Detailed primary sample collection procedures

FORMALIN FIXED PARAFFIN EMBEDDED (FFPE)

Collection of Sample (guideline only)

1. Sample is collected into a fixative to preserve the tissue and the structure of the cells. The most common fixative is a neutral buffered formalin (10%).
2. The tissue is then processed. The samples are immersed in multiple baths of ethanol to dehydrate the tissue, followed by a clearing agent such as xylene and finally paraffin wax to impregnate the tissue.
3. The tissue is then embedded into a hard paraffin block, which is then placed in a mold containing more molten wax and allowed to cool and harden.
4. The tissue can then be sectioned into very thin sections using a microtome. These slices are then placed on a glass slide for staining and/or other testing.
5. Label slides with patient name and one other unique identifier.
6. Cut at least two sections for each probe requested at the appropriate depth (see note) and place on six poly lysine slides.
7. Place slides in a slide container and into a sealed bag with a completed referral form.
8. Please advise IGENZ of the specimen.
9. Arrange transportation of the specimen to IGENZ.

Note: The depth of the sections (microns) will depend on the type of disease required for analysis. Diseases such as Small Round Blue Cell Tumours have small cells, and require sections cut at 2-4 microns to prevent cells overlapping and cause analysis artifacts resulting in false positive or negative findings. Those that have larger cells require sections cut at 4-6 microns. Sections cut at <4 microns may also result in artifacts pertaining to loss of signal and possibly enumeration issues.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	2 poly lysine slides (per probe requested) with a 2-4 micron section on each
Collection container	Slide holder
Additives (if any)	Poly lysine on slides
Transportation time	Sample should be received by IGENZ within 10 working days of the request.
Transportation temperature	Ambient
Tests available	FISH Molecular Testing
Time limits for extra testing	1 week from receipt of sample
Repeat examination due to analytical failure	Lab may request further slides from different blocks if initial preparation fails within 3 days.
Clinical information	Please provide a pathology report and any other relevant information required.
Turn around time	FISH: Urgent 24 hours, Routine 3 working days Molecular Testing: Urgent 72 hours, Routine 5 working days
Special Instructions	Please provide an H&E slide with area of interest marked

BONE MARROW ASPIRATE (BMASP)

Collection Procedure (guideline only)

1. Obtain 1-2mL (paediatric: 1mL) of bone marrow in a lithium/sodium heparin tube. We recommend sending the first aspirate drawn, however the laboratory will accept subsequent draws.
2. If aspirate is required for other testing, place a minimum of 2 mL in a sterile bijoux tube to send to IGENZ.
3. Also make 6 bone marrow smears.
4. Label tube/s and slide/s with patient name and one other unique identifier.
5. Place slide/s in slide holder and place slide holders and tube/s in biohazard bag with completed referral form.
6. Please advise IGENZ of the specimen.
7. Arrange transportation of the specimen to IGENZ.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	1ml or more
Collection container	Heparin Vacutainer Or Sterile bijoux And Bone Marrow smear slides
Additive/s (if any)	Heparin
Transportation time	Sample should be received by IGENZ same day as sample collection.
Transportation temperature	Ambient
Tests available	FISH and Cytogenetics
Time limits for extra testing	6 months (for fixed cell suspension) or 48 hours after issue of final report for all other sample types.
Repeat examination due to analytical failure	If bone marrow aspirate fails to yield a result, the decision to request a recollection of peripheral blood will be requested within 5 working days.
Clinical information	Please provide a haematology report and any other relevant information required.
Special Instructions	Please note that EDTA is not the preferred anti-coagulant and its use is therefore discouraged. Clotted bone marrow is unacceptable, however the laboratory will attempt to culture any sample received.

BONE MARROW TREPHINE (BMTREP)

Collection Procedure (guideline only)

1. The trephine (or part of the trephine if other testing such is required) should be placed in a sterile container and heparinised media.
2. Also draw 7-10mL (paediatric: 2mL) of whole blood in a green-top sodium heparin tube. This is typically sufficient for cytogenetics and FISH studies.
3. Label tube/s with patient name and one other unique identifier.
4. Place tube/s in biohazard bag with completed referral form.
5. Please advise IGENZ of the specimen.
6. Arrange transportation of the specimen to IGENZ.

