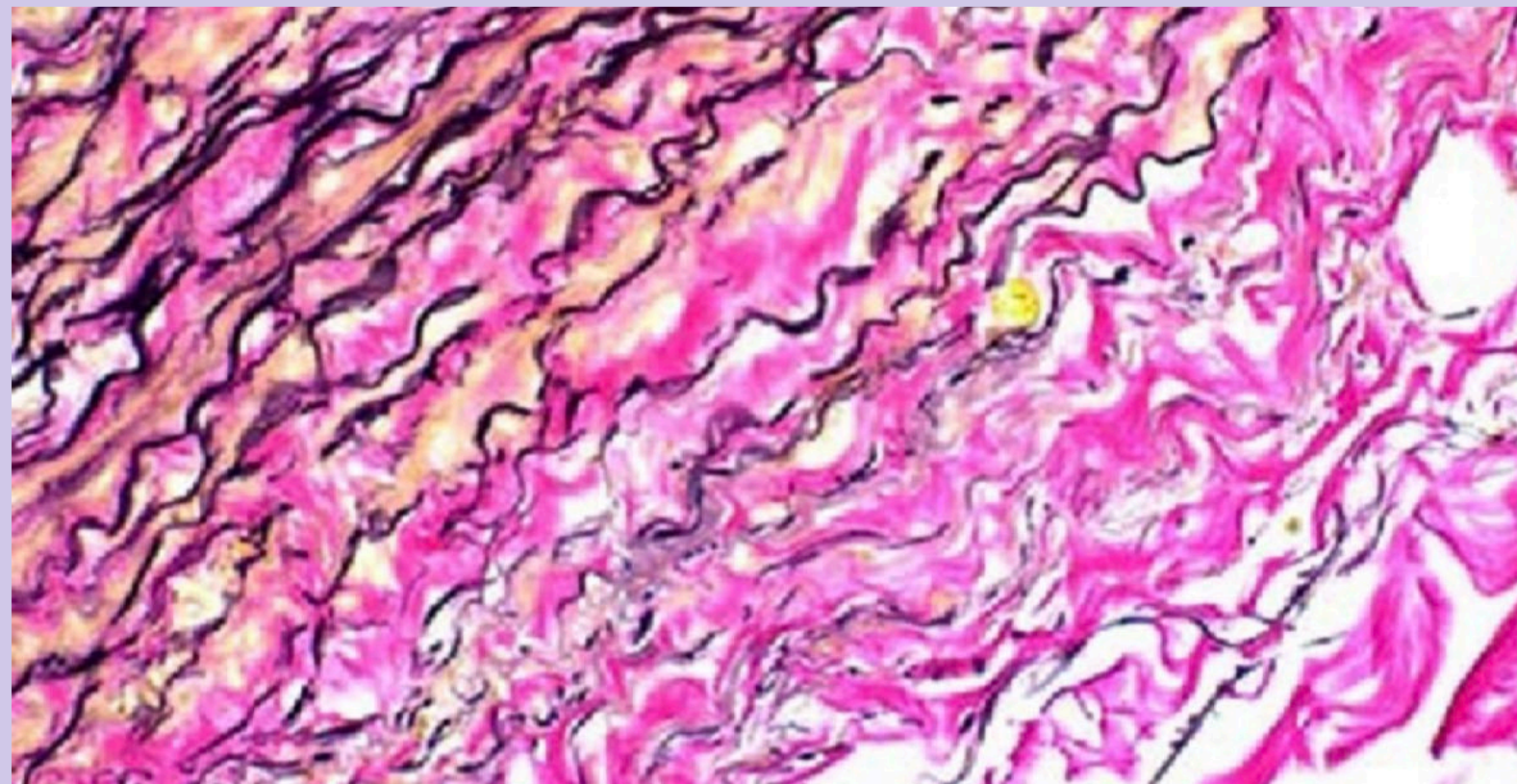


# Mauve to Mounting: A Foundation for Diagnostic Staining

An incidental finding when attempting to synthesise quinine led William Henry Perkin to the discovery of a mauve dye. The outcome of this process resulted in the production of a whole range of other synthetic staining compounds that are applied wherever dyes are used, including in cellular pathology.

