# Purpose

Describes procedures relating to tissue embedding.

# Scope

Applies to all histopathology staff undertaking paraffin embedding.

# Responsibilities

It is the responsibility of all staff in the histology laboratory to follow procedures described in this document. It is the responsibility of the histology Section Supervisor and Shift Supervisor to ensure procedures are followed.

# Definitions

## N/A

# Procedure

Paraffin impregnated tissue are orientated in a base mould, overlain with molten paraffin wax and cooled to create a tissue block ready for microtomy. It is essential accurate specimen identification is maintained throughout the embedding process.

## General Instructions

* Scheduling of tissue processing programmes must occur to ensure completion of processing coincides with commencement of the embedding shift. Delays in embedding may cause serious damage to specimens. Small biopsies, held in molten wax for extended periods beyond processor finish times can become brittle and difficult to section.
* Adjust processing start and finish times accordingly if delays to embedding are anticipated. For day-run and urgent blocks, thought must be given to the program end time to ensure someone is available to embed. Always alert the histology supervisor of short run processing so staff are available to embed the blocks processed.
* Where extended delays are unavoidable, (eg post mortem or research blocks), it is preferable to allow cassettes to harden at room temperature rather than incubating at 60C in wax for long periods. Cassettes, if allowed to solidify, can be rewarmed and embedded at a more convenient time.
* Biopsy embedding is to commence at the time of the first embed roster (generally 6.00 am).
* Cassettes are removed from the processing machine and transferred to the holding tray of the embedding centre.
* When transferring cassettes, processing baskets **must** be placed into a plastic transfer container to ensure wax does not drip onto floor surfaces or seating. Wax drips are a serious slip hazard.
* The temperature of the warm stage and wax reservoir are thermostatically controlled. These must be carefully set to avoid tissue damage. Where the temperature differential between the wax and the cold stage is too high, the wax will crack during solidification, disrupting tissue and causing artefacts in sections.
	+ - Warm stage 65oC +/- 3oC
		- Wax reservoir 60oC +/- 2oC
* Histologists must practice to be able to rapidly identify and orient specimens during paraffin embedding. Taking too long to manipulate and position the specimen may cause more than one layer of solidified paraffin to form when molten paraffin is added to the layer of paraffin that has solidified in the bottom of the base mould. These separate layers may pull apart during microtomy.
* Pieces of tissue should be placed with intention, in one plane, and not just randomly in the block face. Careless positioning will make it very unlikely that a single representative section can be easily obtained.
* Correct by re-embedding, any gross defects in a block such as cracks or air bubbles before allocating for sectioning
* Observe the condition of the tissue whilst embedding, being alert to issues with tissue processing, such as poor fixation, inadequate dehydration, clearing or wax infiltration.
* A "white-ish" appearing or "mushy" texture due to inadequate dehydration in a processed tissue block is most likely in need of reprocessing. Submit this block for reprocessing prior to embedding; it is much easier to reprocess a block prior to embedding than to reprocess from the final cut block.
* Document and correct labelling discrepancies, illegible blocks, or incompleteness of cases during embedding, rather than "passing" these problems on to the microtomy step.

## Identification of Embedding Staff

It is an essential part of Quality Assurance that the identity of the individual responsible for embedding a block is traceable. This is a NATA requirement.

***Place a paper label bearing the name or initial of the embedder into the back of each cassette to identify the person responsible for embedding each block.***

The names of staff participating in the embed 1 and embed 2 roster should also be entered onto the *Daily QC Check Form* (CD\_AP\_FA\_0030) as an easily accessible record of staff responsible.

## Relevance of Cassette Colours

Different coloured cassettes indicate specific processing or protocol requirements.

