as the standard magnification for most digital pictures. Nevertheless, this could be an obstacle to duplicating the procedure for establishing primary diagnoses, as traditional light microscopes consistently include 40× and frequently 100× objectives. Consequently, countless pathologists have an uncomfortable experience when lacking convenient access to these objectives, slowing down the process. Including the 40× objective will therefore increase both the scanning time and file size by four times, leading to an increase in the slide processing time and storage cost [3].

3. Competence of Staff

The upfront cost is a limiting factor in the deployment of a DP system. This pertains to the expenses associated with the acquisition of scanners and the establishment of information technology infrastructure, and additionally the costs related to acquiring more laboratory space for accommodating the equipment. Pathologists must undergo a significant training process in order to shift their industry to digital. This involves acquiring competence in the latest digital pathology workflow, acquainting oneself with the associated instruments, and developing the aptitude to diagnose from WSIs [4, 8]. Pathologists must acquire competence in the manipulation of image files. This entails the necessary skills to obtain patient paperwork followed by the initiation of the WSI interface, whereby users may effectively navigate, manipulate, and magnify the digital slides. Acquiring expertise in the slide scanning procedure, including the ability to recognize scanning artifacts and address focus plane issues, requires investment of time and experience (figure 6) [4, 9].

4. Sharing

For diagnostic purposes, digital copies must, be at least the same quality as the original slides. The scanning procedure spans a duration ranging from one to twenty minutes and provides files ranging in size from several hundred megabytes to several gigabytes, commonly from 200 MB to 1 GB. In order to facilitate the sharing process, an adequate internet connection with high-speed and high-bandwidth abilities is necessary [5, 9].

5. Archiving

A substantial amount of memory is needed for the storage of these whole slide images. Additionally, image compression is necessary for all platforms. A universally accepted file format yet does not exist, and the absence of this standardization in digital imaging systems has the potential to cause challenges during transitions between various platforms [9, 16].

The process of storing traditional glass slides demands physical space and creates the possibility for misplacement, breakage, and gradual deterioration of the slides. A digital library provides a safe and easily accessible way for long-term storage, although it needs suitable IT assistance, a flexible storage capacity, and periodic backup of the archive. In case of a disaster, the backup facility should be placed far from the original place so that no vital information will be lost. Nevertheless, archiving these digital images can incur significant costs. Based on the assumption that a standard digital image has a file size of 500 MB, it may be calculated that a terabyte-sized file has the capacity to contain around 2000 digital images. The yearly expense associated with this particular storage capacity, inclusive of necessary backup and security measures, varies between 3000 and 10,000 dollars [3].

Figure 6: Whole slide scanning and the evaluation of these virtual slides [17].

7. APPLICATIONS

Prior to digitization, it is still necessary to perform fixation, sectioning, mounting, and staining procedures on the tissue samples. Whereas it costs a significant amount to deploy digital pathology in the laboratory due to the need for gear, software, IT infrastructure and protocols, training, backup and storage plans and laboratory operations, digital pathology presents new possibilities for enhancing analytical and diagnostic capacities, and goes far beyond the mere production, conversion and distribution of WSIs [9].

1. Diagnostics

According to the US Food and Drug Administration, digital slide scanners are classified as type III medical equipment, necessitating validation before being approved for primary diagnosis.
Within the pathology community, there has been a continuous discourse surrounding the necessary research design to support the use of digital pathology. Notably, certain experts have suggested that, instead of a broader validation, it may be more appropriate to validate digital pathology for specific indications [3].

The field of diagnostic pathology has undergone a significant transformation, characterized by the adoption of specific new technologies. Examples include digital imaging, complicated artificial intelligence (AI) algorithms, and computer-aided diagnostic tools. These tools are employed to aid, enhance, and empower the field of computational histopathology and AI-enabled diagnostics. This development is a facilitating progress in the field of precision medicine for cancer (Figure 7) [12].

Over the last decade, the diagnosis of cancerous cells has developed rapidly. The more traditional methods of cancer diagnosis were narrowed down to fewer variables such as grade, stage, and a few markers, like receptors for progesterone, estrogen and HER2 for mammary cancer, and prostate-specific antigen for prostate cancer. The aim of the pathologist was to provide a diagnosis assisting the clinician to give the best possible therapy. The recent improvement in image analysis algorithms and tools of digital pathology has made it possible to analyze a much larger set of variables over a brief period [8].

