

# Introduction

The history of medicine shows how societies have changed in their approach to illness and disease from ancient times to the present. Early medical traditions include those of Babylon, China, Egypt and India. The Hippocratic Oath was written in ancient Greece in the 5th century BCE, and is a direct inspiration for oaths of office that physicians swear upon entry into the profession today. In the Middle Ages, surgical practices inherited from the ancient masters were improved and then systematized in Rogerius's *The Practice of Surgery*. Universities began systematic training of physicians around 1220 CE in Italy.

Invention of the microscope was a consequence of improved understanding, during the Renaissance. Prior to the 19th century, humorism (also known as humoralism) was thought to explain the cause of disease but it was gradually replaced by the germ theory of disease, leading to effective treatments and even cures for many infectious diseases. Military doctors advanced the methods of trauma treatment and surgery. Public health measures were developed especially in the 19th century as the rapid growth of cities required systematic sanitary measures. Advanced research centers opened in the early 20th century, often connected with major hospitals. The mid-20th century was characterized by new biological treatments, such as antibiotics. These advancements, along with developments in chemistry, genetics, and radiography led to modern medicine. Medicine was heavily professionalized in the 20th century, and new careers opened to women as nurses (from the 1870s) and as physicians (especially after 1970).

In the previous century medical field progressed by leaps and bounds, both in diagnostics and therapeutics. Invention of microscope brought diagnosis to microscopic level (one that cannot be seen by naked eye). We always believed that seeing is believing. What could be more valuable than seeing at the cellular or tissue level? Thus, arrived the science of cytopathology and histopathology. Medical advancements did not stop at tissue or cellular level either. They went a step further to the molecular level. This stage has been called molecular biology/medicine and includes immunohistochemistry and RTPCRs. For diagnosis molecules or genetic sequences are targeted. Molecular medicine is very highly specific and sensitive. As individual atoms donot exist in nascent form in human body, therefore, further advancements shall relate to molecular pathology only.

## INTRODUCTION TO THE TECHNOLOGY

Technical manual of immunohistotechnology, histotechnology and cytotechnology comprises technical preparation guide for specimen removed out of body during

surgery, aspirates and body fluids. Macroscopic and microscopic examination (*histopathology*—by Greek definition *histos*—tissue; *pathos*—suffering; *logia*—study of) of the specimen enables understanding of changes occurred in the disease and diagnose, on basis of which the medical treatment starts. Tissue has to undergo various stages of processing for preparation to a microscopic slide for evaluation (*histotechnology*). This involves tissue processing, embedding, microtomy, staining and mounting.

*Histotechnicians/histotechnologists*, are trained personnel with excellent mechanical skills, having expertise on equipment and knowledge of anatomy, science of tissue processing and various chemical reactions in staining protocol. They play very important role in preparation of slide for microscopic evaluation. Preparation of slides for frozen sections, routine H & E stain, special stains like PAS, reticulin, masson and many more. Immunohistochemistry, molecular tests have become important ancillary tests identifying type of tumors for diagnosis, prognosis and therapeutic aspect.

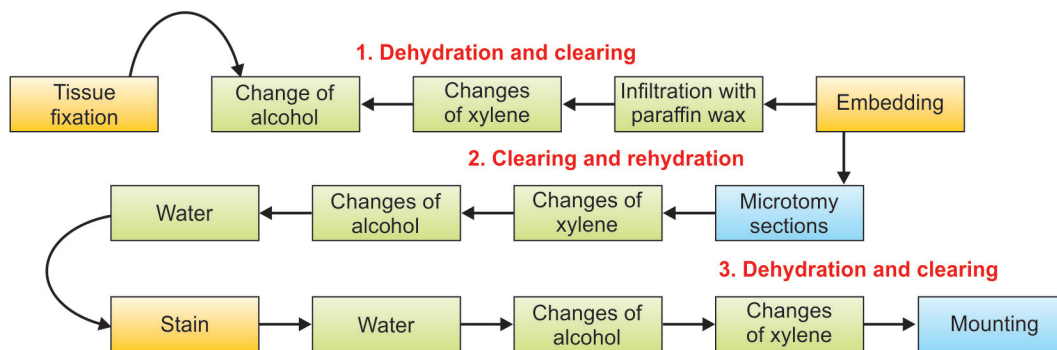
The sensitivity of the technique has improved over a period of years from immunofluorescence labeled antibody system to labeled polymer-based systems and also overcome the effects of formalin fixation for identifying epitopes. With introduction of digital pathology, it is essential to monitor quality of slide preparation.

### Understanding Basics of Tissue Processing (Fig. 1.1)

#### Action for Removal of Water

Water is found in tissue in two forms, free and bound water. The bound water molecule is an integral part of macromolecules of the cell. Proteins, lipids, carbohydrates and nucleic acids are major macromolecules of the cell having their own specific function within living cells. Correct dehydration defines removal of free water, leaving bound water intact.

1. Fixation and gross examination: Fixation is the process of preserving tissue from decomposing, tissue is placed in a solution called fixative. All fixative solutions are aqueous in nature
2. Gross examination is the macroscopic observation and dissection of tissue for purpose of diagnosis
3. Processing and embedding: **Step 1**



Basic of tissue processing cycle—dehydration—rehydration—dehydration

**Fig. 1.1:** Basics of tissue processing

Paraffin wax is universally used for embedding tissue section. Most of the fixatives used for tissue preservation are aqueous solution. Water is removed from aqueous fixed tissue sections using changes of alcohol and alcohol is removed using xylene making place for paraffin wax to infiltrate. This process hardens tissue and helps in section cutting.

4. Microtomy: Microtomy is an art by means of which tissue is cut into thin sections. Microtome is an instrument used for cutting sections.
5. Staining and mounting: **Step 2**  
Staining procedure imparts color to tissue components which can be visualized under microscope. Paraffin wax is removed using xylene and sections are brought to water treating with alcohol
6. Mounting: After staining is complete tissue sections dehydrated, cleared and are mounted using mounting medium DPX: **Step 3**