



3th stage

Tissues Processing

Lab 6

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Tissues Processing

Tissue Processing Sequence

Dehydration

Removing
water from
tissues

Clearing

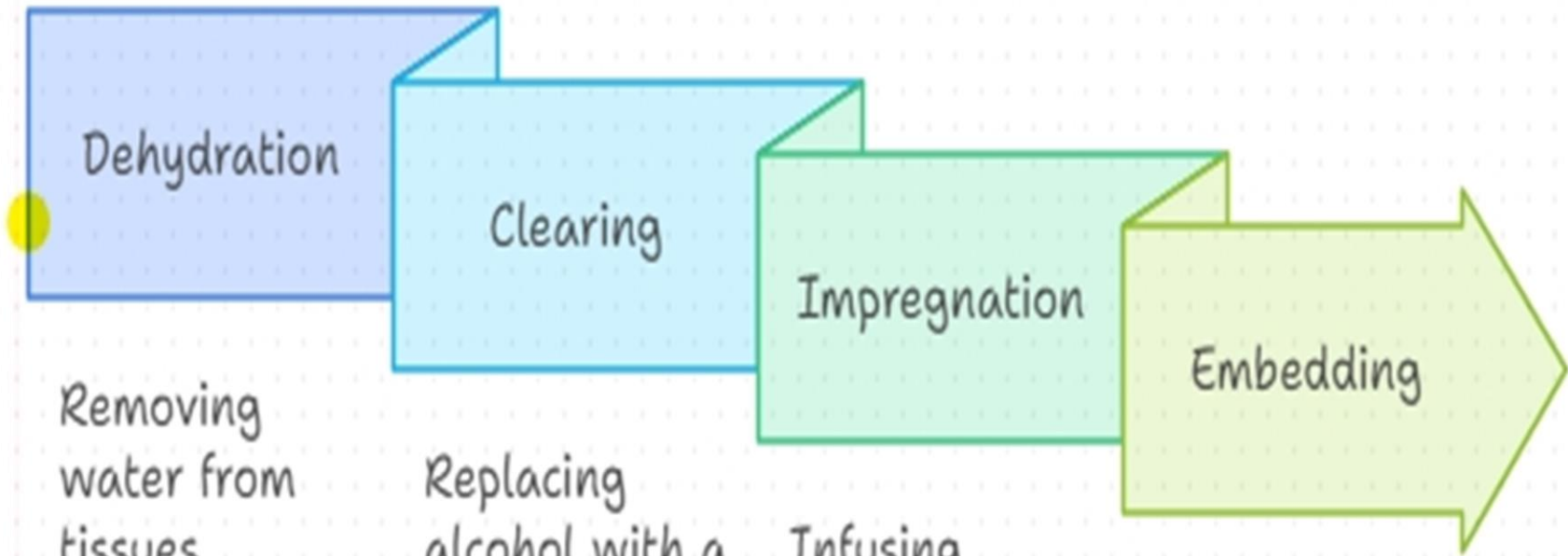
Replacing
alcohol with a
clearing agent

Impregnation

Infusing
tissues with a
paraffin

Embedding

Embedding
tissues in wax



1_Dehydration

Which dehydration agent to use for tissue preparation?

Ethanol

Widely used, safe, effective.