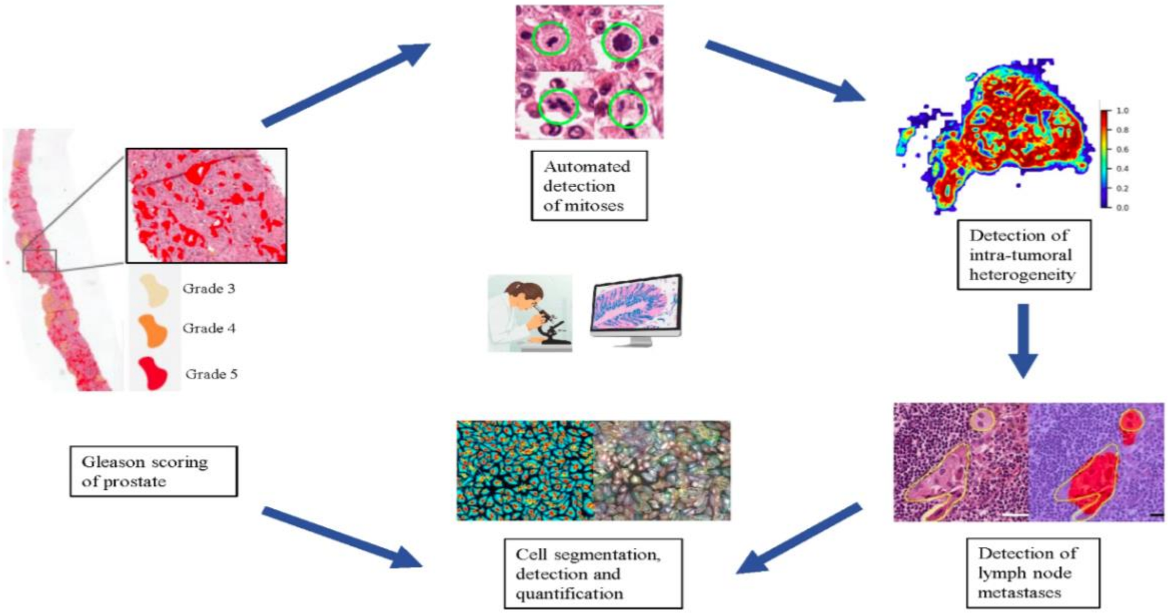

Figure 7: Artificial intelligence (AI) and machine learning approaches to analyze images from tumors [12].

In the case of prostate cancer, an approximation of one million biopsy samples are taken in the US every year where approximately 20% of them have cancerous lesions and the remaining 80% are benign. This indicates that pathologists are wasting valuable time examining non-malignant tissues that can typically be differentiated from cancerous cells. This time and resources could be used for specific cases where the significant lesions are more difficult to identify. Therefore, many scientists have started to develop computer-aided-diagnosis methods to recognize the characteristics of diseases such as neuroblastomas, meningiomas, and breast cancer on digitized tissue slides [8].

In the US more than 60 digital applications have been approved for radiology, cardiology, and internal medicine by the FDA but non for pathology. The Galen Prostate software is an AI-based pathology system applied for routine pathology practice and is capable of prostate cancer cells recognition in core needle biopsy WSIs. In external validation, the specificity was 97.3% and the sensitivity was 98.5%. It completed a second-read evaluation on more than 900 cases in an Israeli pathology department and detected one misdiagnosed case [1]. The Swedish hospitals Kalmar and Linkĥping were among the first to implement digital pathology, completely integrated digital slides and slide scanning into laboratory workflow [12].

AI technologies can also aid in the standardization of scoring systems for a variety of tumors. The Gleason score for prostatic malignancies and the breast cancer grading systems could be better compared with digital pathology methods. It is imperative to consider the diverse range of normal tissue while developing an algorithm, in order to prevent the incorrect detection of microlesions (like benign mimics of cancer) or focal cancerous lesions [12].

An increasing number of artificial intelligence (AI) methods are being used to facilitate the identification of information that is challenging for pathologists to understand. Examples among others are objective evaluation of biomarkers like PD-L1 and Ki67, quantification of cells and recognition of distribution patterns. Artificial intelligence (AI) can be employed for the identification of isolated tumor cells in lymph nodes that are indicative of metastatic neoplasia, hence enhancing the sensitivity of detection in a manner that optimizes time utilization [12].

2. Research

Recent improvements in automated image analysis could enhance histopathologic data reproducibility. It is believed to have the potential to alleviate the burden associated with boring and repeated counting tasks, enhance repeatability by eliminating variability among observers, and enhance the level of objectivity [7].

