

Role of Various Artifacts in Histopathology : A Narrative Review

Abstract:

The noteworthiness of appropriate handling of tissue samples is valuable for histopathological diagnosis. Preciseness of tissue sample diagnosis by pathologists is profoundly reliant on the proficiency of histotechnologists. Artifacts are known as fault or glitches that happens at several stages like collection of sample, during processing, embedding, and mounting of the tissue sample. Thus, it is crucial to determine the frequently occurring artifacts. In this review, literature or information on artifacts is discussed. English language articles were scrutinize in plentiful directory or databases like Pubmed, Scopus, Web of Science, Science Direct, Google Scholar.

Key-words: Artifacts, Diagnosis, Histopathology, Tissue sample

Introduction:

Acc. to Bernstein, Artifact is defined as “an artificial structure or any tissue alteration on a prepared microscopic slide produced by some extraneous factors”[1]. The goal of a desirable histopathological procedure is to provide good microscopic preparedness of tissue samples, but sometimes it is not achievable and tissue distortion is seen[2]. It has been found that some artifacts are efficiently observable from diseases or normal tissue samples whilst some are challenging. The existence of artifacts can result in imprecise diagnosis. Artifacts may consequence in the diversification of normal morphologic features. Therefore understanding various types of artifacts can prohibit misdiagnosis[3,4].

VARIOUS ARTIFACTS IN HISTOPATHOLOGY

1. Formalin pigments:

Formaldehyde has a characteristic predilection to be oxidized, resulting in the production of formic acid. Heme and formaline adhere together to make formalin-heme aggregation. This aggregation in tissue sections emerge as brown-black granules[5].

2. Mercury pigments :

Those fixatives that contains mercuric chloride give rise to granular dark brown deposits[5]

3. Starch artifact :

One of the most frequent impurities of histological and cytological samples is starch powder. In h & e stained region, they present as multiple blue, small structure. In oral cytological smear, they present as refractile, polygonal bodies

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with y-shaped configuration that may be misdiagnosed as pyknotic nuclei. They are PAS positive. In polarized light microscopy, they show “maltese cross” pattern[6].

4. Streaming artifact :

It occurs because of the dissemination of unfixed material to give inaccurate localization because of the placement of material other than its primary location. This type of artifact is most repeatedly observed in formalin-fixed tissue. Streaming artifacts can be abominated by practicing glycogen fixatives[7].

5. Freezing artifact :

This type of artifact emerges as Swiss cheese void in epithelium and tissue zone. They serve as the locations where ice crystals breach the tissue[4].

6. Orientation artifact:

Inappropriate orientation will result in poor arrangement of histological components in slides. For proper orientation of skin specimens, it must be placed in a manner so that the subcutaneous tissue, epithelial edges, and deeper layers are properly oriented in the slides[5].

7. Chatter artifact:

Chatters are defined as thin and thick zones alongside the knife margin. Chatters are most often caused by disproportionate elevated knife angle, extreme brittleness and hardness of the block, and minute vibrations at the edge of the knife[1].

8. Sponge artefacts:

These type of artifacts are observed in places where tissues are placed between the sponges in the cassettes. The intensity of sponge perforation is linked to tissue texture and is reliant on the stromal matrix. For illustration, thin samples ordinarily show larger perforation of sponge compared to harder tissue[9,10].

9. Compression artifact :

This type of artifact was seen most commonly while using a blunt knife or when the knife bevel is too large. Use of blunt knife results in dislocation of tissue sample specially bone[11].

10. Alternate thick and thin sections :

This type of artifact occurs when the blade or block is unconstrained, the wax is much soft for tissue, the microtome is defective, and the clearance angle is deficient. To avoid this artifact, always use ice to cool the block, and hike the clearance angle[11].

11. Residual wax:

Forgetting to eliminate the wax from the tissue section earlier to staining will outcome in insufficient or limited staining in that region. This leads to pink disease artifacts. Nedzel et al observed the existence of intranuclear birefringent inclusions, mainly in lymphocytes[12].

12. Artifact due to mordent of hematoxylin:

In various hematoxylin solutions, the most commonly used mordent is aluminium potassium sulfate. During staining, if the hematoxylin solution is not blended accurately, aluminium potassium sulfate is altered into crystalline forms which leads to the disintegration of the solution and black color pigment will be observed all over section[13].

