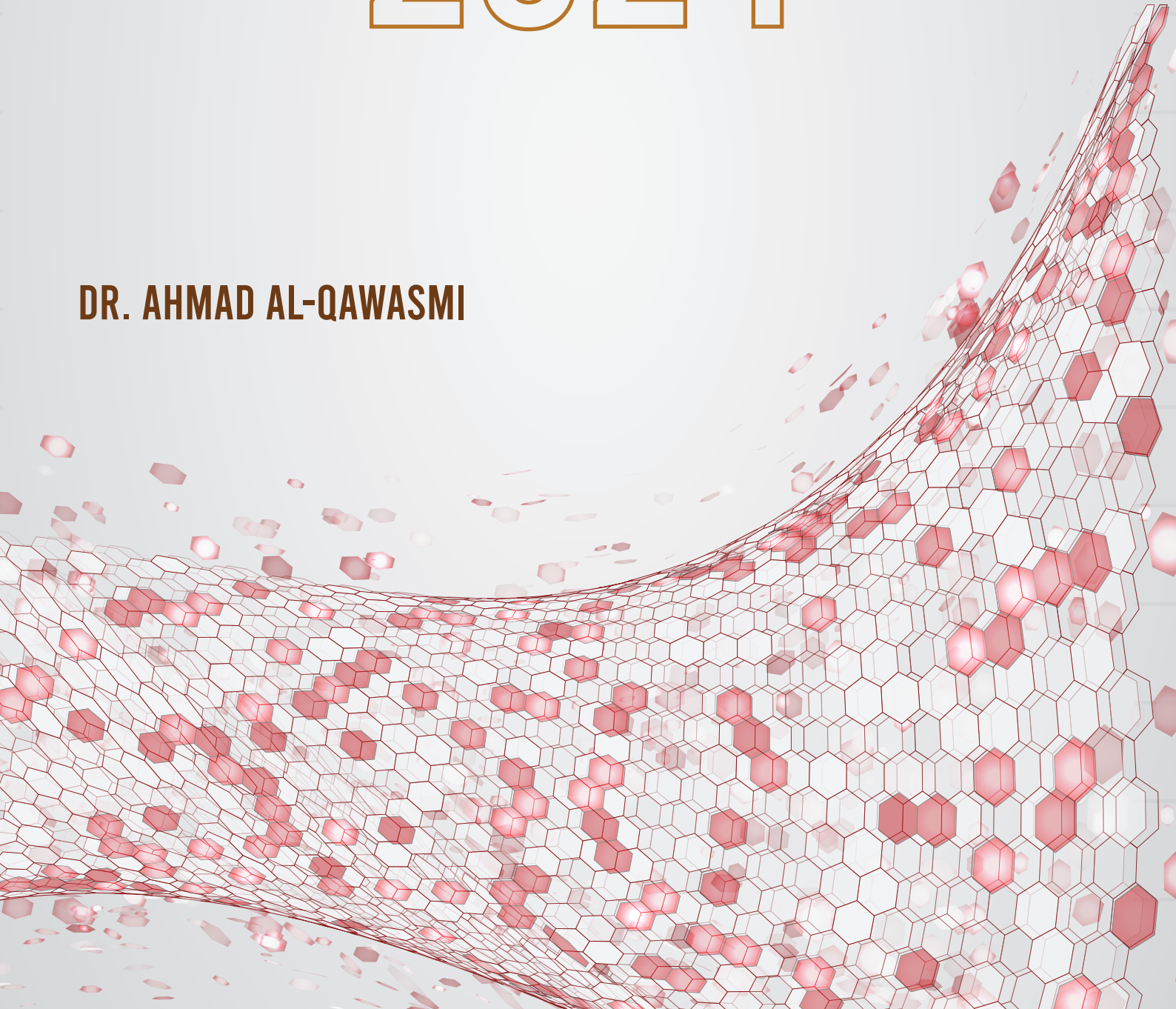


# HISTOLOGY

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## ❖ Introduction to histology

