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**QUESTIONS**

1. Immunohistochemistry and immunocytochemistry is fast becoming an indispensable diagnostic tools in histopathology laboratories discuss.
2. Embalming and plastination compare and contrast.

**ANSWERS**

1. **Immunohistochemistry** is a laboratory method that uses antibodies to check for certain antigens (markers) in a sample of tissue. The antibodies are usually linked to an enzyme or a fluorescent dye. After the antibodies bind to the antigen in the tissue sample, the enzyme or dye is activated, and the antigen can then be seen under a microscope. Immunohistochemistry is used to help diagnose diseases, such as cancer. It may also be used to help tell the difference between different types of cancer.

**Principle**: The selection of antibodies for the immunohistochemical testing is made on the basis of their tumour specificity and the likelihood that they will react with the tumour under evaluation. After tissue sections are incubated with the prospective antibodies, positive reactions (tumour antigen- antibody binding) are identified through the application of one of several detection systems. Those that have the greatest sensitivity use a secondary antibody, reactive against the primary antibody, which is conjugated or linked to an enzyme marker. This system tends to be very sensitive because it allows for the attachment of a relatively large number of enzyme molecules, such as peroxidise, at the antigen site. The colour of the reaction is determined by the selection of a precipitating chromogen, usually diaminobenzidine (brown) or aminoethylcarbazole (red), with which the enzyme reacts.

**The application of immunologic research methods to histopathology** has resulted in marked improvement in the microscopic diagnosis of neoplasm. Although histological analysis of Haematoxylin & Eosin stained tissue sections remains at the core of the practice of head and neck surgical pathology, immunohistochemistry has become a powerful tool in the armamentarium of the pathologist. Traditionally, the goals of diagnostic immunohistochemical studies have been to explore and certify diagnoses by identifying the pathway of differentiation of a given tumour. Immunohistochemistry (IHC) has been proven to be one of the most important ancillary techniques in the characterization of neoplastic diseases in humans, as oncologists demand more specific diagnosis. IHC is a technique for identifying cellular or tissue constituents (antigens) by means of antigen-antibody interactions, the site of antibody binding being identified by direct labelling of the antibody, or by use of secondary labelling method. Immunohistochemical staining methods include use of fluorophore-labeled (immunofluorescence) and enzyme-labeled (immunoperoxidase) antibodies to identify proteins and other molecules in cells. In diagnostic surgical pathology, immunoperoxidase methods are widely used to extract additional information that is not available by H & E staining and light microscopy or by transmission electron-microscopy. The past half century has seen tremendous refinement in the development of the antibody tools and the detection systems. However, the overwhelming majority of this effort has been directed at further analysis of protein expression as a marker of a particular cell, tissue or tumour type. Newer biomolecules which have a role in prognostication or which form the basis of justification of expensive targeted therapy have increased the demands from surgical pathology services.

**Immunocytochemistry** (**ICC**) refers to immunostaining of cultured cell lines or primary cells including smears, swabs, and aspirates. ICC offers a semi-quantitative means of analyzing the relative abundance, conformation, and subcellular localization of target antigens.

Traditional ICC techniques use chromogenic detection in which enzyme conjugated antibodies convert chromogen substrates to a coloured precipitate at the reaction site. However chromogenic detection has lost favour with the advent of immunofluorescent labels.

In immunocytochemistry/immunofluorescence (ICC/IF) assays, the cellular antigens are visualized using either fluorochrome-conjugated primary antibodies (direct detection) or a two-step method (indirect detection) involving an un-labelled primary antibody followed by a fluorochrome-conjugated secondary antibody. By combining different fluorochrome-labelled antibodies, multiplex ICC/IF can detect several antigens in the same sample.

While ICC is often used interchangeably with ICC/IF (immunocytochemistry/ immunofluorescence) and another related term, IF, significant differences exist between them in reference to the method of detection and starting sample types involved.

**Principle**: Immunocytochemistry is a technique used to assess the presence of a specific protein or antigen in cells (cultured cells, cell suspensions) by use of a specific antibody, which binds to it, thereby allowing visualization and examination under a microscope.

Immunocytochemistry harnesses the power of antibodies to give highly speciﬁc binding to unique sequences of amino acids in proteins.

**Application of ICC**: ICC is a powerful, versatile tool for furthering our understanding of the expression, localization and interactions of proteins at the cellular level. This power has only been augmented by more recent developments in molecular biology, where insights into the behaviour of proteins in live cells, or interactions on the atomic scale can now be investigated.

1. **Embalming** in most modern cultures is the art and science of temporarily preserving human remains to forestall decomposition and make it suitable for display at a funeral. Human embalming started in Egypt about 3,000 (three thousand) years ago when the Egyptian noticed the preservation and mummification which took place when they left their human bodies in the desert. The two methods of preserving human cadavers are: the natural method and non-natural method. The ingredients that can be used for making up human embalming fluids are: formalin, phenol, methylated spirit, glycerin and water. Certain factors like temperature, access by insect and access by carnivores can affect an embalmed body. Measures to follow when carrying out human embalming are to always wear laboratory coat, hand (surgical) gloves and rain boots. It has also been recommended that human embalming should be carried out in a place that is equipped with an extractor fan or a well-ventilated area.

**Plastination** is a non-natural method of Embalming. Plastination is a technique or process used in anatomy to preserve bodies or body parts, first developed by Gunther von Hagens in 1977. The water and fat are replaced by certain plastics, yielding specimens that can be touched, do not smell or decay, and even retain most properties of theoriginalsample. In November 1979, Gunther von Hagens applied for a German patent, proposing the idea of preserving animal and vegetable tissues permanently by synthetic resin impregnation. Since then, von Hagens has applied for further US patents regarding work on preserving biological tissues with polymers. With the success of his patents, von Hagens went on to form the Institute for Plastination in Heidelberg, Germany in 1993. The Institute of Plastination, along with von Hagens made their first showing of plastinated bodies in Japan in 1995, which drew more than three million visitors. The Institute maintains three international centres of plastination : in Germany, Kyrgyzstan and China. There are four steps in the standard process of plastination: fixation, dehydration, forced impregnation in a vacuum, and hardening