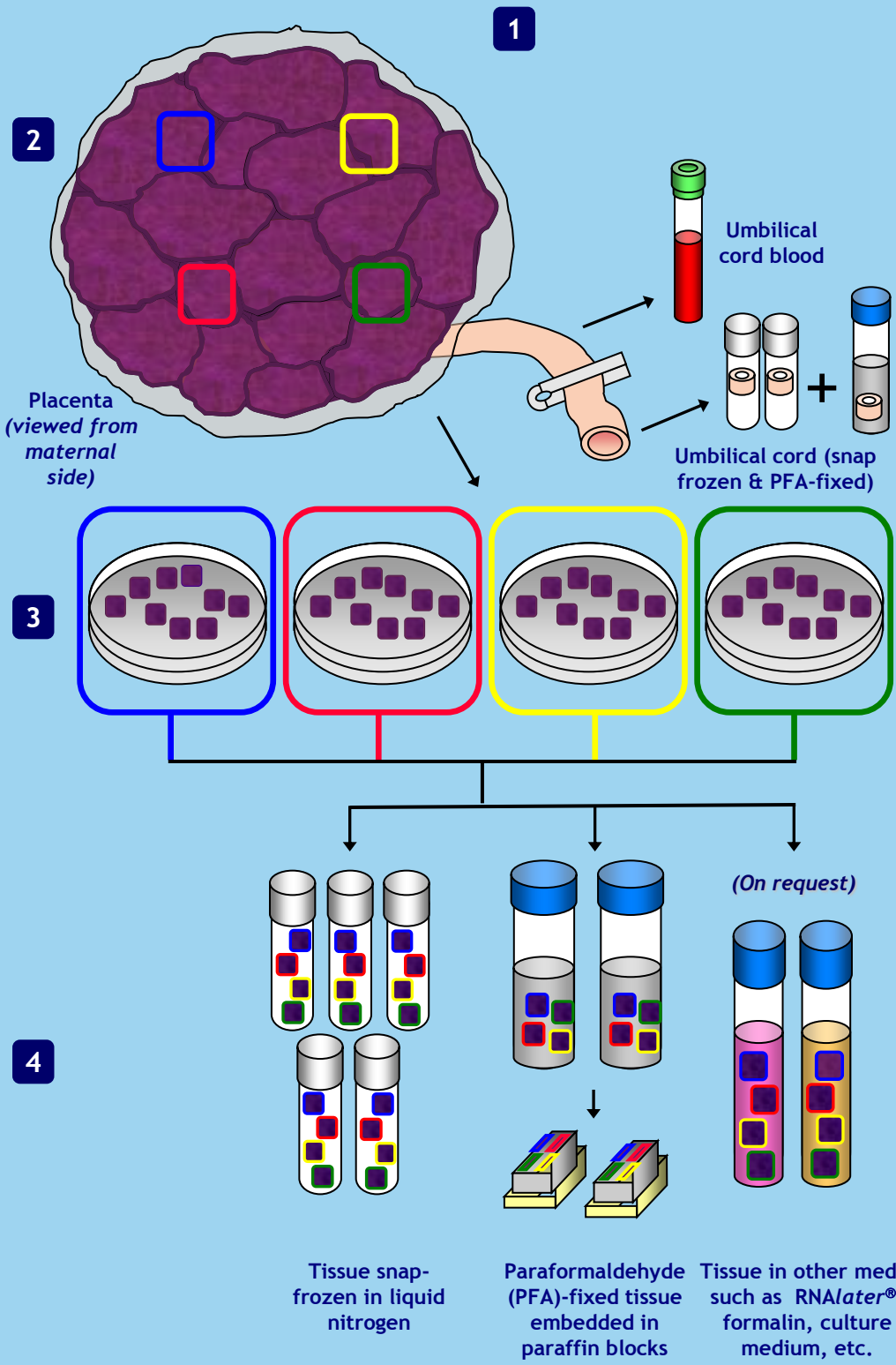


Placenta sampling overview



1. Umbilical cord blood and tissue samples are collected at the start of processing, as required. Placental weight is recorded at the start of processing.

