Phenoxyethanol as a substitute for formaldehyde in the preservation of anatomical specimens

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Introduction:

The human cadaver has been used as a distinct educational tool within the framework of undergraduate medical education. One important pre-requisite for the use of human cadavers in educational settings is the appropriate preservation of the cadaver. Preservation is said to be successful when the cadaver is kept safe from decomposition, harm and destruction. This preservation is attained by the use of certain chemicals to treat the cadaver. This process is called as embalming. Before the introduction of phenols, carbolic acid and formaldehyde in the field of preservation, the materials used for preservation were alcoholic solutions of arsenic of alumina salts prepared in different concentrations.

In terms of historical interest, the sole reason for embalming cadavers was for religious belief and associated with afterlife. The methods of embalming used initially by the Egyptians and around the world includes natural methods of preservation which includes freezing, dessication or exsiccation, by either dry method or cold method, or by the nature of the soil at the burial site. [1] Another method described in history was immersion on honey which was seen among the Persians. The cadaver of Alexander the great was treated with this method. The Nigerians used large quantities of alcohol concentrate, potash, herbal leaf and kernel oil. [2] The earliest known form of artificial preservation was the used of pulverized cinnabar which ensured good preservation. [3]

During the middle ages, embalming comprised of immersion of the body in alcohol and use of preservative herbs which are placed in incisions made in fleshy parts of the body and then wrapping the body in waxed or tarred sheets. In the following renaissance period, the various scientific developments in medicine required human bodies for dissection and hence preservation required more refined embalming techniques. [4] Various chemicals, waxes, naturally occurring substances were injected into arteries for preservation. Arsenic was commonly used in arterial injections for preservation. Phenols were introduced in embalming by Laskowski around the middle of the 19th century. He used phenol and glycerin as the vehicle and later replaced glycerin with alcohol.

The discovery of formaldehyde dates back to 1869 by a German chemist August Wilhelm von Hofmann. This gave excellent results over the years and became the foundation for modern methods of embalming. However, the concentration of formaldehyde was debated to be between 3% and 10%. Formaldehyde is the simplest and oldest widely used chemical preservative for the preservation of biological specimens especially used in dissection. It is prepared by mixing the commercially available formalin solution with tap water in the proportion of 3:1. [3] The immediate adverse effects of using formaldehyde was known to be skin irritation, conjunctivitis, headache and irritation of the respiratory systems. It has been found that formaldehyde increases the risk of nasopharyngeal [6], sinonasal, lymphatic and hematopoietic cancers in occupationally exposed workers [7]. Formaldehyde has been an area of extensive study because of its irritant properties on the eyes, nose and throat, the discovery that it may induce nasal tumors in rodents and that it might increase the risk of certain cancers. Gronroos as summarized that formaldehyde is not appropriate solely as an ideal preservation agent. [8] Hence researchers are looking for a complete alternative to this potential carcinogen.
Phenoxyethanol is a non- toxic chemical which is lightly scented and is commonly used in cosmetic products and first aid products. It is relatively inexpensive, non- flammable, slow to evaporate, an effective anti microbial agent. Because of all these properties, it can be used as a preservative and softener. It is just a preservative. An additional agent is essential for fixation when phenoxyethanol is used. Phenoxyethanol has been shown to be a possible replacement for formaldehyde due to its softness and flexibility. Another use of phenoxyethanol is to wash out excessive formaldehyde from cadavers. The aim of this article is to review the usage of phenoxyethanol as an alternative to formaldehyde in the routine preservation of anatomical specimens.

Discussion:

Chemistry of formaldehyde:

Formaldehyde is a flammable, colourless and readily polymerised gas at ambient temperature. It occurs as a natural compound in umpolluted ambiet air at concentrarions less than 1 microgram/ mm^3. Formaldehyde adopts several different forms. One important derivative is the cyclic trimer metaformaldehyde or 1,3,5-trioxane with the formula $(CH_2O)3$. There is also an infinite polymer called paraformaldehyde. These compounds have similar chemical properties to the molecule CH_2O.

When dissolved in water, formaldehyde forms methanediol, with the formula $H_2C(OH)_2$. This also exists in equilibrium with various oligomers, depending on the concentration and temperature. A saturated water solution, of about 40% formaldehyde by volume or 37% by mass, is called "100% formalin". [9]

The uses of formaldehyde:

Formaldehyde is used commercially in the manufacture of plywood, plastics, paints, lubricants and dyes Formaldehyde appears in automotive emissions [10] or tobacco smoke and is added to many industrial products. It is also a byproduct of fires, cigarette smoke and automotive exhaust [11] [12]. Once formaldehyde is absorbed, it gets easily broken down into a non-toxic compound called formate, which know excreted in urine and is converted into carbon dioxide and breathed out of the body. But formaldehyde can be toxic, allergic or carcinogenic [13]. Though they are harmful, they are still used because of the superior effects they yield.

Why replace formaldehyde?

