

RDO MULTIPURPOSE DECALCIFICATION TECHNIQUE

RDO is a remarkably effective multipurpose decalcifier that can be tailored to suit your specific lab routine. As with most acids, nuclei acids in the cell can become subject to ribonuclease digestion, resulting in a loss of basophilic properties. Careful monitoring should be used with **RDO** to avoid over decalcification. Most decalcification occurs in approximately 4-6 hours or less, depending on the thickness and density of specimens. Overnight decalcification should be avoided. Full Strength **RDO** is used for hard compact bone, ie. femur heads. **RDO** may be diluted with tap water to permit multipurpose use. Dilution with tap water is recommended for mildly calcified specimens such as bone marrow biopsies (core). In either case, the standard procedure should be to check the specimen every ½ -1 hour for mildly calcified specimens and every 1-2 hours for compact bone to determine the endpoint of decalcification.

Techniques for Optimal Decalcification with RDO

Frequent mild agitation or swirling of the specimen in solution will enhance even penetration and decrease the exposure time of the tissue to the acid solution. This will also minimize over decalcification of the outer tissue or bone before sufficient core decalcifying is achieved.

Dilute stock solution of **RDO** with tap water. Dilution factors will be dependent upon thickness and density of a specimen. Dilution will allow you to slow down the decalcification process to suit you specific lab routine.

To avoid over decalcification, check specimens at regular intervals for an endpoint. Every ½ - 1 hour for mildly calcified specimens and every 1-2 hours for compact bone.

Reducing temperature of the decalcifying solution to approximately 20 ° C will promote histochemical staining in procedures such as H & E, Masson's, Van Gieson's, and Azure-Eosin.

To remove sediment, **RDO** may be filtered, if desired, without altering its effectiveness.

To achieve optimal performance, do not reuse **RDO**. Since the nature of a decalcifying agent is to release calcium ions from the bone into the acid solution, as the solution becomes saturated with calcium ions, the decalcification process will slow down.

Addition of an alcoholic solution can aid in preventing undue swelling and hydrolysis of the tissue. It will, however, slow down the decalcifying agent. An 8:2 ratio of stock **RDO** (8) to 80% alcohol (2) can serve as a standard range for this method.

Rinse specimens thoroughly in running tap water after decalcification.

References:

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