Methanol

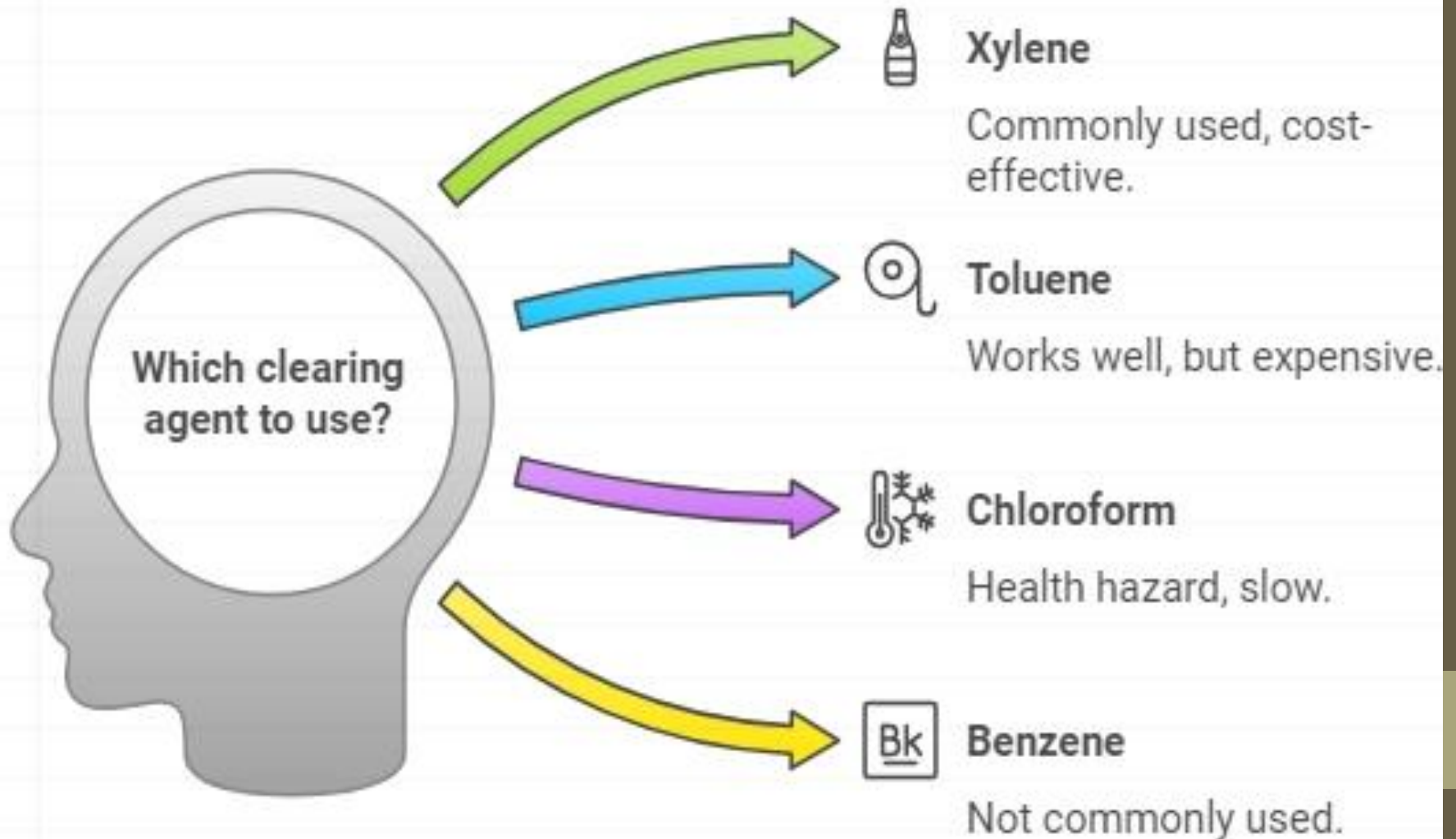
Less common, effective.

Acetone

Fast, but a fire hazard.



2_Clearing



Choice of a clearing agent depends upon following

- The type of tissue to be processed and the type of processing to be undertaken.
- Safety factors
- Cost and convenience
- Speedy removal by molten paraffin wax.
- Minimal tissue damage