Various digital pathology procedures were used in studies evaluating animal tissues. In a previous study, the utilization of whole slide imaging was employed to assess pig liver sections following bipolar radiofrequency ablation done in vivo. The goal of this strategy was to quantitatively evaluate distinct regions of tissue destruction and cellular death. By utilizing a custom algorithm, they successfully conducted an objective comparison of the degree of cellular damage across several tissue slices from diverse animal specimens (Figure 8). A different, less complex method of pixel counting was employed to measure the extent of myocardial fibrosis in a murine model of ventricular hypertrophy induced by pressure overload (Figure 9) [7].

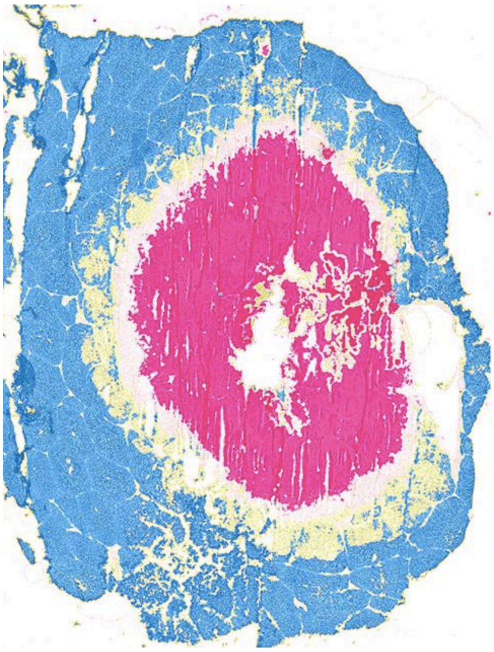

Figure 8: NADH-diaphorase–stained sample of pig liver. An Aperio CS2 scanner was used to digitize the tissue sections, and the Genie Software from Leica Biosystems Imaging Inc. was used to create a macro for viewing the different stages of destruction. Bright pink being full cell destruction, cream is fragmentary tissue destruction, edematous areas are indicated with yellow and normal hepatic tissue is bright blue [7].

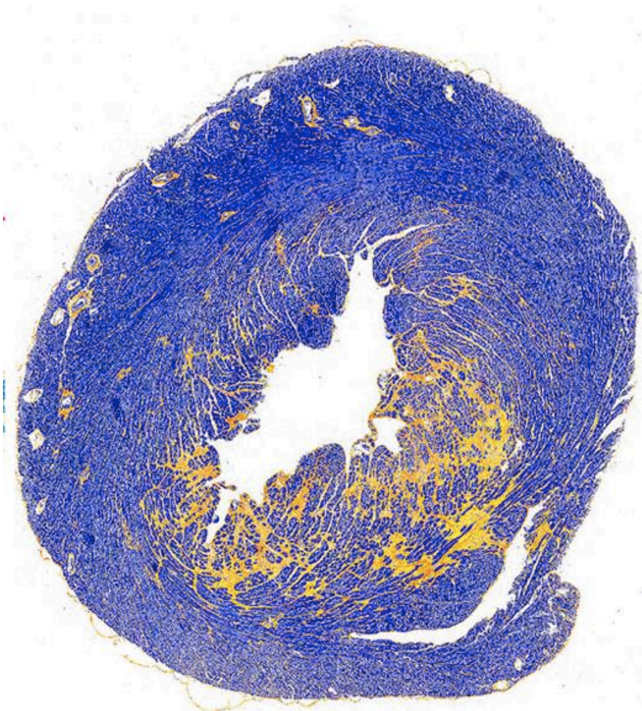

Figure 9: Picrosirius red–stained heart muscle of a mouse. An Aperio CS2 scanner was used to digitize the tissue sections, and the Genie Software from Leica Biosystems Imaging Inc. was used to create a macro to view the fibrotic areas (orange) in the myocardium (blue)[7].

For research and clinical studies, DP provides a variety of benefits related to versatility and adaptability. During the COVID-19 epidemic, limits and restrictions were placed on the research environment, and since then, the criteria for the standard of care has evolved. This led to a decreased in the number of samples and sample types available for research, which has an impact on all research fields that require patients or samples, including cancer studies. Additionally, travel restrictions may hinder transplantation research [18].

3. Education

Originally, the teaching of histopathology and cytopathology has included the use of light microscopy (LM) and physical glass slides for each individual student, as well as the incorporation of images, multiheaded microscopes, or video microscopy. The availability of light microscopes and glass slides are limited to specific times and locations. Additionally, static images lack the ability to enlarge or reduce in size. Multiheaded microscopes are limited to the inspection of a few slides. Video microscopy frequently exhibits poor resolution and presents challenges in terms of handling [7]. Nowadays, digital pathology (or virtual microscopy) has emerged as an extremely successful alternative approach for teaching and evaluating histopathology. Its integration into the educational programs of veterinary faculties is becoming more prevalent, particularly in the area of histology, and parasitology. Virtual microscopy is a useful tool for performing practical exams, enabling self-directed learning, and offering good-quality lectures. However, many veterinary students value being taught with traditional light microscope simultaneously. Some pathologists have concerns that an excessive focus on virtual microscopy in the veterinary education curriculum might lead to a deficiency in the capacity and knowledge about how to work and manipulate glass slides and traditional light microscopes. It is worth noting that the use of conventional light microscopes remains the predominant method in routine veterinary practice, particularly for cytopathology analysis [7].

