

Understanding necropsy and histologic artifacts

Pathologic artifacts are any structures or features that are not normally present in the living animal. Some are minor, easy to distinguish from normal or diseased tissue, and/or do not interfere with the pathologists' ability to provide an accurate diagnosis. However, in some cases, the degree of artifactual damage can be severe or can involve the entire specimen, rendering the sample useless for diagnostic purposes. The aim of this document is to provide a guide for understanding some of the more common artifacts encountered in veterinary pathology and to give suggestions to help minimize their occurrence.

Autolysis

Autolysis and putrefaction result in the rapid degradation and distortion of the tissues, hampering interpretation of postmortem findings. Factors that can increase the severity of autolysis include, but are not limited to, heat, a longer time interval from the moment of death to the time of tissue collection, exposure to the elements, delayed body or tissue cooling, and the type of disease process (e.g., intestinal rupture with exposure of internal organs to gastric enzymes). Autolysis can be minimized by either placing a sample directly into formalin at the time of tissue collection or, if the animal has passed, trying to cool the body as quickly as possible by either placing them in a refrigerator, cooler, or if that is not possible, a shaded area out of direct sunlight covered with ice. As a general rule, the larger the animal, the slower it takes for the internal body temperature to fall, thus increasing the likelihood of autolysis.

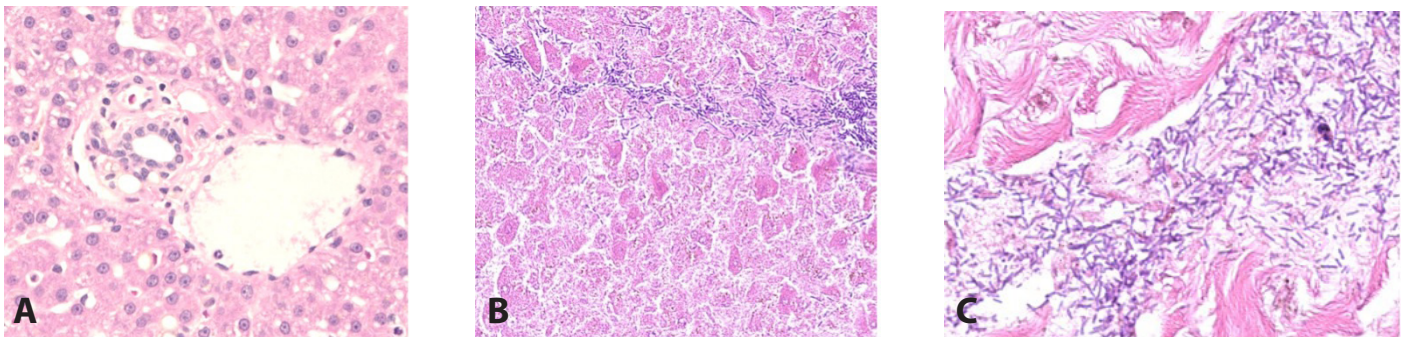


Image 1: A) Normal liver showing good cellular detail with nuclei, vessels, and a bile duct. B) Marked autolysis of the liver with loss of cellular detail and overgrowth of post-mortem bacteria. C) Close up of the bacteria (arrow) within the periportal collagen.

Cautery artifact

Electrocautery is used in surgery to stop bleeding by heating the tissue and clotting blood. When examined histologically, cautery artifact can cause cells to look smudgy with increased eosinophilia (bright pink) and nuclei to look stretch out and be darker compared to normal cells. This is often most prominent at the surgical margins of mass removal sites, which can interfere with determining tissue margins for tumor borders.

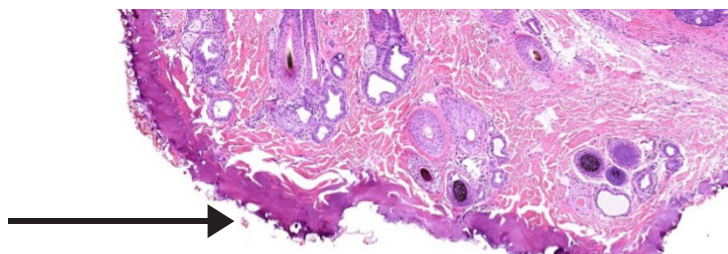


Image 2: The arrow points to the bright eosinophilic area caused by the cautery.