|  |  |  |
| --- | --- | --- |
| **Colour** | **Tissue** | **Protocol Requirement** |
| Lilac | Urgent Bx various tissue types | Urgent handling. Priority process, cut, stain and distribute |
| Lilac | Various special tissues. eg liver core, tumour core  | Special or altered protocol. Stop and check. A case requiring a specific or altered protocol to routine (reduced levels, preservation for molecular, etc). Check before cutting |
| White | Routine Tissue all types | Routine handling, decalcification for calcified tissue |

|  |  |  |
| --- | --- | --- |
| **Colour** | **Tissue** | **Protocol Requirement** |
| Pink | Various | Require levels. Also SLN for Breast Ca |
| Grey | Various | Require special stains |
| Orange | CNS, Kelly Derm,VNLSCytology Cell Block | Cases for Head of UnitCases for Head of Unit/Neuromuscular pathologistCases for Cytology |
| Tan | Various | Additional blocks for an existing case. Routine handling |
| Blue | SLN- melanoma | Sentinel Nodes for melanoma protocol |
| Blue | BMT | Bone Marrow Trephine for decalcification |
| Blue  | Frozen Section | Routine processing  |
| Green | All Autopsy tissue | Routine autopsy tissue |
| Yellow | Various | Tissue for Controls and Research |

## Embedding Priorities

In order to provide an efficient staining and reporting workflow embed cassettes according to the following priority

* Urgent – any case/block identified as Urgent. Includes transplants (Cardiac, lung, renal), renal cores, GVHD skin, paraffin of frozen.
* NPV (smalls) - Single block NPV cases. Prioritised to assist delivery to NPV pathologist (approx. 20 cases / 4 trays)
* Specials (Grey) – Cases requiring special stains. Prioritised for staining workflow.
* Smalls (Pink) – Cases requiring levels
* Smalls (White) – Single block cases
* Medium (White) – Two to Four block cases
* More-on (Tan) – Additional blocks from previous cases
* Large (White) – Multiple block cases.
* Autopsy
* Controls
* Research

## Tissue Orientation - General Rules

* **Know the proper embedding for each tissue**. Incorrect placement of tissues may result in diagnostically important tissue elements missed or damaged during microtomy.
* **Do not rush or become distracted**. Embedding requires thought and concentration. The importance of the embedding step to the diagnostic process must never be underestimated.
* **Place tissue centrally in the base mould**. Arrange so the long axis of the specimen lies parallel with the long axis of the mould and therefore, parallel to the knife edge during cutting. If the long axis is placed perpendicular to the knife the tissue has a tendency to compress and appear distorted.
* **Orientate large dissected specimens as they are placed in the cassette.** Place in the mould cut side down. Example; tonsil, ovary, breast thyroid, placenta, uterus, lipoma, etc.
* **Cut surface:** embed on cut surface of tissue. Example; skin excision
* **Arrange multiple tissue pieces in the block to permit maximum representation on the cut slide.** If two or more specimens are placed in one mould, the tissues should be in contact with one another. If there is paraffin between specimens embedded together the tissue may pucker during sectioning. This puckering can promote wrinkles to form during drying.
* **Arrange multiple tissue pieces across the long axis of the mould. Do not place at random. Maintain any obvious sequence.**
* **Do not layer tissue pieces at different depths in the mould. All pieces must be embedded flat against the base of the mould for appropriate visualisation and sectioning by the microtomist.**
* **Segmental sections of tubular structures:** embed on cut end of segment. Example; fallopian tube, appendix, umbilical cord, vas deferens.
* **Segmental sections of large lumen organs or tissue:** embed on the cut surface to demonstrate the lining. Example; intestines, gallbladder, cyst wall.
* **Orientate to allow for hard and tough surfaces** (eg. capsules of organs, cornified epithelium of skin etc.) should be embedded so they are cut last and do not drag through the adjacent soft tissue.
* Use a magnifying glass if necessary to assist in orientating small fragments or identifying tissue surfaces.