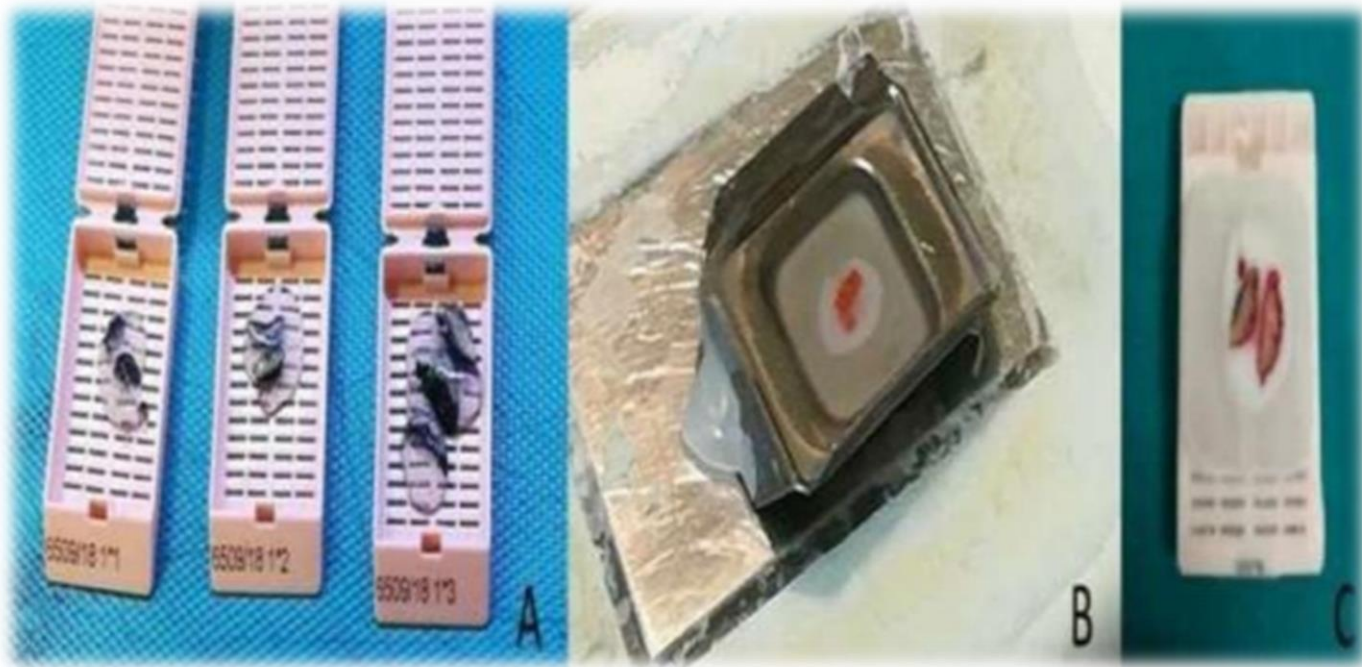
3_Impregnation

- The tissues are put from 2 - 4 hours in hot soft paraffin at 57 °C in the oven.
- The paraffin will penetrate in-between the cells of the tissues.