The most common effects from the exposure to formaldehyde vapor include sensory irritation of the eyes, nose and throat when sufficient concentrations are reached . Continued exposure to formaldehyde can be broken down into those associated with odor detection, followed by sensory irritation and associated upper respiratory tract endpoints when progressing to higher concentrations and effects such as nasal tumors. Chronic exposure to formaldehyde has been said to be associated with immunological hypersensitivity. This is measured by the elevated circulating levels of IgG and IgE antibodies. Also, a decrease in the proportion of T cells was observed which indicates that the immunity is altered. [14]

Formaldehyde is a known nasal carcinogen in rats and considered as a human carcinogen. In addition to sufficient evidence in animal studies for nasal carcinogenicity due to formaldehyde, the IARC concluded that there is sufficient evidence to support that formaldehyde cause nasopharyngeal cancer in humans. [15] It has been found by the NAS (2007) that " The degree of sensory and irritant effects at lower exposure levels depends on concentration rather than duration". The concentration of formaldehyde may be more important than the total daily dose. [16] [17] In order to reduce the risk of the occurrence of carcinomas and even DNA damage due to formaldehyde exposure in living organisms, a potential replacement for formaldehyde especially in the field of specimen preservation is being worked up on.

Chemistry of phenoxyethanol:

The others names of phenoxyethanol include 1-Hydroxy-2-phenoxyethane, β-hydroxyethyl phenyl ether etc. It is an oily, colorless, slightly viscous liquid at room temperature that belongs to the family of the glycol ether. Although Phenoxyethanol can be found naturally in green tea and chicory, it is synthetically produced in a laboratory.

Use of phenoxyethanol:

Chemically, phenoxyethanol can be described as 2- phenoxyethanol or ethylene glycol (mono) phenyl ether or 1-hydroxy2-phenoxyethane, phenoxetol or phenyl cello solve. It has been found that careful use of 2% aqueous solution of phenoxyethanol for maintaining specimens maintained the quality of the specimen at a high level.

Phenoxyethanol has been proven to be a non- irritant, relatively safe, non-toxic liquid with a mild fruity odour. It contains both aromatic and aliphatic components that give it a high boiling point, high organic solubility and low aqueous solubility. It prevents fungal growth and keeps tissues pliable. The antimicrobial property of phenoxyethanol has been widely studied because it is used as an antimicrobial agent in the cosmetic industry. The mode of action of phenoxyethanol has been studied extensively and found to be gross membrane damage resulting in leakage if cytoplasmic constituents, eventually resulting in cell death. Experiments at sub- lethal concentrations showed that phenoxyethanol caused inhibition of DNA and RNA synthesis, thus inhibiting cell growth. Also, inhibition of malate dehydrogenase and protein gradient disruption inhibited protein energy metabolism. In short, all studies showed that phenoxyethanol acted on several targets in the cell depending on the concentration of the agent. Cell death when phenoxyethanol is used is usually a result of the various mechanisms which result in irreversible cell injuries. [18] [19]

Phenoxyethanol plays a fixed role in extracting formalin from iced specimens (Rumph and Williams, 1988). [20] This is done by extracting previously formaldehyde-fixed material for more than 3 months in 1% phenoxyethanol in tap water. Sharks are best preserved in phenoxyethanol. In a study done by P. J. Smith and James Crossland, it was found that Phenoxyethanol is not suitable as a biochemical preservative for plankton samples. They also stated that phenoxyethanol has an application as a protein preservative in areas where freezing facilities are not available. [21]
Phenoxyethanol has not been used as a primary agent in arterial injection solutions for preservation purposes. However, there are two patents by Campbell in 1995 and Margrave in 1998 from the United States according to which a solution of glutaraldehyde, aromatic ether of ethanol like phenoxyethanol, humectant and an alcohol like ethanol. [22] A buffer of an anti oxidant can be added to the solution to maintain the stability of the solution. The pH must be maintained at 7-9. Additional substances can be added to prevent microbial growth in the solution.

Why is phenoxyethanol a better option in comparison with formaldehyde?

From an anatomical point of view, Phenoxetol gives superior results to those of most other preservatives. The color retention is good [23]and the tissue consistency softer and more flexible than that achieved by other types of preservation. Since no mineral salts are used in the preservative fluid, other embalming methods using such salts will definitely be superior in terms of color retention [24] The most important advantage of Phenoxetol in preservation is its low toxicity. Phenoxetol is recommended as an antiseptic ingredient in medical creams and ointments [25]

In addition to avoiding the well-known hazardous effects of formaldehyde, the use of Phenoxetol also prevents the pungent smell and other local and systemic manifestations of its toxicity because phenoxyethanol has a mild fruity smell.

In an article by Brenner in 2014, Phenoxetol has been described as relatively inexpensive, non-flammable, slow to evaporate, effectively antimicrobial and an excellent tissue preservative and softener. Though it is merely a preservative, not a fixative, it appears to be an effective bactericide at a 1% concentration. It has a broad spectrum of antimicrobial activity and is particularly effective against strains of P. aeruginosa. [26]

Conclusion:

With increasing advancements in the field of research, formaldehyde toxicity and its potential carcinogenicity have gained more importance. The evidence for formaldehyde influencing the occurrence of certain cancers has increased. The use of alternative preservative fluids especially phenoxyethanol are on the rise due to its properties related to preservation and non-toxicity. Phenoxetol proves to be a viable replacement of formaldehyde for long-term preservation of cadavers, human tissue and museum specimens.

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