## Tissue Orientation - Specific Rules

* + **Vessels:** embedded on end. Example; temporal artery
	+ **Skin punch (bisected):** embed on the cut edge
	+ **Skin segments:** embed diagonally, facing in such a way that the microtome blade cuts through the fat and dermis first and the epidermis last. Use consistency when orientating and aligning skin sample, .i.e. with the epidermis facing in the same direction in all skin sections.
	+ **Skin shave:** embed on side
	+ **Skin for Alopecia:** requires dissection at embedding and specific orientation that will vary with the clinical presentation. Alopecia samples must only be dissected and embedded following instruction from a pathologist and must only be embedded by senior staff. Refer to the specific Alopecia protocol for further details.
	+ **Cores:** single cores should be orientated parallel across the mould or at a slight angle. Multiple cores should be placed in parallel rows to each other. Ensure all cores are sitting at the same level flat against the base. Use a tamper with gently pressure if required.
	+ **Endoscopic biopsies:** are always small and difficult to orientate. Look for a surface and embed on edge. Embed multiple biopsies in a straight line across the mould to assist with multiple sections to fit on a slide. Gastrointestinal biopsies often curl into a ‘C’ shape during fixation. Embed these on their side.
	+ **Curettage biopsy**: embed fragments longitudinally to show as much tissue as possible. Example; uterine curetting’s, prostate chips
	+ **Polyps:** embed on the long axis to show plane of cut section.
	+ **Placenta:** embed membrane rolls on edge perpendicular to the block face to show membrane layers. Embed cord segments on end to show vessel openings.
	+ **Muscle biopsies**: are sectioned in both transverse and longitudinal planes (embedded by VNLS staff)

Where a particular feature is present on one surface only, this must be embedded face down.

* + **Nerve biopsies:** may be presented as 1, 2 or 3 pieces some of which may be osmicated.
* 1 piece (non-osmicated **or** osmicated). These are embedded on end (TS orientation) **▌**
* 2 non-osmicated pieces. Embed one piece on end (TS) the other flat down (LS) ▌and **▬**
* 2 pieces, one non-osmicated, the other osmicated. Embed both on end (TS) ▌▌

## Preventing Cross Contamination

Good practices and careful attention will prevent cross contamination between blocks from occurring during the embedding step. Staff must remain alert at all times during the embedding process for the potential for tissue to be displaced, adhere to surfaces and transferred between cases inadvertently. To eliminate the potential for cross contamination always;

* Open only **ONE** cassette at a time.
* Open lids, biopsy pads, mesh bags, and lens papers carefully to avoid specimen fragments from "pinging" outward and potentially being lost.
* Wipe forceps between each specimen. Be especially careful with forceps bearing small "teeth" or grooves at the tips.
* Regularly clean the heated wells of embedding centre that holds forceps to avoid cross contamination from any foreign material.
* Ensure non-disposable base moulds are clean before use or re-use. Clean with solvent or detergent periodically.
* Do not use a base mould that has any residual tissue fragments or wax fragments in the base. Inspect each base mould before adding wax to ensure it is clean and suitable for use. Place unsuitable base moulds aside and ensure cleaning occurs before reuse.
* Embed any pieces that appear to "not belong" or to be contamination from cut-up into a far corner of the block face away from what you believe to be the intended specimen.

This communicates that you think it "does not belong" and makes it easy to identify for removal from the block if determined that it is a contamination.

Use a black dot label to flag the block for review and ensure to communicate the issue to a pathologist.

* Ensure fragments of tissue are not left to contaminate the embedding instruments hot work surfaces or cassette holding tray. Any residual fragments produced from control block assembly must be removed from the work surface, embedding moulds or holding tray once blocks are embedded. Do not leave open moulds in the holding tray unattended. Be mindful of the potential for moulds to flip and spill contents into the holding tray.
* Deal with any unidentified tissue found on the embedding centre according to the procedure outlined in 5.8.5

## Embedding Procedures

* Be alert to cassettes requiring special attention or specific tissue orientation. Use cassette colour, notations, inking or written instructions to guide embedding.
* Remove the cassette from the holding module / transfer tray and place on the heated working stage.
* Gently open the process cover from the cassette. Check the inner surface of the cover for any adherent tissue before removing and placing aside on the heated work surface.
* Never open more than one cassette at any time
* If biopsy pads are present, peel back the top pad gently. Check the inner surface for adherent tissue. Place the pad aside on the heated work surface.
* Examine the specimen contained within the cassette. Note any tissue ink, cut surfaces, lumen, or layers that may be present. If you do not recognise the specimen type, check the macro description to verify the tissue type or other important information. If the tissue is unfamiliar or confusing, always seek further information in order to orient the specimen correctly. It is better there is a delay in a block being cut than a diagnosis being compromised by poor orientation or vital tissue being lost at microtomy.
* Select a base mould from the thermal console. The size chosen must allow a 1-2mm (minimum) margin around the embedded specimen(s).
* Note: Visually check the base mould is clean. If any residual tissue or contamination exists do not use. Place aside for cleaning.
* Fill the base of the mould with molten wax.
* Using clean warm forceps place the tissue into the mould. Quickly orientate the tissue into the base mould, taking care to centre and correctly align the entire tissue sample. See section 5.3 for specific orientation requirements.

