Original Article

Evaluation and comparison between smartphone and photomicrography based whole slide imaging

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ABSTRACT

Context: In recent years, a new concept has emerged at the forefront of slide-based diagnosis and telepathology. This is the concept of whole slide imaging. This has very recently also been tried with smartphones using complex software for photo analysis and stitching. This study is aimed to evaluate and compare the use of Jenoptik photomicrograph camera and smartphone camera-based whole slide imaging (WSI) for various microscopic preparations and slides. The study also uses a commonly available IOS smartphone device, a commonly available phone to microscope attachment, and the most commonly used Adobe Photoshop software for all stitching purposes. **Aim:** To evaluate and compare smartphone and photomicrography-based whole slide imaging. **Subjects and Methods:** The study was conducted at the Department of Oral Pathology and Microbiology in Bapuji Dental College and Hospital, Davangere. The samples included in this study are 10 slides of routine hematoxylin and eosin stain slides and 10 ground section slides of teeth and bone samples. Continuous photographs of the entire slide were captured using an IOS device and a Jenoptik camera attached research microscope at 10× magnification. The photographs were stitched using Adobe Photoshop 2017 software to obtain a whole slide image. Then, two observers analyzed the whole slide image for a possible diagnosis and thus compared the efficiency of both the methods. **Statistical Analysis Used:** The statistical Chi-square test and analysis of variance (ANOVA) H test were done using Statistical Package for the Social Sciences (SPSS) 2010 Software. **Results:** Percentage of measure of agreement was 79.5%, 83.7% and 86.3%, 89.1% for the whole slide images taken in the smartphone and photomicrograph and analyzed by Observer 1 and Observer 2, respectively. **Conclusions:** In this study, it can be seen that the WSI by research microscopes is better than that with a smartphone and a compound microscope but still both the methods are equally good and can be followed with accurate results.

Keywords: Adobe photoshop, microscope, photomicrograph, smartphones, whole slide imaging

Introduction

Microscopic observation of the tissues has come a long way from the time of Anton van Leeuwenhoek to today's sophisticated techniques. The humble glass slide has withstood the test of time, relevant, and used even today. These slides are being digitized world over with various companies offering a wide range of whole slide imaging (WSI) digitizers.[1]

WSI, also commonly referred to as "virtual microscopy", aims to emulate conventional light microscopy in a computer-generated manner. Practically speaking, WSI consists of two processes. The first process utilizes specialized hardware (scanner) to digitize glass slides, which generates a large representative digital image (so-called "digital slide"). The second process employs specialized software (i.e., virtual slide viewer) to view and/or analyze these enormous digital files. It shows an example of a contemporary WSI scanner and virtual slide viewer. During the last decade, a wide range of commercially available WSI

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instruments have been developed. These devices are meant to meet the needs of a diverse user base.[2]

In recent years, two alternative solutions have attracted much academic and commercial interest. One solution is aborting the automation feature, thus leaving the operator to control the microscope manually, reducing the product package to a dedicated digital camera and software, and costing as little as the US $10,000. Other attempts have made use of smartphones, which not only have integrated capturing and processing abilities but are also widely distributed among clinical professionals, thus lowering the start-up cost to near zero.[3]

With a mainstream smartphone mounted on the eyepiece of an optical microscope, a pathologist can scan the whole slide into a virtual copy by simply operating the microscope following this normal examination procedure. The image quality, based on the clinical evaluation results, is considered on par with high-end whole slide scanners for most tissue types, as assessed by senior pathologists, and its speed has been proven to be adequate for general applications.[4]

Mobile WSI (mWSI), with the increasing processing capacity, pixels in the camera, and slim mobile designs altogether has become a very powerful digital tool. World over many pathology residents and pathologists capture single field view pictures of important spots on the slide for a late review or reference whereas the mWSI uses the panorama picture capture in the dedicated app, and these images are stitched together using an image processing software.[1]

This study is aimed to evaluate and compare the use of Jenoptik photomicrography and smartphone camera-based WSI for various microscopic preparations and slides.

Subjects and Methods

The study was conducted at the Department of Oral Pathology and Microbiology in Bapuji Dental College and Hospital, Davangere. The samples included in this study are 10 slides of routine hematoxylin and eosin stain slides and 10 ground sections of teeth and bone slides.

The inclusion criteria for the study included slides that are clean with all the major features clear under routine microscopic study; histopathological slides that were prepared less than 5 years ago and hematological, cytological, and microbiological slides that are fresh and prepared less than 2 days before.

The exclusion criteria for the study were slides that were prepared more than 5 years ago, slides that are hazy and unclear, and slides with procedural and staining errors.

The procedure for this study is as follows. Slides that satisfied the inclusion criteria were first observed and pictures were obtained using a Jenoptik photomicrography camera mounted on a Leica RM 125 microscope [Figure 1]. The images acquisition process will take about 15–20 min in the 10× magnification, respectively.