Note: Always include the additional sample of peripheral blood in case the initial preparation fails.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	5mm or more
Collection container	Sterile container with additives
Additive/s (if any)	RPMI with heparin Or MEM with heparin IGENZ can provide containers with media if required
Transportation time	Sample should be received by IGENZ same day as sample collection.
Transportation temperature	Ambient
Time limits for extra testing	48 hours post issue of report or 6 months on fixed cells suspension.
Repeat examination due to analytical failure	If bone marrow trephine fails to yield a result, the extra specimen of peripheral blood will be processed accordingly.
Clinical information	Please provide a haematology report and any other relevant information required.
Special Instructions	It is recommended to include an additional sample of peripheral blood in case the preparation fails.

PERIPHERAL BLOOD (PB)

Collection Procedure (guideline only)

1. Draw 7-10mL (paediatric: 2mL) of whole blood in a green-top sodium heparin tube.
2. A total of 5-10mL (paediatric: 2-5mL) is typically sufficient for cytogenetics and FISH studies.
3. Invert several times to mix blood. (Clotted blood is not acceptable).
4. Label tube/s with patient name and one other unique identifier.
5. Place tube/s in biohazard bag with completed referral form.
6. Please advise IGENZ of the specimen.
7. Arrange transportation of the specimen to IGENZ.

Collection procedure – Liquid Biopsy

1. Draw 8mL of whole blood in a Streck tube with a butterfly syringe or large gauge needle only
2. Gently invert sample 6 times after collection
3. Label tube/s with patient name and one other unique identifier.
4. Place tube/s in biohazard bag with completed referral form.
5. Arrange transportation of the specimen to IGENZ.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	5-10 mL (paediatric: 2-5mL)
Collection container	Heparin Vacutainer OR Steck tube (LBx)
Additive/s (if any)	None
Transportation time	Sample should be received by IGENZ same day as sample collection. LBx – sample should be received within 7 days of collection
Transportation temperature	Ambient
Time limits for extra testing	48 hours after issue of report or 6 months if fixed cells suspension.
Repeat examination due to analytical failure	The decision to request a recollection should be made in no more than 10 working days.
Clinical information	Please provide a hematology report and any other relevant information required.
Special Instructions	Clotted blood is unacceptable, however the laboratory will attempt to culture any sample received. Haemolysed samples are unable to be processed

AMNIOTIC FLUID (AF)

Collection Procedure (guideline only)

1. Draw 20-25 mL of amniotic fluid in a sterile syringe.
2. Discard the first 2 mL of amniotic fluid.
3. Remove the needle and transfer the specimen to two screw capped, sterile centrifuge tubes.
4. Label tubes with patient name and one other unique identifier.
5. Place tubes in biohazard bag with completed referral form.
6. Please advise IGENZ of the specimen.
7. Arrange transportation of the specimen to IGENZ.

Note: Optimal gestation for amniotic fluid specimens is 16 weeks (+2 weeks). If an amniotic fluid has been referred outside the optimal gestation period, non-viable cell numbers may be encountered and an incomplete result obtained. (If gestation is less than 14 weeks, low numbers of cells are available).

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	
5mL (minimum)	FISH
15 mL	Cytogenetics
15mL or more	FISH and Cytogenetics
Collection container	Screw capped sterile centrifuge tube Tubes can be provided by IGENZ if required
Additive/s (if any)	None
Transportation time	Sample should be received by IGENZ same day as sample collection.
Transportation temperature	Ambient DO NOT REFRIGERATE OR FREEZE
Time limits for extra testing	6 months
Repeat examination due to analytical failure	The decision to request a recollection should be made in no more than 10 working days.
Clinical information	Please provide a clinical report and any other relevant information required.

CHORIONIC VILLUS (CV)

Collection Procedure (guideline only)

1. Obtain 20-30mg of chorionic villus specimen by the transabdominal or transcervical method.
2. **EITHER**
    1. Transfer the CV to a petri dish containing transport medium.
    2. Using a stereo microscope and sterile forceps, assess the quality and quantity of the villi.
    3. Remove any blood clots and maternal decidua.
    4. Transfer the CV using sterile technique to a sterile container containing transport medium.