 Notes:

* + Use minimal pressure to avoid damaging the specimen.
	+ Use a tamper for larger pieces or irregular cut surfaces.
	+ Warm forceps in the Bacti-Cinerator, however do not overheat as hot forceps will burn tissue and produce artefacts.
	+ The underside of the base mould may be briefly touched to the cold plate to create a semi-solid wax base to assist positioning and to hold tissue to the bottom surface of the mould. Tissue must be placed parallel to the base of the mould
	+ If orientation or placement is in any way unsatisfactory, remelt and start again. Near enough is never good enough
* Validate the tissue cassette contains the number of pieces documented on the side of the cassette. Refer to section 5 if discrepancies encountered.
* Slide the mould from the heated working area to the "Cold Spot". The wax on the base immediately solidifies holding the specimen in position.
* Place the embedding cassette onto the top of the base mould. Ensure to orientate the angled face of the cassette to the side which will maintain appropriate orientation when placed in the microtome chuck. Dispense additional molten wax into the mould through the holes in the base of the cassette. Fill the back of the cassette until the wax hole are covered.
* If biopsy pads are used. Retain the bottom biopsy pad in the back of the embedding cassette to aid in future investigation and as a signal to the microtomist that the block may contain a small specimen or requires special handling.
* Take care not to overfill with excessive wax. Moulds over-filled requiring scraping of the back and edges of the cassette prior to microtomy. Over-filled blocks may sit unevenly in the microtome chuck causing instability that may lead to the tissue becoming damaged during microtomy

* Slip an embedder identifier label into the back of the cassette. Each block must be accountable and identify who embedded it.

Notes: Coloured dot labels are used to flag specific requirements or alerts. Dot labels may be placed into a cassette at processing or originate at the point of embedding. Specific labels in use include;

* **Orange** dot label. Flags a Neuropathology case for **Prof McLean**
* **Yellow** dot label. Flags a block subject to **pinged tissue at embedding**
* **Green** dot label. Flags a block with a **tissue count discrepancy**
* **Red** dot label. Flags a block with **no tissue in the block**.
* **Blue** dot label. Flags a block with **an altered or non-routine microtomy protocol**.(e.g. core biopsy with altered levels, core biopsy for molecular, biopsy for research/biobank, etc). Also denotes **thin tissue**. A blue dot must prompt a **Stop and Check** before microtomy is undertaken.
* **Black** dot label. Flags a block with any other issue not covered by other labels (e.g. poor processing, staple, suspicious contaminant)
* Transfer the embedded specimen to the cold plate to solidify.
* Remove the cassette cover from the work surface and place into the holding basket.
* If a biopsy pad remains, remove from the work surface and place into a plastic holding container. Cap, date and retain holding containers for 48hrs.
* Ensure the work area is clean before proceeding to the next cassette. Do not allow any tissue residues to remain on the work surface. See section 5.6.5
* Heat forceps in Bacti-Cinerator to incinerate any contaminants. Wipe forceps tips to remove any burnt deposits.
* Repeat the embedding procedure with the next cassette.
* Allow the wax in the block to harden. After several minutes, lift the block from the base mould. **Never force a block from a mould as this may damage the tissue or leave fragments of tissue stuck to the base mould.**
* Place the base mould aside for cleaning before reuse.
* Scrape excess wax from the sides and/or face of the block.
* Inspect for gross defects, such as air bubbles or misaligned cassette back. Melt and re-embed if necessary.
* Sort and transfer blocks for microtomy.
* Always double check the processing basket for missed blocks before placing the basket and cassette lids into the processor for cleaning.

## Embedding Discrepancies & Problems

### **Communicating Embedding Issues**

It is essential to alert the section supervisor as soon as possible to problems or discrepancies encountered during embedding. Blocks missing tissue, blocks containing poorly orientated tissue, blocks contaminated with tissue from other sources or blocks incorrectly labelled can all have serious diagnostic implications.