A smartphone will be used and placed in a mount attached to the microscope [Figures 2 and 3]. The mobiles to be tested are Apple iPhone 7 plus. The images will be acquired in the phones using the default camera app. Image acquisition will be started from one corner of the slide, and multiple serial images will be taken in the horizontal axis until the whole tissue was captured. Then, the field will be changed vertically downward to start capturing again horizontally; in this zigzag pattern whole slide will be captured.

Figure 1: Leica research microscope with Jenoptik camera

Figure 2: Instrument used to attach the smartphone to the eyepiece of the compound microscope

Figure 3: Olympus compound microscope

The images were then transferred to a computer and the Adobe Photoshop CS6 software will be used to stitch the panoramic strips and generated a final complete picture of the slide [Figures 4 and 5]. To stitch all images, the software requirement will be of at least 15% overlap in both the axes.

The images were saved in portable network graphic (PNG) format as well as the joint photographic expert group (JPEG). Histopathology, microbiology, hematopathology, ground section, and cytopathology slides were used to generate virtual slides.

The images will be managed on a computer by making a simple folder arrangement of the files and did not require a dedicated software program to retrieve, review or for archival purposes. The images can be viewed on the computer, mobiles, and tablets without using any additional software.

The images will be assessed by both methods by two investigators with more than five years of proficiency based on the clarity of the image obtained, reproducibility of details, ease of operation, ease of reproducibility, and sending.

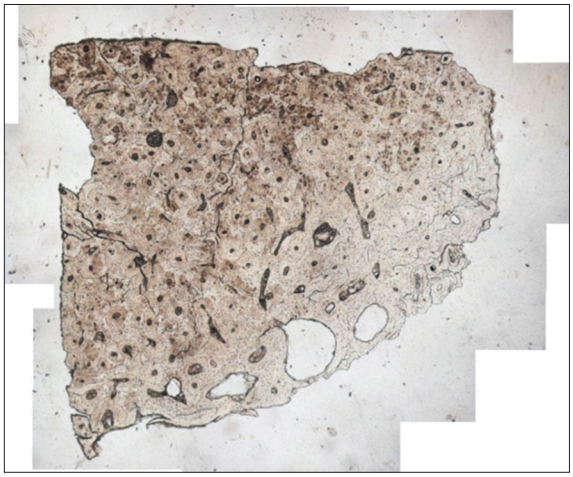
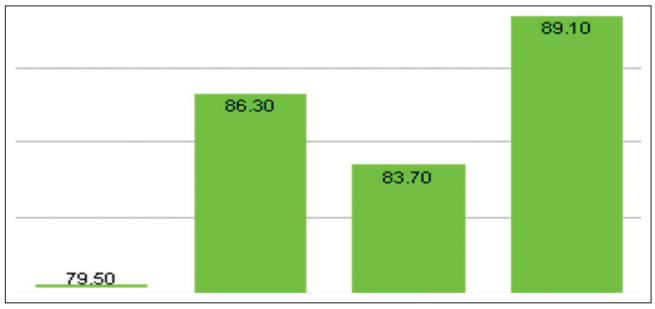


Figure 4: Whole slide image of a ground section of bone

Results

The statistical Chi-square test and analysis of variance (ANOVA) H Test were done using Statistical Package for the Social Sciences (SPSS) 2010 Software. On analysis of the results, it was seen that the percentage of measurement of agreement was 79.5% and 86.3% for the whole slide images taken in the smartphone and analyzed by observer 1 (L1) and observer 2 (L2), respectively. The percentage of measurement of agreement was 83.7% and 89.1% for the whole slide images taken in the photomicrograph and analyzed by observer 1 (P1) and observer 2 (P2), respectively.

Graph representing the percentage of accuracy using both the methods by both the observers.



Discussion

The era of digital pathology continues to evolve at a rapid pace, primarily driven by developments in technology. Persistent gains in computer processing power, data transfer speeds, advances in software and cloud storage solutions have enabled the use of digital images for a wide variety of purposes in pathology.[4]