    OR

    1. If the above facilities are not available transfer the CV into a sterile container containing transport medium.
3. Label container with patient name and one other unique identifier.
4. Place container in biohazard bag with completed referral form.
5. Please advise IGENZ of the specimen.
6. Arrange transportation of the specimen to IGENZ.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	Minimum of 20mg of chorionic villus
Collection container	Sterile container with additive IGENZ can provide containers with media if required Transport media supplied by IGENZ should be stored in the refrigerator for up to three weeks (or six months if frozen) before being discarded.
Additive/s (if any)	RPMI Or MEM
Transportation time	Sample should be received by IGENZ same day as sample collection.
Transportation temperature	Ambient
Time limits for extra testing	6 months
Repeat examination due to analytical failure	The decision to request a recollection should be made in no more than 10 days.
Clinical information	Please provide a clinical report and any other relevant information required.
Special Instructions	None

PRODUCTS OF CONCEPTION (POC)

Collection Procedure (guideline only)

1. Where a foetus is identified, obtain 1 cm^3 biopsy specimen of muscle/fascia from the thigh and about 1 cm3 of placenta including 20mg of chorionic villi.
2. If a foetus cannot be identified collect villus material or tissue that appears to be of foetal origin.
3. Aseptically place each specimen in a screw capped sterile container with transport media.
4. If no transport media is available it may be sent with sterile saline.
5. Label each container/s with specimen type, patient name and one other unique identifier.
6. Place container/s in biohazard bag with completed referral form.
7. Please advise IGENZ on 09 307 3981 of the specimen.
8. Arrange transportation of the specimen to IGENZ

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	1 cm^3 biopsy specimen of foetus and/or Minimum of 20mg of chorionic villus or 1 teaspoon of tissue that appears to be of foetal origin.
Collection container	Sterile container with additive. IGENZ can provide containers with media if required
Additive/s (if any)	RPMI Or MEM Or sterile saline
Transportation time	Sample should be received by IGENZ same day as sample collection.
Transportation temperature	Ambient DO NOT REFRIGERATE OR FREEZE
Time limits for extra testing	6 months
Repeat examination due to analytical failure	No repeat examination possible.
Clinical information	Please provide a pathology/clinical report and any other relevant information required.
Special Instructions	If foetus/placenta is sent please ensure your lab protocols are followed if patient requests foetus/ placenta to be returned.

SKIN

Collection Procedure (guideline only)

1. Biopsy specimens are best taken by punch biopsy and should be 4mm in diameter and include full thickness of the dermis.
2. Where a foetus is identified, obtain biopsy specimen to include full thickness of dermis. If unable to take a punch biopsy take a full skin thickness ellipse.
3. Aseptically place specimen in a screw capped sterile container with transport media.
4. If no transport media is available specimen may be sent without media.
5. Label container with specimen type, patient name and one other unique identifier.
6. Place container in biohazard bag with completed referral form.
7. Please advise IGENZ of the specimen.
8. Arrange transportation of the specimen to IGENZ.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	Punch biopsy Or Full skin thickness ellipse approx 1cm x 0.5cm
Collection container	Sterile container with additive IGENZ can provide containers with media if required
Additive/s (if any)	RPMI Or MEM Or Sterile saline
Transportation time	Sample should be received by IGENZ same day as sample collection.
Transportation temperature	Ambient DO NOT REFRIGERATE OR FREEZE
Time limits for extra testing	6 months
Repeat examination due to analytical failure	No repeat examination possible.
Clinical information	Please provide a pathology/clinical report and any other relevant information required.
Special Instructions	Ensure full dermis thickness

TUMOUR TISSUE

Collection Procedure (guideline only)

1. Place a 0.5-3.0 cm^3 or larger tumour biopsy in a sterile transport container with 10-20 mL of transport media.
2. Label container with specimen type, patient name and one other unique identifier.
3. Place container in biohazard bag with completed referral form.
4. Please advise IGENZ of the specimen.
5. Arrange transportation of the specimen to IGENZ.