8. FUTURE DIRECTIONS AND DEVELOPMENTS

Digital pathology is used in all fields of pathology and offers a unique chance for pathologists to improve their diagnostic, research, and teaching skills, despite current minor weaknesses [7].

Major advantages of digital pathology and whole slide imaging include easy handling, improved ergonomics, quantitative measures, and remote access to digitalized slides. Even though automated image analysis in veterinary pathology is still in its infancy, over the next decade pathologists probably will be able to perform reproducible and reliable qualitative and quantitative diagnoses with it. Pathologists and researchers must investigate how they can implement it into the daily workflow rather than leaving it to IT specialists. To stay up with the impending digitization of pathology, current and future pathologists should learn information technology and image processing in order to effectively adapt to the changing and evolving process of digitization within their field [7].

The discipline of digital pathology is rapidly changing, with the emergence of new diagnostic and prognostic tools in the market and under research, demonstrating promise for further improvements in patient care. The subdiscipline is developing quickly, and a pathologist's workload can become less demanding and more efficient by using equipment for automated mitotic counting, tumor grading, and microorganism detection. These tools save the valuable time of pathologist, so that they can prioritize it and can focus on the more challenging parts of diagnosis. Future image analysis of digital slides is most likely going to become a part of the normal practice, which will allow for even higher simplicity of the diagnosis process [19].

10. DISCUSSION/CONCLUSION

Digital pathology is a rapidly evolving field that has changed pathology practice. It involves collecting, organizing, disseminating, and interpreting pathology data in a digital context. Through the utilization of artificial intelligence (AI) and deep learning, the digitization of glass slides has opened new diagnostic opportunities and allowed pathologists to diagnose effectively and precisely a large number of cases.

Digital pathology has the potential to revolutionize the practice of veterinary pathology. The ability to scan and digitize slides for histology and cytology can make the process less labor-dependent and increase access to remote working pathologists. Moreover, with the rapid referral of cases, not only is there the possibility for quicker diagnosis, but it also gives the business access to a greater staffing pool, including better access to expert advice and second opinions. It also provides our pathologists a better work-life balance.

In summary, digital pathology is a promising area that has the potential to transform the way we diagnose and treat diseases. With the accessibility of new technologies, the future of digital pathology looks promising. In the veterinary field, digital pathology has the capability to revolutionize the daily work of veterinary pathologists, making the process less labor-intensive and increasing access to working off-site. However, it is crucial to note that the global shift in the direction of digital pathology and AI should be done with caution and consideration, as it is still a relatively new technological field and necessitates additional research and growth to ensure its safety and efficacy.

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I hereby confirm that I am familiar with the content of the thesis entitled

…………VETERINARY DIGITAL PATHOLOGY…………………………
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……………– LITERATURE REVIEW………………………………...…..

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Date: Budapest, …16….day……11…..month …2023…….year

……HORVÁTH DÁVID GÉZA……………………

……. Supervisor name and signature

…………………………………………………

……PATHOLOGY……………………………

……. Department

Thesis progress report for veterinary students

Name of student: MARIA SLAPGAARD

Neptun code of the student: N2OTFJ

Name and title of the supervisor: HORVÁTH DÁVID GÉZA, ASSISTANT PROFESSOR

Department: PATHOLOGY

Thesis title: VETERINARY DIGITAL PATHOLOGY – LITERATURE REVIEW

Consultation – 1st semester

Timing				Topic / Remarks of the supervisor	Signature of the supervisor
	year	month	day		
1.	2022	06.	26	—	[signature]
2.	2023	02.	22	—	[signature]
3.	2023	03.	18	—	[signature]
4.	2023	05.	19	—	[signature]
5.	2023	05.	26.	—	[signature]

Grade achieved at the end of the first semester: 5

Consultation – 2nd semester

Timing				Topic / Remarks of the supervisor	Signature of the supervisor
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1.	2023	09	1	—	[signature]
2.	2023	09	7	—	[signature]
3.	2023	09	14	—	[signature]
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5.	2023	09	28	—	[signature]

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The thesis meets the requirements of the Study and Examination Rules of the University and the Guide to Thesis Writing.

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