

Mustansiriyah University

College of science

Biology Dept.

Zoology

4th class

Laboratory Technique {*Histological Technique*}

(7)

Histochemistry

Histochemistry can be defined as 'the identification, localization and quantification, in cells and tissues and by chemical or physical tests, of specific substances, reactive groups and enzymes.'

Application

- ❑ Identify, quantify, and localize
 - chemical substances
 - gene expression
 - biological structures, organelles
 - specific cell types
- ❑ Clarify cell and tissue structure and morphology.
- ❑ Demarcate functional boundaries.

Special Stains for Tissue Types

- Carbohydrate
- Nucleic Acid
- Lipid
- Amyloid
- Stains for Micro-organisms
- Connective Tissue Stains
- Pigments and Minerals

Enzymes Histochemistry

- **Enzymes** are proteins that catalyse chemical reactions without them being changed chemically.
- A **catalyst** is a substance that changes the rate of the reaction of chemicals without being consumed by the reaction.
- A **substrate** is usually either an organic compound or ion becomes more chemically active than it would normally be towards another reactant.

Factors that influence Enzyme demonstration

- **Enzymes** are removed or destroyed by fixation, while others are sensitive to freezing and thawing, so compromises have to be made.
- **Non-optimal substrate**: sometimes optimal substrate concentration can't be obtained because of poor substrate solubility.
- **Non-optimal temperature**: there is an optimal temperature for enzyme activity, Enzyme activity is usually destroyed at temperatures greater than 56°C .
- **Non-optimal pH**: most enzymes are best demonstrated at a pH near 7.0, however, there are exceptions, as with acid and alkaline phosphatases.
- **Inhibitors**: an excess of diazonium salts in the substrate, fixatives, heat and some metallic ion may decrease or completely abolish enzyme activity

IMMUNOHISTOCHEMISTRY

Immunohistochemistry :- is a technique for identifying cellular or tissue constituents (antigens) by means of antigen antibody interactions, the site of antibody binding being identified

Principle

- ❑ The principle of immunohistochemistry is the localization of antigens in tissue sections by the use of labeled antibodies as specific reagents
- ❑ Antigen-antibody interactions that are visualized by a marker such as fluorescent dye, enzyme, radioactive element or colloidal gold.