

RISK ASSESSMENT AND STANDARD OPERATING PROCEDURE

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| **1. PERSON CARRYING OUT ASSESSMENT** |
| **Name** | Nanet Willumsen | **Position** | Research technician | **Date** | 27/04/2020 |
| **2. DESCRIPTION OF ACTIVITY (include storage, transport and disposal if relevant)** |
| **Generic Immunohistochemistry protocol for FFPE tissue****Notes:** Nitrile gloves will dissolve upon continued exposure to xylene. Use utensils to place/remove slides racks in/out of xylene. When coverslipping slides, use tweezers to remove single slides from the rack submerged in xylene.1. In fume hood: Dewax and dehydrate slides; x2 in xylene, x1 in 100% ethanol, 5mins each
2. On bench continue to dehydrate slides in 100% ethanol, 90%eth, 70%eth and dH20, 5mins each.
3. Quench slide in peroxidase: 1:30 H2O2to 1XPBS, 30mins (10ml 30% H2O2 + 290ml PBS).
4. During quench step, place citrate buffer (CB) in steamer with a foil lid, turn on steamer and allow CB to warm (30ml 10X stock CB + 270ml H2O).
5. After quench, wash slides in dH2O, 5mins.
6. In the steamer, place the slides in the warm 1X CB for 20mins.
7. After 20mins turn off steamer and transfer the CB containing the slides and place on ice for ~15mins until slides are at room temperature (RT).
8. Wash slides x1 in dH2O, and x1 in 1XPBS, 5mins each.
9. Block in appropriate blocking serum if necessary.
10. Add ~130µl of the primary antibody (10 antibody in primary diluent (PBS-Tx 0.3%)).
11. Incubate at RT for 1HR or 4°C overnight.
12. Wash in 1XPBS, 5mins.
13. Incubate with secondary antibody.
14. Wash x3 in 1XPBS, 5mins each.
15. Incubate in amplification step if necessary, followed by x3 5min 1XPBS washes.
16. Incubate in DAB – benchtop for kits, hood for ‘bucket’ DAB.
17. Wash in dH2O, 5mins.
18. Wash in running dH2O, 5 mins.
19. Incubate in Mayer’s haematoxylin (~2 mins).
20. Wash in running tap water, 5mins.
21. Dehydrate tissue through ethanol: 70%, 90%, 100%, 100%, 5mins each.
22. In fume hood: Clear slides x2 in Xylene for 5mins each, followed by a final >30 min incubation in xylene.
23. In fume hood: Coverslip slides with DPX mountant and leave to dry.
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| **3. LOCATION** |
| **Campus** | Hammersmith | **Building** | Burlington Danes | **Room** | 4th Floor |
| **4. HAZARD SUMMARY** |
| **Accessibility** | X | **Mechanical** | X |
| **Manual Handling** | X | **Hazardous Substances** | Y |
| **Electrical** | X | **Noise** | X |
| **Working at height** | X | **Extreme temperature** | X |
| **Falling objects** | X | **Pressure/steam** | Y |
| **Trip hazards** | X | **Other** | X |
| **Lone Working Permitted?** | **Yes** **[x]  No** **[ ]**  | **Permit-to-Work required for planned maintenance?** | **Yes [ ]  No [ ]  N/A [x]**  |
| **5. Who might be harmed and how?** |
| **Staff / students [x]**  |       | **Cleaners, engineers etc [ ]**  |       |
| **Support staff [ ]**  |       | **Other**       |       |
| **6. How often is the process being carried out?** |
| Once a day [ ]  Once a week [x]  Once a month [x]  Every 6 months [ ]  Annually [ ]  Other – give details       |
| **7. Brief description of the procedure** | **Precautions (Controls) in place** | **Is risk high, medium or low?** |
| Xylene | Wear PPE and use in a chemical fume hood. Nitrile gloves **will dissolve** in xylene. Dispose via chemical waste route only. Store in fume cabinet separate to flammable solvents, keep away from sources of ignition. | Low |
| Ethanol | Wear PPE. Store locked in flammable cupboard. Avoid use near flames, heat or sparks. | Low |
| Steamer | Wear PPE, keep away from hot steam outlets. Use forceps to transfer racks into boiling solutions. Do not use glass in steamer. Allow solutions that are too hot to cool before handling/disposing. Do not place steamer directly under plug socket. | Low |
| H202 (30%) | Wear PPE. Store away from combustible materials. Use in well ventilated area/fume cupboard. | Low |
| Bucket DAB | Wear PPE and use in a chemical fume hood. Dispose via chemical waste route only. Store in fume cabinet separate to flammable solvents unless at -20 storage. Keep away from sources of ignition. | Low |
| DPX | Wear PPE and use in a chemical fume hood. Dispose via chemical waste route only. Store in fume cabinet separate to flammable solvents, keep away from sources of ignition. **D DO NOT USE IF PREGNANT.** Has the H360FD code – May damage fertility, may damage the unborn child **– contact Occupational health for advice.** | Low |
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| **8. Are extra precautions needed? If no please tick box and move onto next section** **[x]**  |
| **If yes, please describe** | **Who has been asked to do this?** | **By what date?** |
|       |       |       |
| **9. EMERGENCY ACTIONS** |
| **First Aid** treatment for skin contact with xylene and ethanol:Remove any contaminated clothing. Rinse skin well with water for a prolonged period. If necessary, seek medical attention.**First Aid** treatment for inhalation:Remove person to a well ventilated area. If person seems to become dizzy or loses consciousness call security and wait for emergency services to arrive.**First Aid** treatment for eye splash:Rinse thoroughly for at least 15min using the eye wash station. If necessary, seek medical attention.**First Aid** treatment for ingestion:Do NOT induce vomiting. Rinse mouth with water and seek medical attention immediately.**First Aid** for scalds (from steam):Remove from heat source. Cool burn with cool water for >20mins, removing any clothing near burn unless stuck to skin. Can cover with cling film. Seek medical attention if burn is major. **Chemical Spillage**Within the laboratory but outside any primary containment facility such as a fume cupboard:If the spillage is small <2ml, clean up with paper towel and water. See Local Rules. If spillage is Xylene and is large, evacuate immediate area. Notify laboratory manager and building manager. Spill will need to be cleaned up using the chemical spill kit. Facemask, lab coat and xylene resistant gloves must be worn. Dispose of waste in yellow chemical waste bag provided and send through Imperial hazardous waste route. If spillage is ethanol and is large, use chemical spill kit immediately. Within a fume cupboard (if relevant):If there’s a spillage in the fume cupboard. Lower sash as low as possible. Clean spill using chemical spill kit. Dispose of via Imperial hazardous waste route. De-contaminate area with water and paper towel.**Emergency support through security: 4444 (+442075891000)****In all instances** of accident OR near miss, notify the safety department and complete a SALUS report. <https://www.imperial.ac.uk/safety/safety-by-topic/accidents--incidents/>Occupational health contacts: <https://www.imperial.ac.uk/occupational-health/> email: occhealth@imperial.ac.uk phone: +44 20 7594 9401 |
| **10. Monitor and review** |
| Controls should be monitored: daily [ ]  weekly [ ]  monthly [ ]  6 monthly [ ]  annually [x]  other [ ] I will review this risk assessment at least every 6 months [ ]  every 12 months [x]  **Immediately in the event of process / location change or incident or accident** |
| **11. Training record – use this section to record the names and date of any persons you are training in this risk assessment and associated procedures** |
| Name | Date | Name | Date |
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<https://www.imperial.ac.uk/safety/forms/> for all specific risk assessment forms.