Histochemistry combines the techniques of biochemistry and histology in the study of the chemical constitution of cells and tissues. The importance of histochemistry has decreased as IHC methods have developed.

The goal of histochemistry is to provide color and contrast to microscopic images. The field uses disparate techniques to accomplish the specific labeling of biological structures.

STRUCTURE AND CHEMICAL COMPOSITION OF ORAL TISSUES

Oral structures are primarily composed of connective tissue and epithelial linings and associated glands. An understanding of these structures and their chemical composition is important in the consideration of biologic problems related to oral health. Significant chemical constituents of these tissues are proteoglycans, glycoproteins, mucins, and enzymes. Connective tissue Connective tissue is derived from the mesenchyme and consists of various types of cells and fibers that are embedded in an amorphous, semigel, colloidal ground substance.

Histochemistry specifically stains constituents of cells and biochemical tissues: mucins, lipids, nucleic acids, amyloid, microorganisms, and other proteins. The most frequently used stains are the following: Mason's Trichrome (stains collagen green or blue), Van Gieson (stains for elastin), Von Kossa (stains tissue calcification), Safranin-O (stains glycosaminoglycans red), PAS (stains glycogen, neutral mucins, basement membranes, many fungi and parasites red magenta), Congo Red (stains amyloid deposits red or apple green when viewed with polarized light), Prussian blue (stains iron intensely blue), Giemsa (highlights some microorganisms, e.g., *Helicobacter pylori*, *Leishmania*), Ziehl–Neelsen (stains acid–alcohol resistant bacteria bright red)

PAS reaction (Periodic Acid Schiff)

Periodic acid–Schiff (**PAS**) This is one of the most popular a staining method used to detect polysaccharides such as glycogen, and mucosubstances such as glycoproteins, glycolipids and mucins in tissues, It has been shown to be one of the best techniques for demonstrating carbohydrates in tissue.

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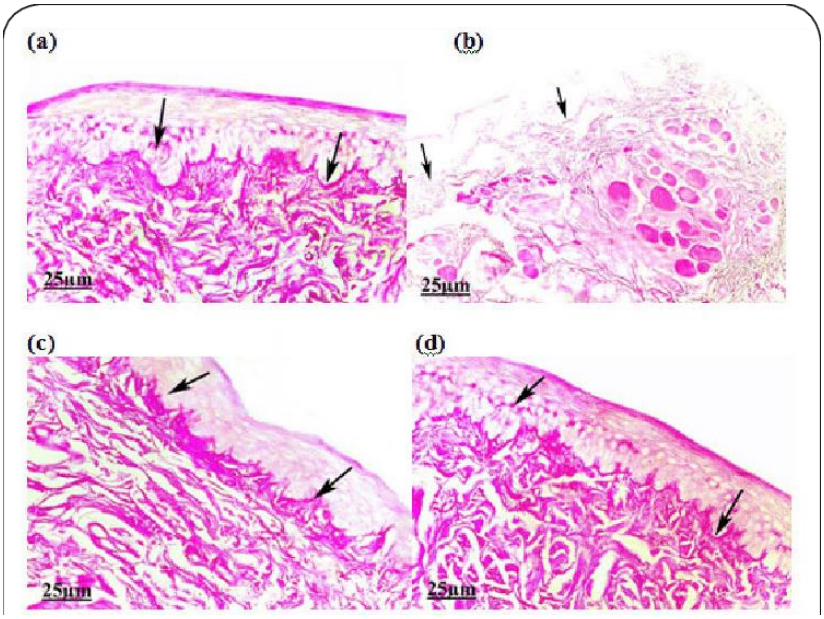
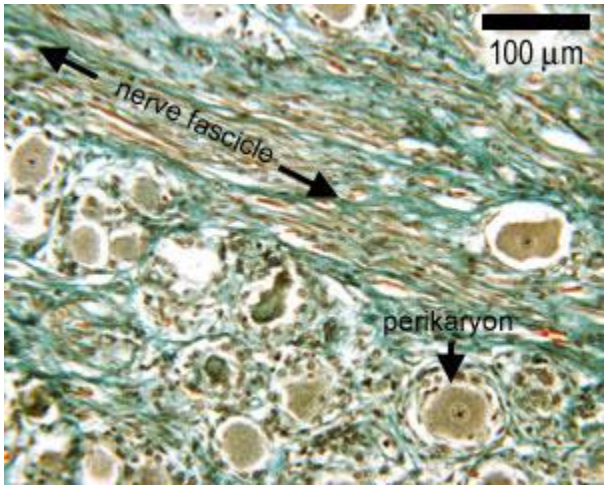
• PAS diastase stain (PAS-D) is PAS stain used in combination with diastase, an enzyme that breaks down glycogen.



photomicrograph of buccal mucosa stained with Periodic acid Schiff (PAS). (a) Buccal mucosa from group I showing intense PAS positive reaction (arrow) [PAS, X400, scale bar=25 µm]. (b) Buccal mucosa from group II showing weak PAS positive reaction (arrow) [PAS, X400, scale bar=25 µm]. (c) Buccal mucosa from group III showing intense PAS positive reaction (arrow)[PAS, X400, scale bar=25 µm]. (d) Buccal mucosa from group IV showing intense PAS positive reaction (arrow)[PAS, X400, scale bar=25 µm].

Masson's trichrome.

This is often used to stain connective tissue. Trichrome - means the technique produces three colours. Nuclei and other basophilic (basic-liking) structures are stained blue, cytoplasm, muscle, erythrocytes and keratin are stained bright-red. Collagen is stained green or blue, depending on which variant of the technique is used.



Immunohistochemical techniques.

These techniques employ antibodies (with antigen specificity) to visualize substances (for e.g. cellular proteins or surface receptors) in tissue sections or cytological cell preparations. These antibodies are connected chemically to enzymes (in immunohistochemistry). Alternatively, fluorescent dyes (as in immunofluorescence) are used. Immunohistochemistry has become more popular than immunofluorescence because the latter requires a microscope modified for ultraviolet illumination and preparations are often not permanent because they fade with time.

In immunohistochemistry the end product is a deposit of opaque or colored material that can be seen with a conventional light microscope and does not deteriorate. The list of substances detectable by these techniques has been greatly enlarged by the development of monoclonal antibodies.