

Preservation of Cadavers and Gross Specimens

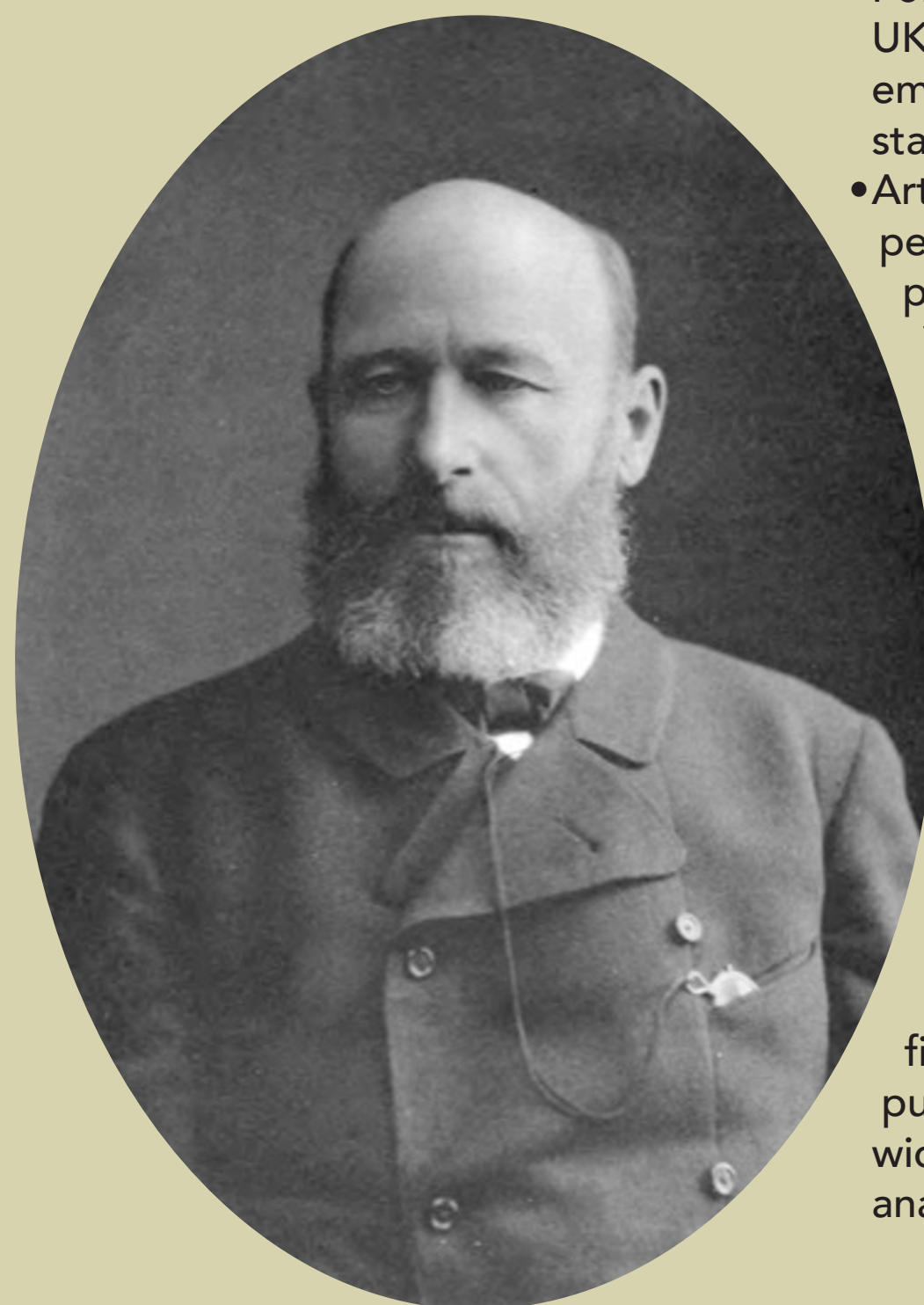
While the polymath Leonardo da Vinci is credited with describing a method of venous injection to preserve cadavers in the late 15th century, anatomical dissection of unpreserved bodies continued to be widely performed until the end of the 18th century.



