

+ Special stains and TUNEL

Special staining includes a multitude of staining techniques some of which are indicated below with a brief description of their use.

Alcian Blue is one of the most widely used cationic (it has many positive charges on the molecule) dyes for the demonstration of glycosaminoglycans and mucopolysaccharides.

Prussian Blue is used to demonstrate ferric iron and ferritin. This is not a true staining technique rather, it is a histochemical reaction.

Congo Red histology stain is used to stain amyloid.

Fontana-Masson stains argentaffin granules and melanin black.

Cresyl Violet will stain both neurons and glia. It bonds well with acidic parts of cells such as ribosomes, nuclei and nucleoli.

Van Gieson-This histology stain can be used to differentiate collagen and smooth muscle.

PAS (periodic acid Schiff)-This an all-around useful stain for many things. It stains glycogen, mucin, mucoprotein, glycoprotein, as well as fungi. A predigestion step with amylase will remove staining for glycogen. PAS is useful for outlining tissue structures--basement membranes, capsules, blood vessels, etc. It does stain a lot of things and, therefore, can have a high background. It is very sensitive, but specificity depends upon interpretation.

Oil red-The oil red O (ORO) stain can identify neutral lipids and fatty acids in smears and tissues. Fresh smears or cryostat sections of tissue are necessary because fixatives containing alcohols, or routine tissue processing with clearing, will remove lipids. The ORO is a rapid and simple stain. It can be useful in identifying fat emboli in lung tissue or clot sections of peripheral blood.

The **VonKossa** stain is a silver reduction method that demonstrates phosphates and carbonates, but these are usually present along with calcium. This stain is most useful when large amounts are present, as in bone.

The **Trichrome** stain helps to highlight the supporting collagenous stroma in sections from a variety of organs. This helps to determine the pattern of tissue injury. Trichrome will also aid in identifying normal structures, such as connective tissue capsules of organs, the lamina propria of gastrointestinal tract, and the bronchovascular structures in lung.

* No guarantee of success when an uncharacterized antibody is tested.

*TUNEL

The TUNEL method identifies apoptotic cells *in situ* by using terminal deoxynucleotidyl transferase (TdT) to transfer biotin-dUTP to these strand breaks of cleaved DNA. The biotin-labeled cleavage sites are then detected by reaction with HRP conjugated streptavidin and visualized by DAB showing brown color.

It should be noted that results with this technique are intimately associated with the quality (fixation etc) of the tissue(s). Thus if paraffin blocks are provided by the researcher, success cannot be guaranteed.