

PROTOCOL FIXATION AND INCLUSION IN PARAFFIN

- Withdrawal of the anatomical piece;
- Place the piece in the cassette between two sponges;
- Immerse the cassette in 4% paraformaldehyde in 0.1 M PBS or in 4% buffered formalin, for at least 48 hours;
- Wash the cassette in running water O.N;
- Place the cassette for 12-24 hours in 70% ethanol (changing the ethanol after the first hour) and continue increasing ethanol scale, 90% ethanol and then 100%, with incubations of 2-3 hours (changing the ethanol after the first 30 minutes);
- Xylol for 30 min/1 hour;
- Xylol for 30 min;
- Inclusion in hot paraffin diluted;
- After two hours from the inclusion in paraffin, recover the piece, place it on a steel mold, where previously a small amount of liquid paraffin had been placed, and pour the paraffin over to form the block. This mold containing the piece will be kept at RT to allow the paraffin to solidify;
- The block obtained will be cut to the microtome.

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4/06/2015

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