Mrs Martin van Butchell on display in the Hunterian Museum.

Embalming and Exhibition

- William Hunter, in the late 1700s, is often credited with being the first to report the use of arterial and cavity embalming using a mixture of turpentine and camphor. It was, however, Gabriel Clauderus who, in 1695, described a method using cream of tartar and salammoniac for arterial injection and immersion of the body, then drying it in the sun.
- In 1775, John Hunter embalmed the body of Mrs Martin van Butchell. Her will specified that her husband had control of her fortune as long as her body remained above ground. In 1814, after her death, her husband dressed her in fashionable clothes and displayed her embalmed remains in a glass lid case in a sitting room. Subsequently, her body was displayed in the Hunterian Museum.
- The philosopher John Bentham left instructions that after his body was dissected it would be permanently preserved and be his memorial. His remains are still on public display in the entrance of University College London.
- Other early embalming fluids included turpentine, camphor, lavender oil, mercury sulphide, wine, alcohol, salt and saltpetre.
- Alcohol was famously used for preservation. Following the 1805 Battle of Trafalgar the body of Admiral Horatio Nelson was immersed in a cask of spirits. It is said that when the body was removed from the cask, the sailors present drank the spirits, giving rise to the naval term for rum as 'Nelson's Blood'.



- Following the passing of the UK Anatomy Act of 1832, embalming of cadavers became standard practice.
- Arterial embalming is not perfect and even such carefully prepared remains as those of Vladimir Lenin, who died in 1924, and still on view at the Kremlin, need periodic retreatment.
 - Although Alexander Butlerov discovered formaldehyde in 1859, its ability to act as a tissue preservative was not recognised until 1893 by the German physician Ferdinand Blum. Working with a 4% solution, he noticed that the tips of his fingers became leathery. His publication of this led to the wide use of formaldehyde in anatomy and histology.