- **Histology:** The study of the tissues of the body and how these tissues are arranged to constitute organs
  - **Tissue:** A group *cells* surrounded and impeded in *ECM*
  - **ECM:** Extracellular matrix is a solution between the cells, consisting of water, glycoproteins, electrolytes, ...
- Tissue processing for histology:
  - **Fixation:** Small pieces of tissue are placed in solutions of chemicals that *cross-link* proteins and *inactivate degradative enzymes*, which **preserves** cell and tissue structure
    - ✓ Most popular fixative is *formalin*
  - **Dehydration:** The tissue is transferred through a series of *increasingly concentrated alcohol* solutions, ending in 100%, which **removes all water**
  - **Clearing:** **alcohol is removed** in an *organic solvent* in which both alcohol and paraffin are miscible
  - **Infiltration:** the tissue is placed in melted *paraffin* until it becomes completely **infiltrated** with it
  - **Embedding:** the tissue is placed in a small *mold* with melted paraffin and allowed to **harden**
  - **Trimming:** the paraffin block is trimmed to **expose** the tissue for sectioning (*slicing*) on a microtome
    - ✓ The perfect thickness of the section is 7-10  $\mu\text{m}$
  - **Staining:** adding a stain (pigment) to enhance the contrast of the image

Levels of organization of the body:

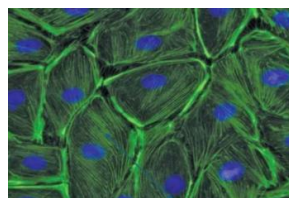
- Chemical (Atoms, Molecules)
- Cellular
- Tissue
- Organ
- System

## ❖ Microscope

- **Light Microscope**
  - Uses *visible light*
  - Maximum resolution *0.2  $\mu\text{m}$*
  - Has 2 main lens, ocular and objective lens
    - ✓ The magnification power of the microscope equals the **multiplication** of the 2-lens power
    - ✓ Permit magnification of 1000- 1500 time
- Types of light microscope:
  - *Bright-field microscopy*
    - ✓ **Stained** tissue is examined with *ordinary light* passing through the preparation
    - ✓ Includes an optical system and mechanisms to move and focus the specimen
    - ✓ The **condenser** collects and focuses a cone of light that illuminates the tissue slide on the stage
    - ✓ Objective lenses enlarge and project the illuminated image x4, x10
    - ✓ The two eyepieces or oculars magnify this image another x10 and project it to the viewer, yielding a total magnification of x40, x100, or x400
  - *Fluorescence microscopy*
    - ✓ In fluorescence microscopy, tissue sections are irradiated with **Ultraviolet (UV)** light and the emission is in the visible portion of the spectrum

**Resolving power:** the *smallest distance* between two structures at which they can be seen as *separate* objects (high quality)

- Objects smaller or thinner than  $0.2 \mu\text{m}$  (such as a single ribosome or cytoplasmic microfilament) cannot be distinguished
- It determines the quality, clarity and richness of detail, and depends mainly on the quality of its *objective lens*



**Fluorescence:** when certain cellular substances are irradiated by light of a proper wavelength, they emit light with a longer wavelength

- ✓ The fluorescent substances appear bright on a dark background
- ✓ For fluorescent microscopy the instrument has a source of UV or other light and filters that select rays of different wavelengths emitted by the substances to be visualized
- ✓ It can use an Immune-Fluorescent staining which is more specific (using antibodies)

➤ *Phase-contrast microscopy*

➤ *Confocal microscopy*

➤ *Polarizing microscopy*

- **Electron Microscope** uses a *beam of electrons*, and it has 2 types:

➤ *Transmission electron microscope (TEM)*: produces **2D images** where the electrons penetrate the cell and can produce images for the internal structures and organelles

➤ *Scanning electron microscope (SEM)*: produces **3D images** where the electrons can't penetrate the surface producing images for the cell surface