It is essential pathologists are also informed of any situation that arises at embedding which has the potential to affect diagnosis. It is paramount that issues concerning patient identity (eg labelling, block duplication, etc) or specimen identity (pinged tissue, lost tissue, count discrepancy, block duplication, etc) are communicated to the pathologist prior to reporting.

Document and communicate embedding issues to the pathologist using the *Laboratory Technical Quality Feedback Form* (CD\_AP\_0327)*.* Include the form with slides arising from a problem block when presenting to a pathologist or when placing slides on the pick-up table.

The pathologist can also use this form to raise an issue with the laboratory where a discrepancy or technical issue occurs with a block or slide.

No matter who instigates the issue raised on the form, investigation, follow-up and sign-off of all *Laboratory Technical Quality Feedback Forms* is the responsibility of the histology supervisor for that shift.

### **Pinged, Flicked or Dropped Tissue**

It is essential staff performing embedding concentrate on their actions. Cassettes need gentle handling. Always open lids carefully, in a controlled manner and with close observation. Even for the most careful embedders, tissue can ping from a cassette or drop from forceps. You need to be alert to your actions and movements in case of such events to ensure tissue is not lost or contaminations introduced into the patient sample.

Undertake the following steps if a cassette is opened and tissue flicks out of the cassette or drops/pings from forceps.

* If a landing site is witnessed. Pick up the tissue and embed as required.
* If the landing site cannot be identified. Check the side of the cassette for notation on the number of pieces that should be present.
* Look carefully for the tissue piece on the embedding surface. Check the cassette lid. Check the sides of the cassette.
* Check the front of your gown, including sleeves. Check fingers, hands and wrists (do this before standing up). Check surrounding surfaces.
* If tissue is found. Examine closely to determine if it fits the macroscopic description. Pick up and embed as required.
* Place a **YELLOW** coloured dot in the back of the cassette to flag the cassette as subject to a “ping” event at embedding. The dot should have the initials of the supervisor written on it.
* If no tissue can be found **always** have a second person (or even a third) verify and help check for something you may have missed.
* If the tissue cannot be located in addition to the **Yellow** dot place either a **RED** coloured dot as a “No Tissue” flag (i.e the cassette now has no tissue in the block) or a **GREEN** coloured dot as a “Tissue Count Discrepancy” flag (i.e the cassette now has less tissue fragments than the side of the cassette and macro indicate).
* Document what happened via a *Laboratory Technical Quality Feedback Form.* Use this form to alert the reporting pathologist. The form must accompany any stained slides and/or the affected cassette for presentation to a pathologist. The reporting pathologist needs to be aware of the possibility of a specimen mismatch with pinged, flicked or dropped tissue irrespective of whether tissue is subsequently found and embedded in the block.

### **No Tissue in Cassette**

Undertake the following steps if a cassette appears to have no specimen is within the cassette on initial opening.

* Look carefully at the cassette lid (surfaces and hinge) and the interior corner and crevice of the cassette. If the specimen is on a biopsy pad or in lens paper, use a magnifying glass to check all surfaces. Use a clean, warm pair of angled or curved tip forceps to scrape the surface of pads or paper and transfer any particles to a prefilled mould.
* Check the side of the cassette for notation on the number of pieces that should be present.
* If no visible tissue is found, **always have a second person verify** and help check for something you may have missed.
* Check the macroscopic description. It may indicate the specimen was minute or of a nature that might not survive processing.
* If there is a possibility that the tissue flicked/pinged out the cassette also follow the steps outlined in section 5.7.2
* Retain biopsy pads or paper in the cassette.
* Place a **RED** coloured dot in the back of the cassette labelled “No Tissue” to flag the cassette as having no tissue in the block. The dot should also have the initials of the supervisor.
* Advise a pathologist to a missing specimen as soon as possible.
* Retrieve the original specimen container and check for tissue. If the original specimen was small and tissue remains in the pot it is possible tissue transfer was overlooked. If tissue was sampled from a larger specimen, a pathologist will determine if secondary sampling is appropriate.
* Document what happened via a *Laboratory Technical Quality Feedback Form.* Use this form to advise the reporting pathologist. The case will still require reporting as the requesting doctor and patient are expecting a result.