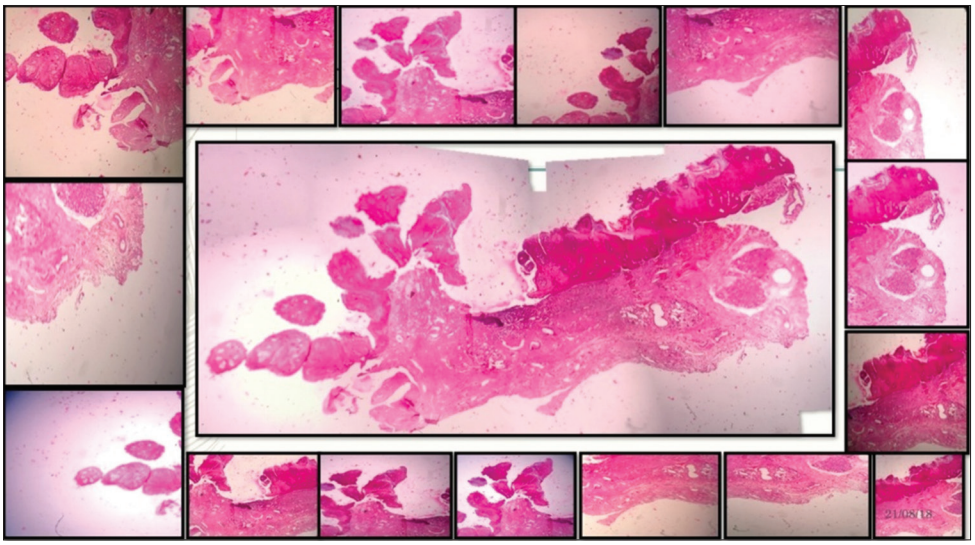


Figure 5: Image with the separate photographs taken and the stitched whole slide image

WSI, also commonly referred to as "virtual microscopy", aims to emulate conventional light microscopy in a computer-generated manner. Practically speaking, WSI consists of two processes. The first process utilizes specialized hardware (scanner) to digitize glass slides, which generates a large representative digital image (so-called "digital slide"). The second process employs specialized software (i.e, virtual slide viewer) to view and/or analyze these enormous digital files.[5,6]

The first WSI scanners, introduced in the late 1990s, were quite primitive compared with their contemporary counterparts.[7] Prior to the introduction of WSI, digital imaging in anatomic pathology relied largely on microscope-mounted cameras to produce "static" digital images.[4]

These static images were of limited clinical utility because they captured only specific regions of a glass slide. Robotic microscopy was accordingly utilized because it allowed the telepathologists to remotely review an entire glass slide.[8] Such standalone robotic systems are no longer vendor-supported and are therefore infrequently being used today.[4]

With the data quality and speed improvements of automated microscopes and whole slide scanners,[9] telepathology has become a major component in pathology labs.[10] Providing remote interpretation of digitized microscopic images and virtual whole slides, it allows diagnosis without physical transportation of samples but just data transfer over the internet. Telepathology not only greatly reduces the financial and time cost but also improves the availability and accessibility of priceless expert resources.[11]

The reliability and practical value of virtual slides compared with the traditional glass version have been extensively assessed and recognized.[12] However, the high financial cost of WSI solutions, and especially the upfront portion, has not seen a significant reduction after years of maturity, limiting its penetration into developing countries and regions, or remote hospitals in the developed world.[13] As supplements for these situations, where limited manual operating is preferred over expensive automation, manual and low-cost alternatives of automated WSI have been studied and developed.[14]

With a mainstream smartphone mounted on the eyepiece of any optical microscope, a pathologist can scan the whole slide into a virtual copy by simply operating the microscope following this normal examination procedure. The image quality, based on the clinical evaluation results, is considered on par with high-end whole slide scanners for most tissue types, as assessed by senior pathologists, and its speed has been proven to be adequate for general applications.

However, the potential and assessment of smartphone-powered WSI have not yet been fully explored. On the one hand, the previous clinical evaluation was limited to working with the cryosection.

In this study, a standard IOS device (iPhone 7 Plus) was attached to a standard microscope which is more convenient and financially feasible as compared to the use of a Jenoptik camera attached with Leica research microscope, which is a more scientifically accurate camera but uses a complex microscopic system and financially not as feasible as compared to the prior. The study also included ground section and hematoxylin and eosin-stained histopathological slides as they are the most commonly used method in micropathology and are obtained as routine cases in a normal pathological setup. The software used for photostitching was the commonly used Adobe CC 2017 Photoshop software as it can be easily worked with two interpreters that were used to diagnose with the whole slide images to increase the accuracy of the study.

In this study, it was seen that on an average, the percentage of accuracy was comparatively more while using a research microscope with a Jenoptik camera (at 83.7% and 89.1%) as compared to that with a smartphone mounted on a compound microscope (at 79.5% and 86.3%) even though the percentage of accuracy of both the methods are fairly high.

Although the scalable WSI (sWSI) solution is clinically recognized by pathologists and achieves its goal of trading full automation for saving financial cost by multiple orders, it suffers from a few technical weaknesses. Many weaknesses are caused by the inherent data model of the image stitching and distortion correction algorithm and thus may not be resolved with further development without switching to a different kernel. Underqualified sample preparation may limit the sWSI's spatial coverage of samples. The diagnosis error introduced by reviewer bias may have underrated the quality of the sWSI virtual slides.[3,15]

WSI helps public health in primary care by avoiding the use of microscopes; patients themselves can carry the image soft copies and report to physicians. Pathologists can also share soft copies via emails and it reduces breaking of slides, paperwork, turnaround time, and long-term costs.

Digital sharing of cases support the group of pathology practices in distant locations by providing ease in diagnosing the rare cases and even allowing the patients to access their complete records. Thus, WSI helps in primary diagnosis and awareness of public health.

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Conflicts of interest

There are no conflicts of interest.

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