## Perkin's Colouring Matter

A serendipitous discovery, in Perkin's own words... "I took a cold solution of sulphate of aniline, or a cold solution of sulphate of toluidine, or a cold solution of sulphate of xylidine, or a mixture of any one of such solutions with any others or other of them, and as much of a cold solution of a soluble bicarbonate as contains base enough to convert the sulphuric acid in any of the above-mentioned solutions to a neutral sulphate. I then mix the solutions and allow them to stand for ten or twelve hours, when the mixture will consist of a black powder and a solution of neutral sulphate. I then throw the mixture upon a fine filter, and wash it with water until free of neutral sulphate. I then dry the substance thus obtained at a temperature of 100 degrees centigrade, or 212 degrees Fahrenheit, and digest it repeatedly with coal tar naphtha, until it is free from a brown substance which is extracted by the naphtha. I then free the residue from the naphtha by evaporation, and digest it with methylated spirits ... which dissolves out the new colouring matter".



Verhoeff's Haematoxylin and Van Gieson stain.

328, 238, 434, and 27.6%.

- **1956:** At an event in the Midland Hotel in Manchester the attendees gathered to celebrate "100 Years of Mauve". One exhibit gave the CIE system coordinates for the colour that changed the world. These define the redness, greenness, blueness, and lightness of the colour. So, if you want to see what 'Mauve' really looks like, the answer is above...