### **Tissue Count Discrepancy**

Undertake the following steps if a cassette is opened and the number of pieces notated on the side of the block does not match the number of pieces within.

* Check the cassette lid. Look carefully at the all surfaces including the hinge. If the specimen was submitted on a biopsy pad, recheck all surfaces of the pad. Re-check the cassette paying particular attention to interior corner and crevice of the cassette. Use a magnifying glass to assist.
* If no visible tissue is found **always** have a second person (or even a third) verify, help check for tiny fragments or something the embedder may have missed.
* Check the macroscopic description. It may indicate that the specimen was minute or of a nature that might not survive processing.
* Place a **GREEN** coloured dot in the back of the cassette to flag the cassette as subject to a count discrepancy. The dot should also have the initials of the supervisor.
* If there is a possibility that the tissue flicked/pinged out the cassette also follow the steps outlined in section 5.7.2
* Document what happened via a *Laboratory Technical Quality Feedback Form.* Use this form to alert the reporting pathologist. The form must accompany any stained slides arising from a problem block when presented to a pathologist or placed on the pick-up table. The reporting pathologist needs to be aware of the possibility that a block or slides may not represent the sampling that occurred

### **Unidentified Tissue (Found Objects)**

It is essential the embedding centre, including the holding tray, hot work surfaces, moulds, mould warming tray and wax hopper are kept clean at all times. Any residual fragments of tissue found need to be dealt with appropriately.

Fragments of blot clot may be discarded but other tissue fragments still belong to a patient and may be diagnostic. Tissue fragment(s) may represent an irreplaceable sample, explain a count discrepancy or assist with a diagnostic irregularity.

Undertake the following steps if a fragment of tissue is found in the embedding area.

* Notify the section supervisor
* Label a cassette with UT + Date + A (e.g UT-17/9/20-A). Any additional fragments on that day would go into a cassette labelled B, C etc
* Complete a Unidentified Tissue form
* Cut a section from the block for H&E
* Present the slide and UT form to the duty pathologist, who will review the slide and determine the next steps.
* File the block and slide together in the “Unidentified Tissue” box in the histology laboratory. Retain blocks and slides for a minimum of 3 years. File the form in the UT Log folder.

### **Under-processed Tissue**

It may be obvious at embedding that tissue is not adequately processed. Tissue may appear processed around edges but is soft and mushy in the central portion.

Inadequate processing can be a product of;

* Tissue cut too thick
* Processing times being inadequate for proper processing (Incorrect program selection).
* Poor quality of reagent (contamination, saturation or incorrect concentration)
* Reagent containers in the wrong position
* Processor fault or failure.

Where staff are aware of unprocessed tissue at embedding the following steps are followed.

* Do not embed the tissue. Replace the cassette lid and put aside.
* Alert the section supervisor who will decide on the appropriate next steps in consultation with the duty pathologist.
* If the tissue is excessively thick a pathologist will determine if slicing the tissue to make it thinner is an option or if reprocessing in its entirety on a longer cycle is required.
* If reagent quality is the issue, the supervisor will determine if all blocks need reprocessing or just those above a certain size.
* Perform reprocessing according to the procedure described in CD\_AP\_0027

# Maintenance of Embedding Centres

## Wax Levels

* At the completion of embedding check the wax level of the dispensing chamber. Replenish paraffin (should be 2 parts dry pellets to 1 part molten). With sufficient wax for the following day’s embedding. Maintain level at a minimum of half-filled. Take care to not overfill. Checking wax levels is the responsibility of the person embedding.

## Cleaning of Base Moulds

* Base moulds need to be free of residual wax and tissue debris before use and reuse.
* Check moulds at the block release step to ensure they are clean.
* Scrape away any excess wax from edges of the mould
* Stack moulds and place in the cassette rack.
* Cleaning embedding moulds in the tissue processor chamber during the purge cycle to allow the xylene to remove the paraffin is best avoided. Doing so increases the wax load on processor cleaning solvents, reducing cleaning effectiveness and increasing potential for instrument malfunction

## Cleaning of Cassette Lids

* Cassette lids need to be free of residual wax and tissue debris before use and reuse.
* Cassette lids are cleaned in the tissue processor chamber using the cleaning cycle as the xylene will remove paraffin residues from the surface. Make sure the surface of the lids is free of wax scum. Attention needs to be paid to the condition of the cleaning xylene and ethanol as heavily contaminated reagent will not clean adequately.
* Plastic cassette lids are not as durable as stainless steel lids and cannot be reused indefinitely.
* All lids must be sorted and checked for cleanliness before they are placed back into cut-up to eliminate the potential for cross contamination.