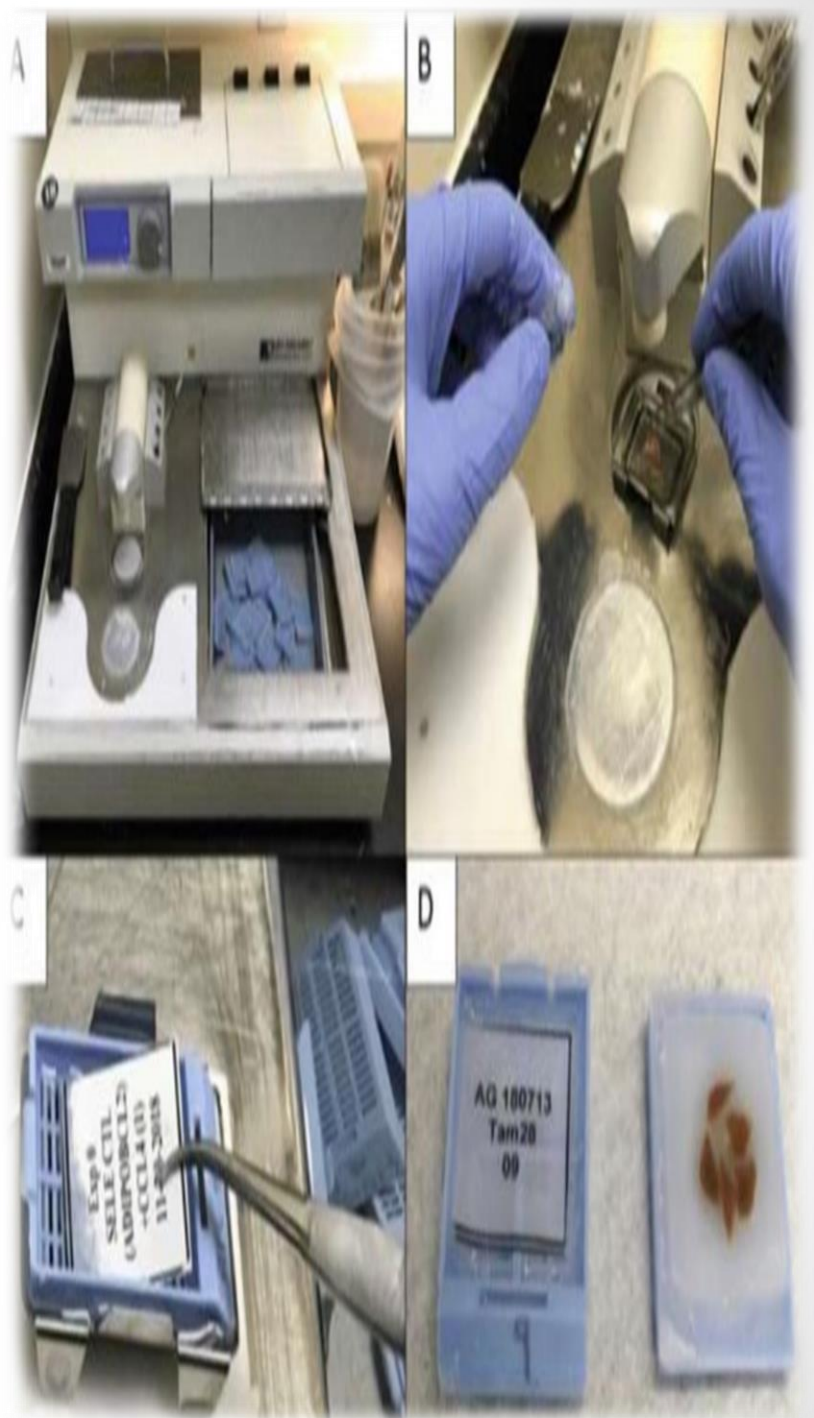
4_Embedding

- Impregnated tissues are placed in a mould and then fresh melted wax is poured in it and allowed to settle and solidify.
- Once the block has cooled sufficiently to form a surface skin it should be immersed in cold water to cool it rapidly.



Trimming

After preparation of was templates preferably trimmed with a sharp blade so that the specimen be in a position Suitable for cutting edges so that they become parallel and can be applied to edge of the knife of microtome.



Summary of Paraffin Wax Embedding

1. Dehydration:

- a. 70% alcohol 1 hour
- b. 90% alcohol I 1 hour
- c. 90% alcohol II 1 hours

2. Clearing:

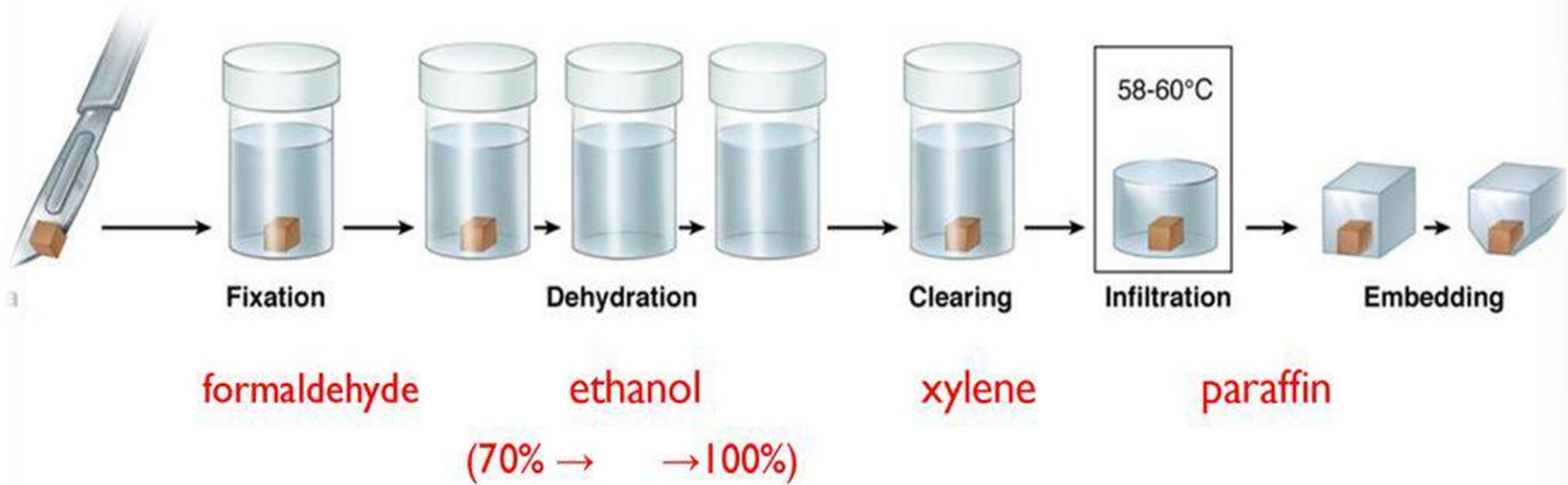
- a. Xylene I 15 minute
- b. Xylene II 15 minute

3. Wax Impregnation:

- a. Paraffin wax I 1 hour
- b. Paraffin wax II 1 hour
- c. Paraffin wax III 1 hou

4.Embedding

Sample preparation for the microtome sectioning



Thank You