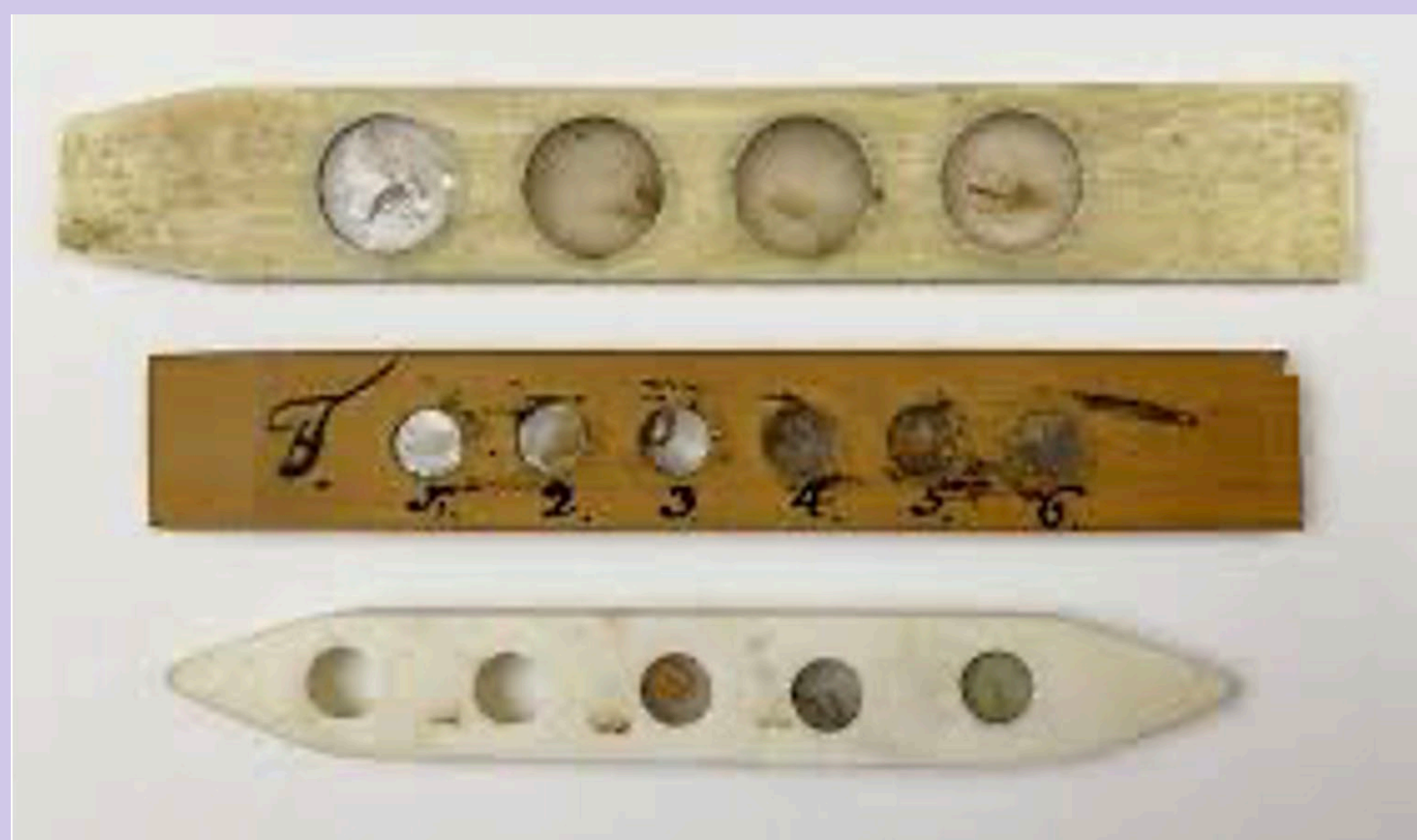
## Eosin: A Mythology Method

Eos (dawn) was, in Greek mythology, the Titan goddess of the dawn, who rose from her home at the edge of Oceanus, the Ocean that surrounds the world, to herald her brother Helios, the sun. As the dawn goddess, she opened the gates of heaven (with 'rosy fingers') so that Helios could ride his chariot across the sky every day.

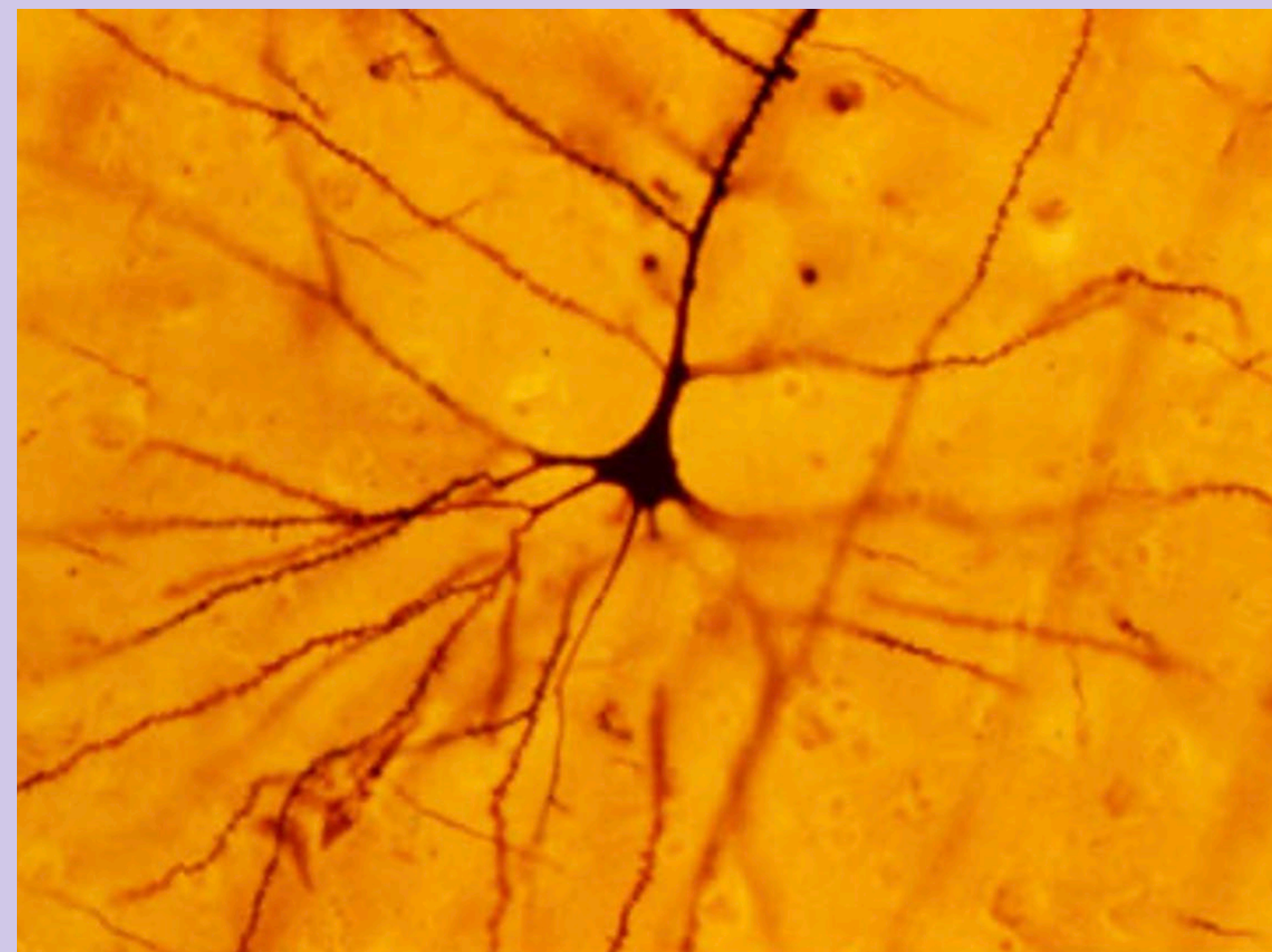
- The term eosinophilic is literally "a love for eosin" and is used to describe any structure that has an affinity for the dye eosin (eg eosinophils, erythrocytes, intra- and extra-cellular proteins).