2. Four placental cores (1.5x1.5cm, full thickness excluding the chorionic plate) are taken from representative sites in each quadrant, at least 1.5cm from the edge.

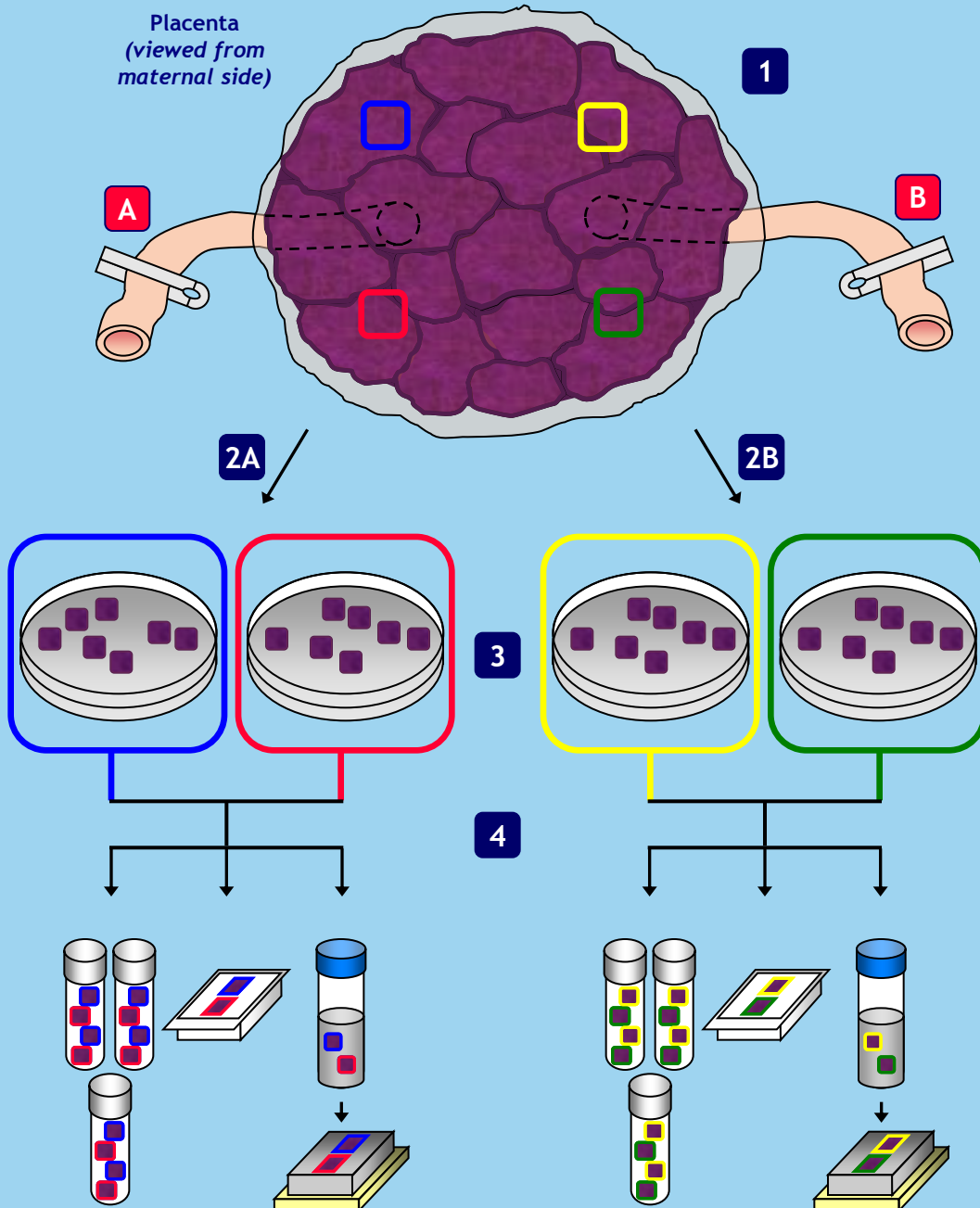
3. Each core is rinsed in PBS, blotted briefly to remove excess PBS and cut into multiple pieces on a blue absorbent pad.