## Daily Cleaning of Embedding Units

* + The tissue embedding surfaces must be clean before commencing any embedding session. It is the responsibility of each person embedding to ensure the work area is clean and free of any residual tissue or potential contaminants before embedding starts.
	+ Clean embedding surfaces with paper towel or cloth wipes. Clean hot areas when the unit is on and the wax molten. Pay attention to grooves and crevices for tissue fragments and debris.
	+ Repeat the cleaning process at the end of the embedding session. Once again, this is the responsibility of the person embedding. Cleaning and removal of debris (including found object processing) must be performed at the end of each session.
	+ General cleaning of embedding units can occur towards the end of the day. Clean the external surfaces of the consoles to remove spilt wax. Scrape off wax using a plastic spatula. Empty wax collection trays and condensation traps. Remove condensation from cold plates and cold spots. Clean the heated wells of embedding centre that hold forceps to avoid cross contamination.
	+ Avoid the use of xylene to remove wax deposits. Xylene is a flammable liquid. Clean surfaces with a non-flammable agent such as Para-klean. Ensure switches are in the OFF position when cleaning around the display console.
* Complete the tissue embedder maintenance log ANA\_HPM\_002.

## Periodic Extended Cleaning of Embedding Units

* Periodically remove all wax from the wax chambers of the embedding centres and thoroughly clean inside of hoppers. Depressing the finger- switch dispenser control and allowing the liquid wax to run into a container does this. Remove the mesh filter and clean with xylene. Turn the "HEAT" to the OFF position and wipe the walls of the wax chambers with a clean dry cloth.
* Vacuum the air filter located on the back of the cryo-console.

# Operation of Embedding Centres

## Thermostat Adjustment

### Tissue-Tek TEC 6 Embedding Centre

The temperature adjustments for the wax chamber, working platform (hot plate), transfer trays and cryomodule are located as electronic controls on the front panel of the unit. Refer to the Operating Manual for the Tissue-Tek TEC 6 Embedding Centre for detailed instructions..

Note: The thermostats for the wax chamber, working platform and transfer trays are independently adjustable from 50oC to 75oC. The cryomodule is adjustable from -10oC to 0oC. The operating temperature is shown on the display.

## Wax Flow Rate

### Tissue-Tek TEC 6 Embedding Centre

Rate of wax flow can be adjusted by turning the control knob on top of the paraffin dispenser. To increase flow turn anticlockwise, to decrease turn clockwise.

## Timer Operation

### Tissue-Tek TEC 6 Embedding Centre only

The wax dispensing section and the working platform of the unit is set to turn on automatically at 5.00a.m, and turns of at 5.00p.m., Mondays to Fridays. For the operation of the timer please refer to the Operating Manual for the Tissue-Tek TEC 6 Embedding Centre.

Note: The cryo-module of the unit is set to auto.

# Related Documents

* CD\_AP\_FA\_0003 Daily QC Check Form
* CD\_AP\_FA\_0327 Technical Quality Feedback Form
* ANA\_HPM\_002 Embedding Station Maintenance Log

# References

* Sakura Tissue-Tek TEC 6 User manual
* Carson FL, Hladik Cappellano C. Histotechnology: A Self-Instructional Text. 5th ed. Chicago, IL: ASCP Press; 2020.
* Spillan BS. *Proper Tissue Embedding Practices*. Histologic Magazine. April 1976;VI(2):79.
* Winsor L. Tissue processing. In: Woods A, Ellis R eds. Laboratory Histopathology: A Complete Reference. New York: Churchill Livingstone; 1994
* Suvarna SK, Layton C, Bancroft JD, eds. *Theory and Practive of Histological Techniques*. 8th ed. Philadelphia, PA: Churchill Livingstone Elsevier; 2019