Freezing artifact

When a body or tissue samples are frozen, there is formation of ice crystals. Even if the tissue is placed in formalin, ice crystal gaps or clefts will remain. The ice crystals cause architectural distortion of tissue, hampering diagnostic quality. Once thawed, the nuclei in cells can appear smaller with condensed darker-staining nuclei. The nuclear and cytoplasmic detail is also not as well defined. Cellular color can be affected as well making detection of certain pathologic lesions challenging. Ideally, samples should be placed in a refrigerator if they are unable to be shipped immediately. Once shipped, send either in sealed formalin-filled containers or if fresh, with multiple cold packages to keep chilled. To avoid tissue freezing artifact in very cold environments, an insulated shipping container should be used or isopropyl alcohol added to the formalin fixative (1 part alcohol to 10 parts formalin).

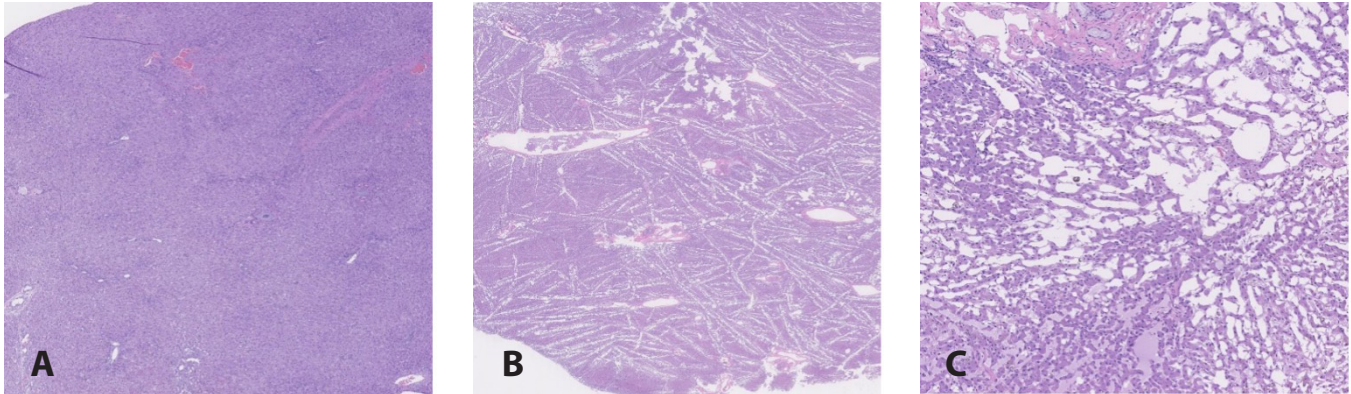


Image 3: A) Normal liver. B) Liver that was frozen. C) Close up of liver B. The multiple clear spaces are artifact from ice crystal formation.

Crush artifact

A crush artifact is a false distortion of tissue caused by iatrogenic compression of a tissue. Tissue crushing is usually the result of firm handling with forceps, though other instruments and fingers can also cause crushing damage. Distortion can be caused by the slightest compression of tissue and can provide difficulties in diagnosis. Hypercellular tissues are more susceptible to crush artifact as they are more fragile, and crushing can cause elongation of cellular nuclei, inhibiting cellular identification. Crush artifact is most problematic with samples that are tiny, such as with endoscopic biopsy sections, where the crush artifact can completely hinder histologic examination. In larger samples, crush artifact may interfere with margin evaluation. Crush artifact can be minimized by removing a sample from a trocar or needle biopsy by using the tip of another needle to slide the sample into the formalin jar instead of picking up the sample with a forceps, or for larger samples, by handling the samples more gently.