4. Tissue samples from each representative area are transferred to cryovials, fixatives, or miscellaneous media as required.



**Monochorionic twins - Placenta
sampling overview**

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1. Placental weight is recorded at the start of processing and the placenta is positioned to allow clear identification of tissue regions corresponding to each twin.

2. For each twin, two tissue cores (1.0x1.0cm, full thickness excluding the chorionic plate) are taken from cotyledons around the periphery of the placenta, at least 1.5cm from the edge.

3. Each core is cut into multiple pieces in a culture dish containing PBS.

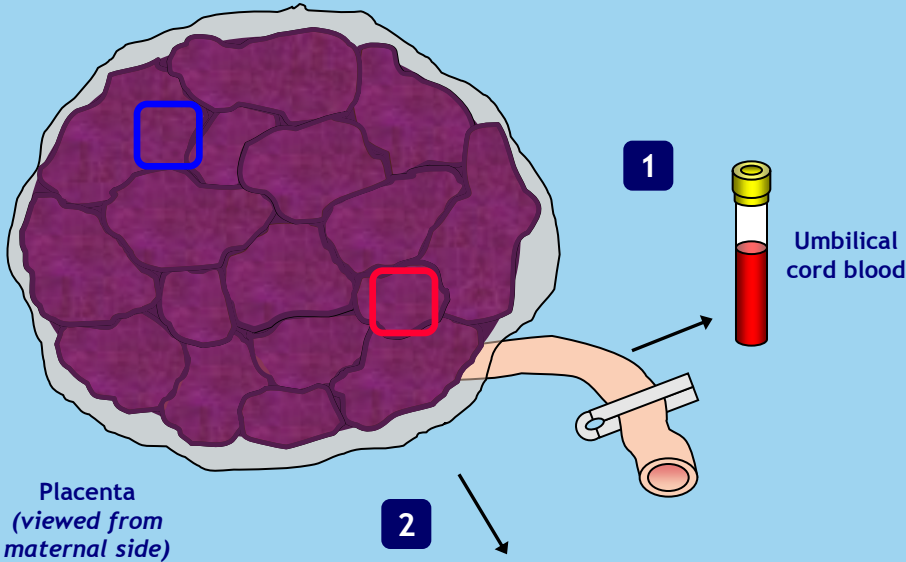
4. Tissue samples from each representative area are blotted briefly to remove excess PBS and transferred to cryovials (3/twin), cryomolds (1/twin), or paraformaldehyde (1/twin).

NOTE: Dichorionic twin placentas are to be sampled using the protocol for singleton placentas, treating each respective twin as a singleton



Investigator-specified study #1 -
Placenta sampling overview

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Placenta
(viewed from
maternal side)

Umbilical
cord blood

1. Umbilical cord blood is collected at the start of processing as required.

2. Two tissue cores (1.5x1.5cm, full thickness excluding the chorionic plate) are taken from cotyledons around the periphery of the placenta, at least 1.5cm from the edge. Each core is cut into multiple pieces in a culture dish containing PBS.

3. Tissue is transferred to a tube containing RNA later and inverted several times to ensure distribution of tissue within solution. Samples are stored at 4C.