Note: The lab will accept Tissue Imprint slides for urgent FISH analysis

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	Minimum 0.5 cm^3 Or 1 cm long needle biopsy
Collection container	Sterile container with additive IGENZ can provide containers with media if required
Additive/s (if any)	RPMI Or MEM Or Sterile saline
Transportation time	Sample should be received by IGENZ same day as sample collection.
Transportation temperature	Ambient DO NOT REFRIGERATE OR FREEZE
Time limits for extra testing	6 months
Repeat examination due to analytical failure	No repeat examination possible.
Clinical information	Please provide a pathology report and any other relevant information required.
Special Instructions	None

CYTOLOGY

Collection Procedure (guideline only)

1. Specimens can be cell suspension (e.g. ThinPrep® or cytospin preparations) or a fine needle aspiration (FNA).
2. Follow laboratory procedure to make slides from the primary sample. For FNA's slides can be either smears or cell blocks.
3. Contact IGENZ to determine the amount of slides required for the test.
4. Label slide/s with patient name and one other unique identifier.
5. Place slide/s in slide container/s and place the container/s in a biohazard bag with completed referral form.
6. Please advise IGENZ of the specimen.
7. Arrange transportation of the specimen to IGENZ.
8. Note The laboratory requires a reason for referral with each specimen. This ensures the appropriate testing and interpretation of the specimen.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	1 or more slide/s PLEASE CONSULT IGENZ
Collection container	Prepared slides
Additive/s (if any)	None
Transportation time	Sample should be received by IGENZ within 10 working days of the request.
Transportation temperature	Ambient DO NOT REFRIGERATE OR FREEZE
Time limits for extra testing	48 hours after report issued
Repeat examination due to analytical failure	The decision to request a recollection should be made in no more than 10 working days.
Clinical information	Please provide a pathology report and any other relevant information required.
Turn around time	Routine 3 days Urgent 24 hours

FIXED CELL SUSPENSION

Collection Procedure (guideline only)

1. Follow laboratory procedure to fix cells.
2. Put suspension in screw top eppendorf tube and top up with fresh fix.
3. Label eppendorf with patient name and one other unique identifier.
4. Place eppendorf in biohazard bag with completed referral form.
5. Please advise IGENZ of the specimen.
6. Arrange transportation of the specimen to IGENZ.

Note The laboratory requires a reason for referral with each specimen. This ensures the appropriate testing and interpretation of the specimen.

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	1.5mL in eppendorf
Collection container	Screw top eppendorf
Additive/s (if any)	Fix (methanol:acetic acid, 3:1)
Transportation time	Sample should be received by IGENZ within 10 working days of the request.
Transportation temperature	Ambient DO NOT REFRIGERATE OR FREEZE
Time limits for extra testing	6 months
Repeat examination due to analytical failure	The decision to request a recollection should be made in no more than 10 working days.
Clinical Information	Please provide any relevant clinical information
Special Instructions	None

BUCCAL SWABS (DNA Parentage)

Collection Procedure (guideline only for LEGAL tests)

1. Label the external container of each swab with a minimum of the full name and date of birth of each individual to be tested.
2. Write full name and date of birth of each individual to be tested on the correct sticky label
3. Obtain signatures on the sticky label from individual whose sample is to be taken (guardian for children under 16 years of age), witness/identifying person and sample collector
4. Two swabs are to be taken from each individual – swab firmly against the inside of the cheek for ~20 seconds and insert the swab back into the swab container. Repeat with second swab.
5. Secure swabs together with the pre-completed sticky label
6. Place swabs in biohazard bag with completed referral form
7. Arrange transportation of the specimen to IGENZ via courier

IGENZ Requirements

Labelling	2 unique identifiers identical to the referral form
Amount	2 swabs
Collection container	Swab container
Additive/s (if any)	NA
Transportation time	Sample should be received by IGENZ within 7 working days of collection
Transportation temperature	Ambient
Time limits for extra testing	6 months
Repeat examination due to analytical failure	The decision to request a recollection should be made in no more than 10 working days.
Clinical Information	Please provide any relevant clinical information
Special Instructions	None

References

ISO15189 for medical laboratories – particular requirements for quality and competence.
Requirements for Cytogenetic Testing, NPAAC