DOUGLASS HANLY MOIR PATHOLOGY

Quality is in our DNA

Effects of varying thickness, processing and fixation time on formalin-fixed surgical specimens

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Introduction

Proper fixation and processing time are vital in diagnosing surgical specimens from small biopsies, fatty tissue, fibrous tissue, skin, mucosal tissues, bone and others. This poster demonstrates the importance of proper fixation and processing time with fatty tissue (breast) and non-fatty tissue (fallopian tube).

Method and materials

Breast

- Fatty breast tissue is fixed with 10% neutral buffered formalin.
- Varying thicknesses of the breast tissue are cut and are processed on the 12-hour processing run.
- Processed fatty breast tissues are embedded and standard 3µm H&E section were performed on the tissues.

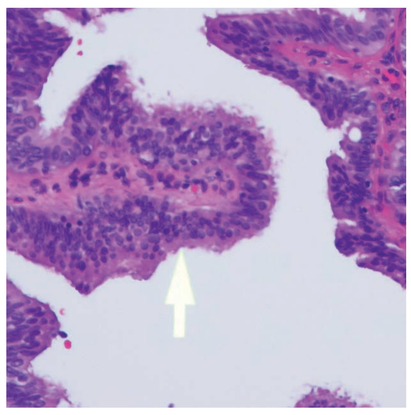
Fallopian tube

- Fallopian tube is fixed with 10% neutral buffered formalin.
- Sections of the fallopian tube are taken and are processed on the 1-hour, 6-hour and 12-hour processing run.
- Processed fallopian tube tissues are embedded and standard 3µm H&E section were performed on the tissues.

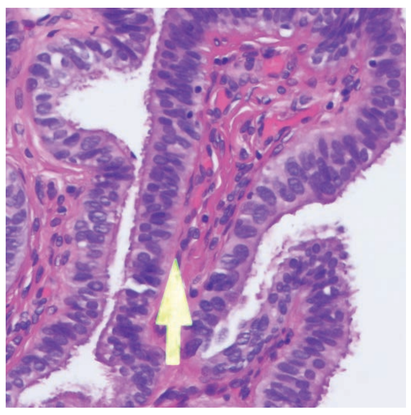
For both tissues, 10% neutral buffered formalin supplied from Muraben lab was used for fixation, routine processing time is performed according to the Leica Peloris rapid tissue processor machine, Leica Paraplast surgical wax was used for embedding the tissues and routine H&E is performed according to the Olympus Tissue-Tek Prisma staining machine.

Fallopian tube.

Cross-section of fallopian tube.



1-hour processing run; 200x magnification.



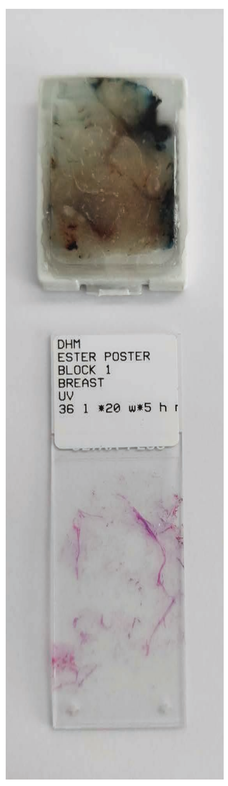
6-hour processing run; 200x magnification.

Discussion

Proper fixation (with the best fixation reagents, optimal time and with the specimen sliced to allow penetration of reagent) and optimal processing by quality machines will give the best sections for histologic diagnosis.
If the tissue is not adequately fixed or processed the following problems will occur:

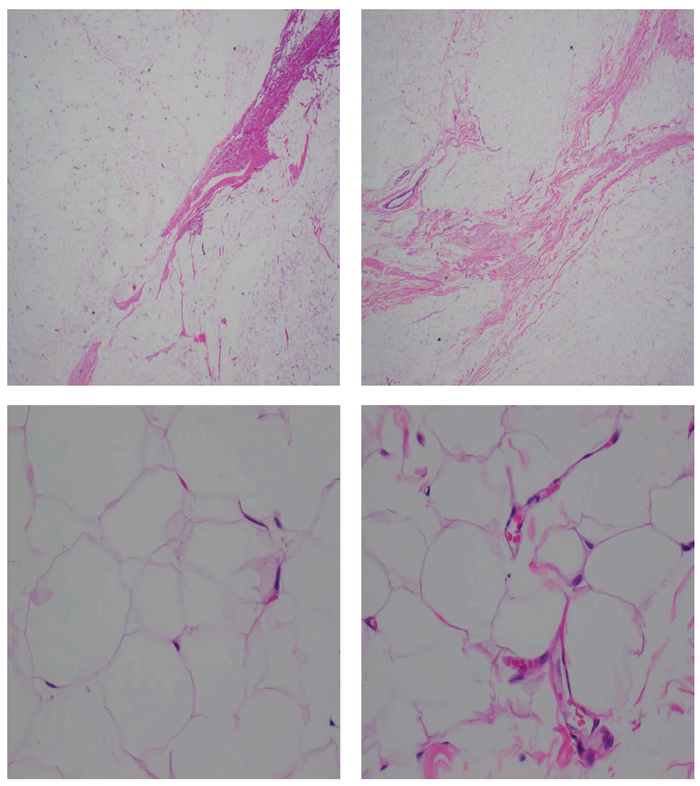
- Sectioning with microtome will give scarring, distorted and disrupted sections.
- H&E staining will not provide nuclear and cytoplasmic details.
- Special stains will not stain appropriate tissues.
- IHC staining will give either false positive or negative results.

Thick fatty sections tend to be more under processed compared to smaller surgical tissues due to the lack of fixation and processing time given. Multiple text books and other literature have repeatedly claimed the importance of fixation and processing. However, we have repeatedly used the above exercise to optimise surgical specimens in fixation and processing to give the best sections possible for histologic diagnosis, which is vital for our patients.



Fatty breast tissue of 5 mm thickness.

Fatty breast tissue of 1 mm thickness.



High and low magnification histological stained fatty breast tissue appearance for 5mm and 2mm thickness.

Conclusion

Breast

The best section is the 1 mm thickness, which fits exactly in the mould. The details of peripherally placed nuclei, complete cell membranes and fibrous bands are best seen.

Fallopian tube

The 6-hour run provides a clearer microscopic view of cell details, such as cilia from the cuboidal to short columnar cells.

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