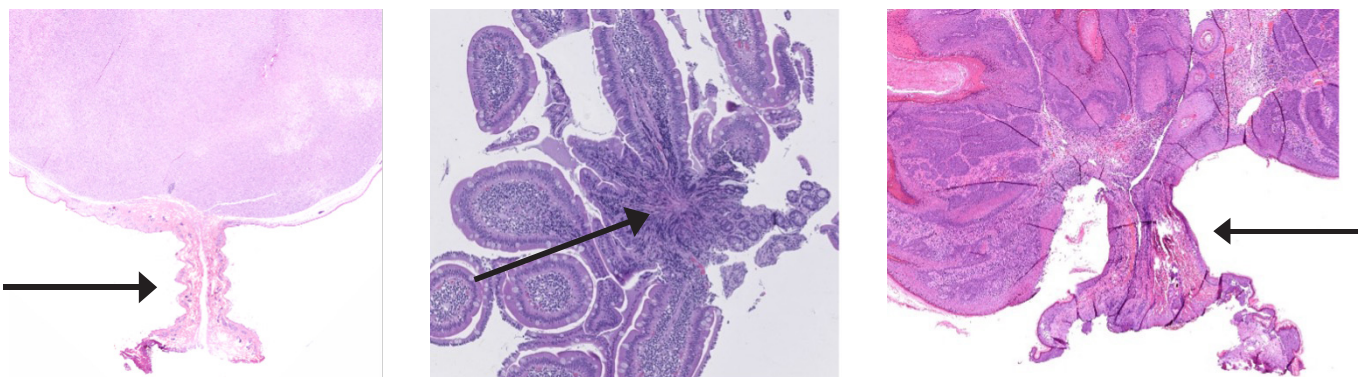


Image 4: Arrows point to the sites of crush. In some sections (A) you can make out the teeth imprint of forceps.

Sponge artifact

When pieces of individual sponge barbs become entrapped in tissue sections or they produce clear triangular impressions within tissue during processing, this is known as sponge artifact. Generally, this is of little importance. However, in certain instances such as endoscopic biopsies of the intestinal tract, it can obscure important information. Sponge artifact can be avoided by ensuring the sponge is pre-moistened with formalin before placing the specimens on the sponge. Gauze sponges are not recommended. Lens paper wrapping of samples within a cassette can also be utilized.

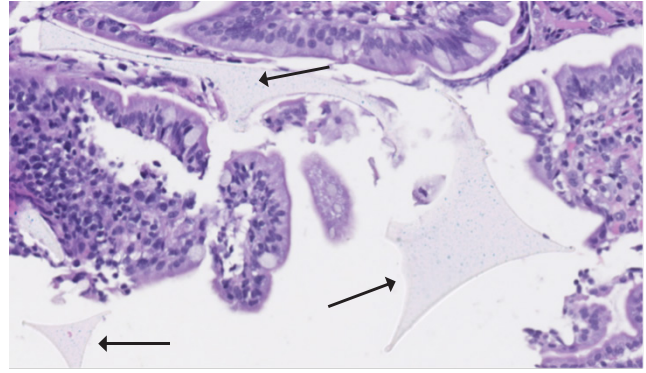
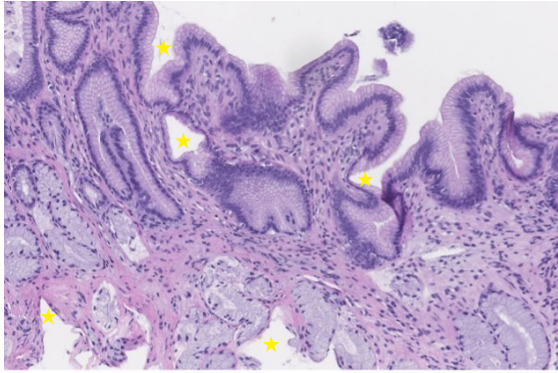


Image 5: Stars denote triangular spaces left behind by sponge. Arrows denote sponge material adhered to the tissue and carried through in processing.

Hopefully, this document aids in the understanding of tissue artifact. At times artifacts are unavoidable but they can be minimized. Should you have any questions on specimen preparation, collection, or tissue shipment, please contact us at the laboratory at 979-845-3414 for assistance.

- Kamstock DE, Ehrhart E, Getzy D, et. al. (2011). Recommended guidelines for submission, trimming, margin evaluation, and reporting of tumor biopsy specimens in veterinary surgical pathology. Vet Pathol. 48. 19-31. 10.1177/0300985810389316.