

Quick Gelatin-Albumin Embedding

Egg Albumin 45 gm in 100 ml 0.1 M phosphate buffer
Gelatin 0.75 gm in 50 ml 0.1 M phosphate buffer

Warm 100 ml of buffer (or PBS) to about 35° C, add albumin and remove from heat (or maintain the low temperature, otherwise you end up with boiled egg). Stir for about 2 hours or so to dissolve the albumin.

Warm 50 ml of buffer to dissolve the gelatin. Let this solution cool before adding to the albumin solution.

Filter the gelatin-albumin mixture through a single layer of gauze to remove any large lumps. Filtering is easily accomplished with a completely unfolded 2X2 gauze pad draped over a pee cup or specimen jar and anchored with a rubber band.

Add 1 ml of 2.5% sodium azide to each 100 ml (1.5 ml per 150 ml batch) and store in the refrigerator for up to several months.

Note: We use Sigma #A 5253 Chicken Egg Albumin, Grade II. Some other grades don't gel very well.

To Embed:

Orient the specimen in a mold. We use Peel-A-Way molds. Samples may be pinned into the preferred orientation. Small samples may be immersed into polymerizing Quick-Embed in a mold.

Pour 20 ml of Quick-Embed into a small disposable container, such as a pharmacy cup or pee cup. Add 1.0 ml of 25% glutaraldehyde and stir like mad for 10-20 seconds. Add the mixture to the tissue. Hardening occurs in 30-90 seconds (slower if the mixture is cold). If it doesn't harden, try adding more glutaraldehyde.

Let the block sit in air for 10-15 minutes, then immerse in PBS or buffer until needed. Trim the block and glue it onto the vibratome chuck. The original protocol recommended vibratoming with a cold bath, but this is not always required.

Glutaraldehyde notes:

1. This embedding method is not recommended for tissues to be labeled by immuno-labels or by fluorescent methods.
2. Glutaraldehyde is a strong fixative that may denature antigens. It is expected that it is mostly binding to the Quick-Embed components but some may penetrate the surface of the tissue. Sections should be thoroughly rinsed in buffered saline as they are cut to remove any unbound fixative.
3. Glutaraldehyde is brightly fluorescent in the green to yellow wavelengths. It might be possible to quench the autofluorescence by methods such as Sudan Black.

(original protocol from Sue McConnell, Stanford University, 8/11/89, edited by Glen MacDonald, 1989-2018)