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# Histopathology: An Old Yet Important Technique in Modern Science

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## Abstract

Histopathology is a scientific study of disease at the tissue and cellular levels. Despite an old practice, the histopathology reserved one of the substantial sections of disease studies, both in medical and veterinary fields in the modern scientific era. During the current molecular age, some improvements have been made in this practice. The early modification in histopathology is the introduction of immunohistochemistry, which plays an incredible role in tumor diagnosis. The new developments, including digital pathology, multiplex immunohistochemistry, immunofluorescence, brain mapping, neuroimaging studies and artificial neuronal networking are emphasizing novel technologies and almost changed the previous ordinary diagnostic methods. The existing molecular pathology, which evolved mainly from biopsy and autopsy. Currently, the revolutions in molecular biology and in the technology of gene array have developed. The telepathology helping the society and deals with histopathological pictures. It is not far, when molecular techniques would be applied to the lesions prior to its paraffinizations, and the histopathological experts would previously recognize what to study in the sections. The productive move from a visual morphological explanation to obscure molecular science, may be delay, but ultimately be there. This chapter tries to express few of such characteristics of the histopathological practice which assured to be the fast progressing portion of the modern science.

**Keywords:** histopathology, developing countries, diagnosis

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## 1. Introduction

Histopathology is being exercised in most parts of the world and is still in the developmental phase in various developing countries. This branch contributes a significant portion in

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the cutting edge effective diagnosis in pathology through highlighting the unique microarchitectural and morphological results. For long, the histopathologists studied their pathological diagnostic reports exclusively on the tissue growth patterns and cell morphology with usual hematoxylin and eosin (H&E) and few (if any) special stained slides. Today's developed technology makes the computerized histomorphometric diagnosis and prognosis possible, and now the results are more scientific and reproducible. The gene array measures thousands of gene expression, facilitating the researcher for pursuing new and rapid markers for disease diagnosis. By this way a molecular diagnosis of the pathological lesions would be derived prior to the preparation of paraffinized sections. The micro array analysis of DNA and proteomics make likely to figure a comprehensive gene expression belongs to tissue neoplasia and helps in diagnosis, susceptibility and prognosis. Such tests are being done in conjunction with preceding histopathology for better results. Artificial neuronal networking is introduced by a surgical pathologist in whom an artificial neuron is working like physiologically normal one by passing information. The telepathology is dealing with obtaining, spreading, and broadcasting of histopathological pictures through the telecommunication networks viz. internet and satellite. This practice will make the study of whole-slide easy and will let the prompt distributions of the images for early diagnosis and detail disease process. Now most of the shortcomings are being overcome and the imminent archetype of histopathology is hypothesized to be digital in the near future. By this means the histopathologists will confirm diagnosis via virtual images analysis on computers instead of as usual morphometry and the digitized tissue could categorize into various histological grading for quantitative analysis, which results in provision of rapid and improved prospects for diagnosis and treatment of tumorous tissues. In the current thrilling time of pathology, we are challenged with the novel boundary of cutting-edge science and technology, which progress and speed up the diagnostic histopathological technique. But still it is not less than a challenge for achieving such procedures successfully so as to route all the information enclosed.

## 2. History

The histopathology refers to the examination of prepared tissue under microscopic and the practice is however in the development phase in most of the developing countries [1]. It has been found that most of the important lesions which are easy to get during biopsy are still not biopsied in those countries and if so, then most of the important surgically isolated tissues are not being processed accurately for histopathology. Most of the existing gaps that need to be filled is the lack of availability of important good quality chemicals, reagents and instruments viz. unavailability of Microtomes for fine sectioning and electron microscopes for tissue study etc., and unavailability of various very common tests including, immunohistochemistry. Various teaching and research institutes are lacking these facilities and are focusing on H&E only [2]. The concept of staining a tissue is for clear visualization of its microstructure. Most commonly used eosin is an acidic coloring chemical that stains basic structures having negatively charge i.e. the cytoplasm. The other companion color is the hematoxylin which is basic in nature and is imparting the color to the acidic portion of the cell i.e. nucleus [3]. Special staining of the tissue is also out of use, but, if available, then its results will not be appropriate. Only