13. Residual water and air bubbles :

When the mounting medium is very thin, air bubbles are produced beneath the cover slip. When it dries up, extra air gets entrapped beneath the edges. To avoid this, mounting medium having sufficient thickness is used[8].

14. Suture artifact:

The existence of suture material in tissue can impair the microtome knives which results in the tearing of tissue section[15].

15. Artifact related to excessive use of mounting media :

Foggy appearance will be observed when more amount of mounting media is used. This type of artifact can be avoided by using sufficient quantity of mounting media having adequate consistency[7].

16. Fulguration artifact :

This type of artifact is observed when laser or electrosurgical cutting of tissue is done. This cutting results in an area of tissue deformity and thermal necrosis. Because of the protein

coagulation, amorphous appearance is demonstrated by epithelium and connective tissue[8].

17. Bone dust artifacts :

Dust is formed when undecalcified bone sections are cut. Some of these bone particles may be included in much deeper in the sections. These particle stains actively with hematoxylin upon h & e staining. In undecalcified sections, this results in a fallacious diagnosis of metastatic calcification[15].

18. Improper embedding :

Because of the improper embedding, tissue becomes fragile and cutting of such tissues will result in cracks[16].

19. Prolonged floating of sections on the water bath :

If tissue is floating in a water bath for a long time, the tissue may expand further their initial size and become deformed. The epithelium shows acantholytic presentation imitative oedema[7].

20. Contamination :

With dust, and remaining cells of the earlier tissue section, the water bath becomes contaminated. It is suggested that repeated cleaning of water bath should be done. Also, use of distilled water is recommended in place of tap water[7].

21. Squeeze artifact :

This type of artifact occurs when tissue is compressed by various surgical instruments. It consists of splits, hemorrhage, pseudo-cyst, fragmentation and crush. On viewing microscopically, the tissue with crush artifacts shows distorted cellular and nucleus details[3,4,17].

22. Microwave fixation artifact:

45° c–55°c is the most appropriate temperature for microwave fixation. Overheating results in pyknotic nuclei and brings out vacuolation, while underheating shows substandard tissue sectioning properties[7].

23. Improper dehydration:

A great degree of tissue shrinkage is seen when tissue is placed in higher aggregation of alcohol. It is seen that when tissue is not dehydrated entirely, paraffin will not intrude into

the tissue adequately. This results in strenuous cutting of tissue that leads to tearing of the tissue. When this condition happens, it is advisable to rehydrate the section of the tissue and again perform the processing[11,16].

24. Artifacts due to poor processing:

Because of the inadequate or poor processing of the tissue, considerable deficit architectural details occur. Reasons for poor processing of tissue include wrong selection of reagents, altered processing cycle, and use of debilitated reagents[16].

25. Folds and wrinkles in section :

This type of artifact is seen when delicate paraffin sections are unwillingly expanded haphazardly around another structure. This type of artifact emerged as darkly stained strands. Folds or wrinkles in the tissue section can be extirpated by expanding and soft tapping with the help of forcep. During these measures, sometimes overstretching may result in tissue section tearing which shows an acantholytic image in epithelium[7,11].

26. Improper clearing :

Tissue becomes brittle when it is placed in xylene for an extended time. It results in the disintegration of tissue during cutting. Contrarily, if the tissue sample is not cleaned accordingly in xylene, paraffin will not enter into the tissue perfectly and results in tissue mutilation at the time of sectioning[11].

27. Improper prefixation :

Sometimes tissue will go through autolysis because solution like normal saline is not able to attach the tissue. Microscopically, such tissues display epithelium and connection tissue segregation and also hallmark of autolysis artifact[18].

Conclusion:

Artifacts are experienced in many histopathologic tissue sections which perform character in the explanation of diagnosis. Adequate processing of tissue sample outcomes in tissue samples appropriate for diagnosis. Identification of various artifacts and conquering these artifacts is the greatest question in the laboratory. Appropriate management of samples and eluding the faulty approach will scale down the artifact and help in making diagnosis better and more accurate.

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