## Sliders, Slides and Coverslips

- Early microscopists attached specimens for examination to a needle, or dry mounted them compressed between two pieces of mica fixed in ivory or wood.
- Several specimens could be mounted on one 'slider', so called because they were inserted between the leaves of a sprung carrier and slid along for examination.
- As mica had a tendency to split, specimens were later held between two thin sheets of glass, sealed with wax and often held together with decorative and informative coloured paper wraps.
- There were many skilled commercial makers of slides for enthusiastic amateur microscope users. Parts of insects such as butterfly wing, animal hair, diatoms and such like were popular.
- With the development of staining of thin wax embedded sections, it was found these could be easily adhered to glass slides, but the size of slide was not standardised.
- **1839:** The Microscopical Society of London proposed a standard of 3 inches by 1 inch (75 mm x 25 mm). This remains the most frequently used size of slide.
- **1779:** Thin glass cover slips were first described, but they were not generally available – frequently microscopists cut their own from blown glass bubbles.
- **1840:** Chance Brothers began selling coverslips and went on to exhibit them at the Great Exhibition of 1851.



Examples of 'sliders'.



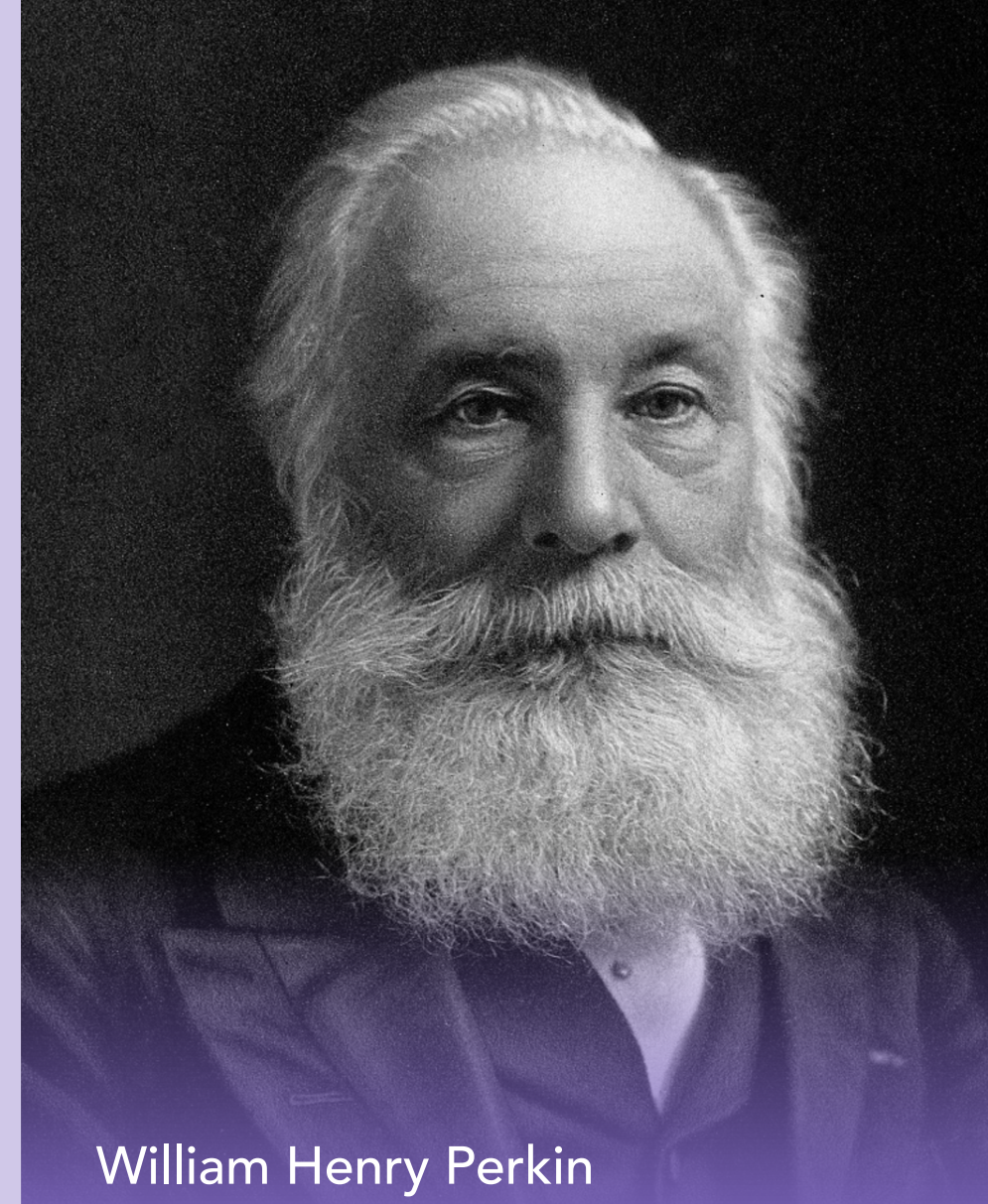
Golgi's method uses silver chromate to stain neurons