few private sectors are claiming that they are producing the slides of a good quality, but the methods are still needed to be standardized. If in case any slide is made up as per routine use of advanced countries, there is a bridge of availability of technically trained persons who can read the slides exactly. State of the art facility of equipment's needed to available along with skilled personals that can get the specimens, processed, stained and could read the lesions. The tumor pathology practice is increasing with the time [4] and the histopathology practice is also needs to be rectified as this is a tool of diagnosing the tumor rapidly and is reliable [5]. The developing countries need to accurate the practice of histopathology with the intention that they could help the civilians in diagnosing the existing regional disease conditions. So that the pictures of good results could be cited in international literature. The histopathology is skinning new grace in finding cause and pathogenesis of diseases [5]. A lot of developing countries faced funding's, trained technicians, equipment's and materials problems [6]. One of the emerging trends in the current scientific era is invention of digital histopathology [7], which is considered as a break-through and celebrity in finding and studying numerous varieties of cancers. The current chapter highlighting the use and application of histopathological old techniques and also highlighting the simultaneous use of old and emerging new techniques in the field of histopathology and also debated about its future recommendations.

### **3. Position of histopathology in the modern science**

Disease is developed from the molecular level followed by cellular and tissue level [8]. It is utmost necessary to grab any disease in the stage when it creates changes in the tissues [9]. In histopathology we study scientifically the changes in the affected tissues under the microscope. Though this is an ancient procedure, and still being adopted in medical sciences [10]. This section of pathology enjoying a considerable portion of detail study of most of the humans as well as animal ailments. This outlet of scientific study reserved a substantial position in the modern techniques in effective disease diagnosis. In histopathology the micro-architectural detail of tissue is being highlighted. For this the tissues are being stained with various categories of stains. Although this practice is a time consuming process, however, some improvements have been made in such protocol during the modern era. The new developments in the current novel technologies improved the earlier conventional disease diagnostic procedures, making enabled such practice in a rapid way [11]. The manual protocol is replaced with the automated machines. In recent times the histological image of life sciences is processed in medical sciences, same to that of the engineer [12]. Tremendous improvements have been made in such medical image processing technology. Due to the improvement of the information technology, the tissue imaging technique is the most acceptable, efficient and reliable mean to detect the cancer and other diseases.

### **4. Improvements in histopathology**

The histopathology is being practiced sidewise to the molecular techniques in the technologically developed world, and the less developed countries are still competing to standardize

such technique. The experts in histopathology used the prepared slides entirely on growth patterns, infiltrated cells and tissue morphology with ordinary staining protocol [5]. The H&E staining are widely being used in histopathology and currently, various specific stains are being established [12]. The aim of these stains is to identify the specific affected tissues after imparting it with different dye. Several special stains have been developed and are being practiced successfully in renowned established laboratories. The special stained make the slides easy in differentiation and identification, which are then subjected into rapid computerized histomorphometric diagnosis.

## 5. Position of histopathology in the modern diagnostics

The major ailment around the world is the cancer and is considered as a chronic disease of the age [13]. Numerous death rates owing to cancer have been reported in human and animal population, and its rate is increasing astonishingly with time. The foremost change and upgradation in histopathology is through the use of immunohistochemical methods [11], which playing an incredible role in tumor diagnosis. The applicable prognosis and early diagnosis of biopsy specimens is possible, and the scientific results obtained are now more reproducible. Other than disease diagnosis, most of the tissues are being developed in the laboratories through culture methods [14]. The tissue culture labs are the leading player of the tissues being cultured. These labs recently claimed the culturing and the introduction of the artificial neuronal networking, in whom an artificial neuron is working like physiologically normal one by passing signals and effective information [15].

The existing molecular pathobiology was evolved mainly from biopsy and autopsy [8]. Currently, the revolutions in molecular biology and in the technology of gene array have developed which measures thousands of genes expression, facilitating the researcher for pursuing new and rapid markers for disease diagnosis [14]. By this way a molecular diagnosis of the pathological lesions would derived prior to the preparation of paraffinized sections. The micro array analysis of DNA and proteomics make likely to figure a comprehensive gene expression belongs to tissue neoplasia and helps in diagnosis, susceptibility and prognosis. These tests are being done in conjunction with preceding histopathology for better results [16]. The telepathology helping the society and deals with obtaining, spreading, and broadcasting of histopathological pictures through the telecommunication networks viz. internet and satellite [17]. This practice will make the study of whole-slide easy and will let the prompt distributions of the images for early diagnosis and detail disease process.