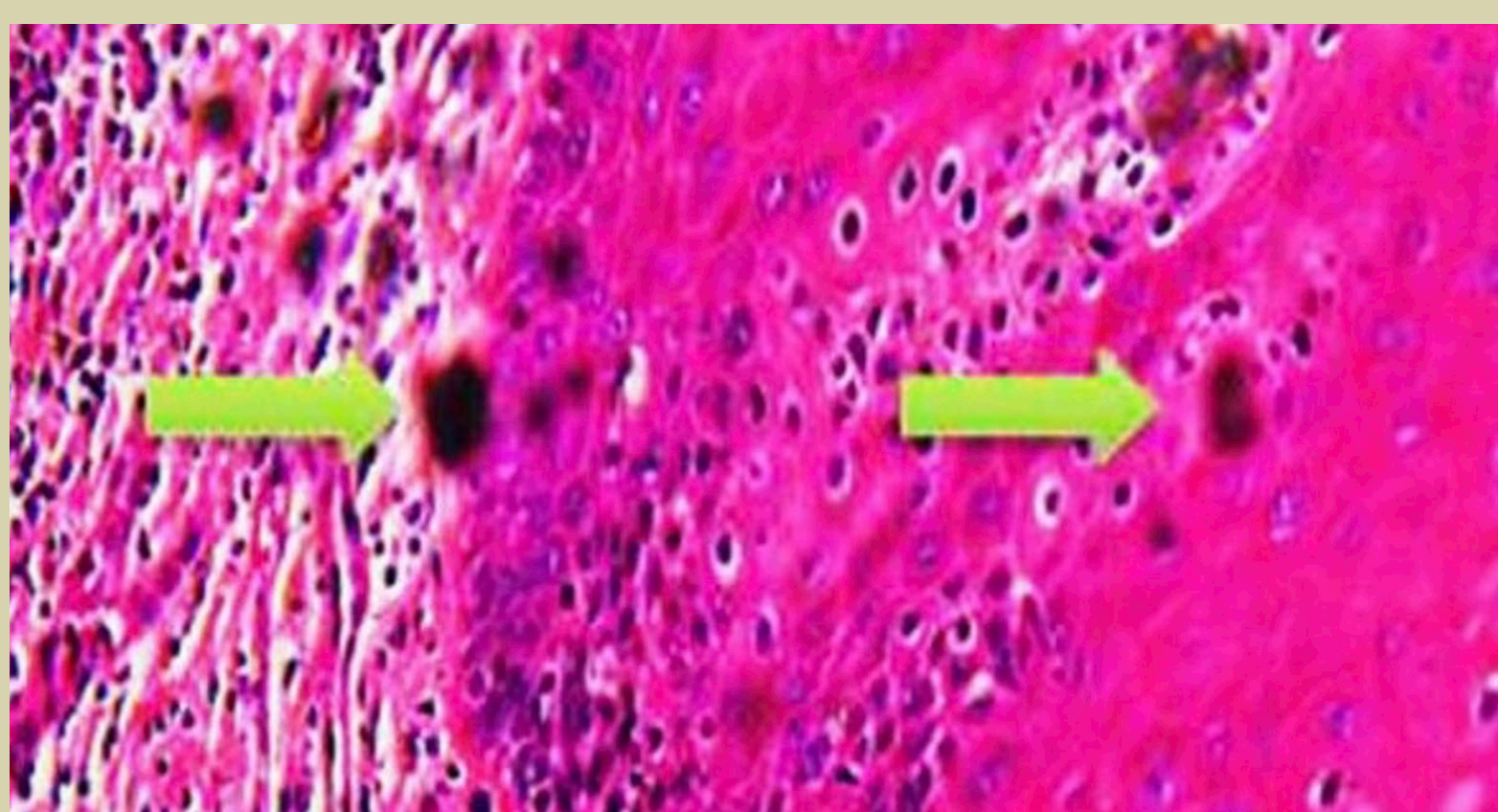
Alexander Butlerov discovered formaldehyde in 1859.



Jeremy Bentham, whose remains are still on public display in the entrance of University College London.

Preservation of Specimens for Microscopy

- Robert Hook described 'cells' in cork in 1665, and Marie Francois Bichat had described 21 types of tissue, but the finer details of tissue and cells were not visible until improvements in microscope optics in the early 1800s. With this came the recognition that better preservation of specimens was needed and this in turn led to parallel improvements in tissue fixation, embedding and microtomy.
- Until the mid-1800s, 'sections' were cut free hand from soft fresh tissue.
- Marcello Malpighi, the founder of microscopic anatomy, reported in 1666 hardening brain by boiling.
- Freezing of tissue prior to sectioning was published by FV Raspail in 1825.
- Fixation as we now know it may be said to start in 1833 when Jacobson suggested the use of chromic acid for hardening tissue.
- Up until the early 1900s, a wide range of chemicals were advocated for use either singly or in combination for hardening and fixation. These included: chromium trioxide (1840), mercuric chloride (1846), acetic acid (1851), potassium dichromate (1860), picric acid (1875), chloroform (1887), osmic acid (1890) and formaldehyde, (1893).
- Named composite fixatives included Zenker's fluid (1894) – mercuric chloride, potassium dichromate, sodium sulphate and acetic acid (Helly later replaced the acetic acid with formalin); Heidenhain's Susa (1916) – mercuric chloride, sodium chloride, trichloroacetic acid, acetic acid and formalin; Carnoy's (1887) – alcohol, chloroform and acetic acid; Bouin's fluid (1897) – picric acid, formalin and acetic acid.
- Culling's 1960s textbook lists 10 individual chemicals and 19 named fixatives. Most being highly toxic, corrosive, inflammable or explosive, and both unbuffered formol-saline and mercuric chloride produced pigments which interfere with staining.



