

COLLECTION AND TRANSPORT OF SPECIMENS FOR EPIDERMOLYSIS BULLOSA DIAGNOSIS

PRELIMINARY STEPS

Contact us

Please fax the referral form to Prof Dedee Murrell so we know that you intend to send us the biopsy, to 02-9113-2906. This way we can best plan to read the results in a timely fashion for you and, if necessary, organise the necessary transport medium, fixative and paperwork to be sent.

Our Contact Details

Professor Dedee Murrell
Phone: +61-2- 9113 2543/3418
Fax: +61-2- 9113- 2906
Email: d.murrell@unsw.edu.au"

You will need to have the following available:

Transport Media

Specimens are to be submitted in the following solutions:

IF mapping - Michel's Medium

EM examination - 2.5% glutaraldehyde

Paraffin sections- 10% neutral buffered formalin (however, we can do H&E on the IF slides to save an additional biopsy)

Paperwork

The following paperwork is required:

Consent forms: a consent form routinely used for the local hospital and a consent form for participation in the EB research program. This consent form has been approved by our ethics committee and should be signed by the parent/patient. The original should be submitted with the specimens and a copy given to the patient and one in their file. This consent form and information sheet are available from the website www.blisters.org.au under the lab referral section.

Request form: Either a local request form or an EB request form. Request forms are also available on the lab referral section.

The request form must include the following:

Full patient details including **Medicare number and, patient/parental signature** in the appropriate portion of the request form (this is to avoid the patient receiving a bill).

The **Referring Consultant's contact details** including provider number, address, mobile phone number and fax number. **(NB not just the registrar details please)**

Clinical history including patient history, pedigree, age of first blisters, family history, clinical impression (simplex, junctional or dystrophic), site of the biopsy and whether it is a rubbed area or non-rubbed normal area.

If from overseas, please indicate who is to be billed, the doctor/hospital or the patient on the pathology form. Contact us about charges.

TAKING THE BIOPSY

After informed consent and the proband or the parent, as appropriate, has signed the consent form: (see above for details.)

Choose an area of skin that appears clinically unaffected but near where the patient gets their blisters and at a site where in future the scar will not be too obvious, e.g. overlying the appendix area.

You will need to rub this site with an eraser/ pencil eraser about 20 times first; it may turn red but you should wait at least 5 minutes for a blister to develop microscopically if it is a newborn. If it is an adult, or a child with what appears to be a less severe form of EB, such as DDEB or EBS, ideally mark with a pen the site (e.g. near the site on their leg/feet where the blisters usually occur) and have them take the pencil eraser to the coffee shop and rub on the area themselves many times to turn it red if it did not do so when you were rubbing it. If the skin starts to tear/peel then they should stop. Or, ask them to do whatever exercise induces blisters the day before to be sure you are taking a biopsy of a fresh blister. They may have to do this the day before or wait at least 3 hours after rubbing.

Clean the skin with an alcohol wipe or other antiseptic solution.

Anaesthetise with 1% lignocaine (plus adrenaline to minimise bleeding) to raise a bleb. Do not use EMLA as this has been shown to induce changes that mimic epidermolysis bullosa simplex (Hoss *et al.* J Cutan Pathol. 1999; 26:100-4).

Take a 3 mm punch biopsy from the rubbed area and place it in Michel's solution (the same transport media used for immunofluorescence specimens). If you can get the specimen to St George the same day then liquid nitrogen or normal saline is preferable.

Take a 2 mm punch biopsy from the (same) rubbed area and place in 2.5% glutaraldehyde solution for electron microscopy.

If possible, it is useful to also take a 3 mm punch biopsy from an unaffected, NON rubbed area, usually the inner upper arm, because sometimes the blistering will inform you of the layer of blistering but the reduction of protein staining, if present, can be more easily assessed on skin that has not actually blistered. This would be transported as for the other IF specimen, in a separate Michel's media container, labeled as normal non-rubbed skin and the site.

Suture with 4/0 ethilon depending on the site, and apply mupirocin ointment to the site bd until stitches are removed in 1 week. Alternatively, catgut absorbable sutures can be used that fall out on their own.

For more detail, see Intong LRA & Murrell DF: How to Take Skin Biopsies for Epidermolysis Bullosa. *Dermatol Clin N Am*, 28 (2): 197-200, 2010.

SENDING THE BIOPSY

Send the specimen at 4°C with coolpacks (do not freeze) by courier to:

Prof Dedee Murrell / Wei Melbourne
Anatomical Pathology
4th floor Clinical Services Building, SEALS,
St George Hospital
Gray St, Kogarah
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