Some commonly used histopathological techniques and stains in the developed countries routinely for diagnosis and the need of such methods and stains in the pathology set up of third world countries are.

## 6. Telepathy

It is basically the communication of thoughts among people without the use of common senses, which are specified for thinking, ideas and physical interaction purposes. This is being

done without using body language or words. The main users/players of this technology is called mind readers or telepaths [18].

## **7. Digital pathology**

This histologically based assessment revealed the lymphocytes that are infiltrating in the tumors as a substitute of the host immunity linked responses are being presented to be predictive and possibly chemopredictive in triple-negative and HER2-positive breast cancers. Though, the cooperation of the said lymphocytes, mediators, tumor cells, microenvironmental features, their quantity and associations are still awaiting to be explored. A tool named a digital pathology is anticipated to be used to evaluate the said features targeted and of chemotherapeutic reactions in patient. Based on digital pathology an image-analyzing algorithm are being developed to recognize lymphocytes, stromal, neoplastic cells in addition to obtaining of an image from the slides stained with H&E [19].

## **8. Multiplex immunohistochemistry**

The prototype of molecular form of histopathology is fluctuating from a simple immunohistochemistry (single-marker) to multiplexed immunohistochemical recognition of markers to comprehend the multipart pathological procedures in an improved manner. This method is currently being used to explore the expression of those proteins pattern that is concerned in controlling of immune related checkpoints. In this histopathological technique the microarchitectural environment mapping of tumors is being done. This technique contributes in the understandings of syndrome heterogeneity and delivering all kinds of information you demand on time [20].

## **9. Immunofluorescence**

This is a cell imaging technique and is being used commonly in laboratories. A fluorescent microscope is used in this technique and is run primarily for microbiological studies. The antibodies used in this method are conjugated chemically with fluorescent dyes which are then called labeled antibodies. These antibodies are later on attached to the specific cellular antigens. This largely valid method commonly practiced by the scientists to evaluate the localized and endogenous levels of protein and antigens expressions [21].

## **10. Brain mapping**

Brain mapping is also including in the future of histopathology in which the positive and negative command is being generated in various segments of the brain. The technology used to detect the specified cortical regions of the brain during mapping is via very sensitive fMRI techniques [22].



## 11. Neuroimaging studies

It is a sort of new technique used for brain imaging. Through this technique the possible changes are visualized in the diseased brain. This technique is being used extensively in the infants and young babies. Because in young age the changes in the brain are being observed very rapid. The imaging techniques of the brain authorize a friendly procedure of neuro-receptor binding and neurophysiology. These imaging is sensitive and powerful tools for research purpose especially during pathophysiological studies of some foremost depressions. Currently, this practice is being used to observe the working capabilities of the brain and how the brain is fulfilling various major tasks including language processing [23].

Some commonly used histopathological stains in the developed countries routinely for diagnosis and the need of such stains in the pathology set up of third world countries are including as Stro-1 and vWF stains [24]; Simplified myeloperoxidase stain using benzidine dihydrochloride [25]; triphenyl tetrazolium chloride and tetrazolium red [26]; silver stain [27]; Giemsa, Diff3 and Warthin-Starry stains [28]; Gram and Steiner stains [29]; Toluidine blue, Masson's trichrome, Mallory's trichrome, Alcian blue. Reticulin, Azan, van Gieson, carmine, silver nitrate stains [1, 30].

Now most of the shortcomings are being overcome and the imminent archetype of histopathology is hypothesized to be digital in the near future. By this means the histopathologists will confirm diagnosis via virtual images analysis on computers instead of as usual morphometry and the digitized tissue could be categorized into various histological grading for quantitative analysis, which results in provision of rapid and improved prospects for diagnosis and treatment of tumorous tissues.

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