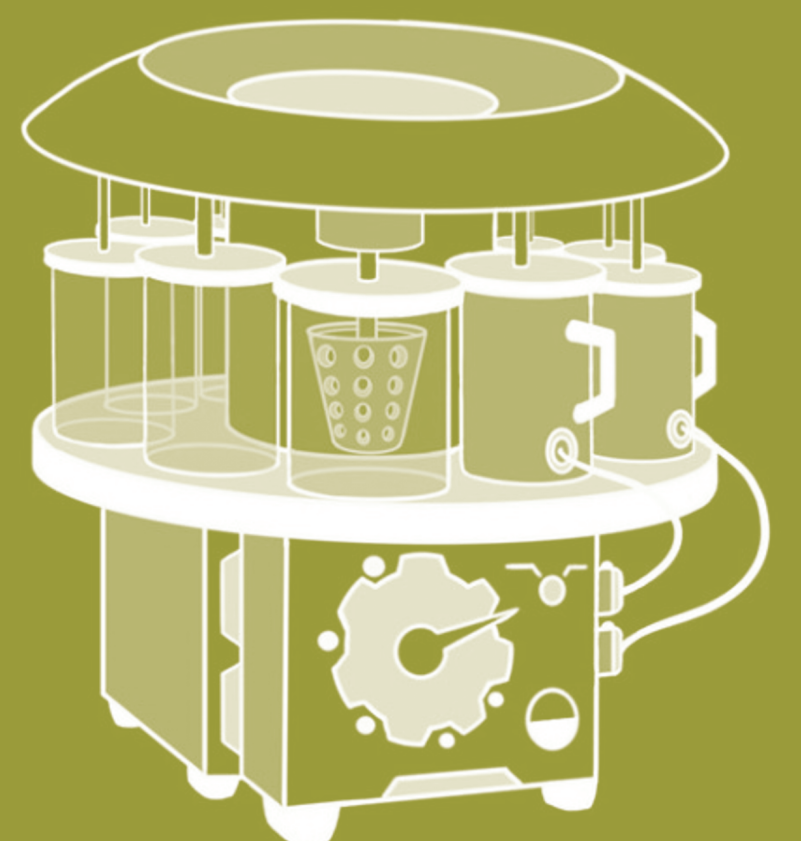
Pigment interferes with staining when tissue is fixed in unbuffered formol-saline.



Marcello Malpighi, the founder of microscopic anatomy

Embedding and Impregnation

- The term 'Embedding' was originally used for methods in which the tissue was surrounded by a supporting material to keep it steady to aid the cutting of sections. Among the materials used were pith, hardened egg white and beeswax.
- The introduction of paraffin wax embedding is attributed to Edwin Klebs in 1867. However, he was following the earlier method of Salomon Strickler who surrounded embryos in a mixture of hot stearin and beeswax. Klebs rejected the method as paraffin wax did not infiltrate tissue.
- In 1868 the Swiss embryologist Wilhelm His, developed an impregnation method. He dehydrated chicken embryos with alcohol, cleared them in lavender oil and dripped hot paraffin wax onto them.
- Ethanol became the most widely used dehydration fluid, but acetone, dioxane, and Cellosolve (2-ethoxyethanol) have also been used.
- 'Clearing agents' were originally used to make the specimen transparent for microscopy and included aniline oil, cedarwood oil, clove oil, benzene, toluene, xylene and chloroform. Not all clarified the specimen but did clear out the alcohol and allowed the wax to impregnate.
- All the steps of this process were manual, the tissue being transferred to each fluid in succession or the fluid decanted and replaced.
- Although G Arendt described an automated tissue processor in 1909, it wasn't until 1945 that the Histokinette (arguably the first piece of pathology laboratory automation) became commercially available.
- Except for the introduction of automation and no longer using the extremely dangerous materials employed during much of the 20th century, the methods used today are still based on those devised towards the end of the 19th century – *Fixation, dehydration, clearing, impregnation and blocking out in wax.*



The Histokinette, which became commercially available from 1945.