Camillo Golgi, the 1906 Nobel Laureate in Physiology or Medicine.

## Aniline dyes in pathology

Synthetic dyes are derived from the modification of the benzene ring, either as benzene, quinone or aniline, by the addition of a chromophore. This can be a paraquinoid ring, an orthoquinoid ring, two azocoupled benzene rings, or a nitro-group attached to a benzene ring. However, these dyes are not 'fast' without the addition of an auxochrome, an NH<sub>2</sub> group being one of the most common of these and formed the basis of the original dye industry. Among the dyes used in pathology that are derived from these structures are:

- Basic and Acid Fuchsin
- Crystal Violet
- Aniline Blue
- Eosin
- Thionin
- Methylene Blue
- Neutral Red
- Carminic Acid
- Bismarck Brown



William Henry Perkin

## A Trio of Staining Eponyms

- **Ira van Gieson** (1866–1913). An American neurologist, psychiatrist, bacteriologist and neuropathologist, van Gieson introduced his Picric Acid / Acid Fuchsin stain initially for use in neurohistology.
- **Camilo Golgi** (1843–1926). Golgi's method was initially named the black reaction (la reazione nera) by Golgi, but it became better known as the Golgi stain or, later, Golgi method. Neuronal cells are filled by microcrystallisation of silver chromate.
- **Santiago Ramon y Cajal** (1852–1934). Cajal's most famous studies were on the fine structure of the central nervous system. He used the histological staining technique developed by his contemporary Camillo Golgi.

## Mounting Media

- Historically, specimens were either dry mounted or an aqueous mountant was used.
- **1795:** Abraham Ypelaar used Venetian turpentine (balsam tree resin dissolved in turpentine) to varnish and preserve his preparations.
- **1832:** Andrew Pritchard introduced gum/isinglass but this was rapidly overshadowed by the introduction, in the same year, of Canada Balsam, the resin of *Abies balsamea*. Its use originally was suggested by Mr J T Cooper and then used by Mr Topping; a prestigious maker of slides. Dissolved in xylene, it was widely used until the latter part of the 20th century.
- **1939:** Kirkpatrick and Lendrum published a formula for a synthetic resin for mounting which became known as DPX – Distyrene, a Plasticiser (tricresylphosphate) and Xylene, which eventually superseded Canada Balsam.
- Many other media have been described, often for specialist applications, including automated cover slipping machines. However, it is likely that coverslips will be superseded by directly applied film.

### Dyeing for a Diagnosis

The topic covered by a History Committee display presented at a previous Congress event. A QR code for these posters is available on the stand.

**Bodies to Blocks:  
A Brief History of Cell Science**  
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