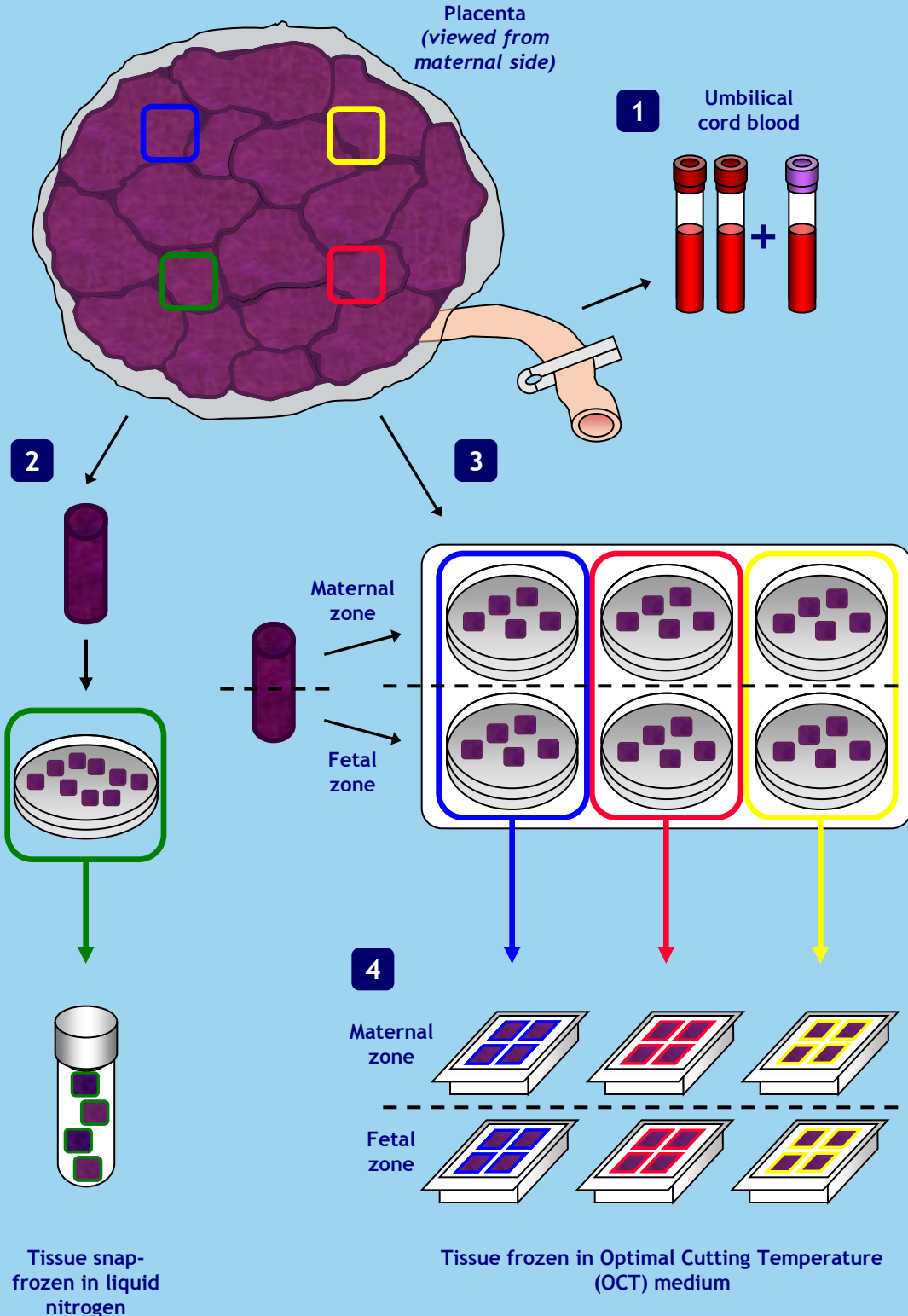
Tissue in RNA later®



**Investigator-specified study #2 -
Placenta sampling overview**

<http://biobank.lunenfeld.ca>
rcwih.biobank@lunenfeld.ca

Placenta
(viewed from
maternal side)



1. Umbilical cord blood samples are collected at the start of processing: 2 redcap tubes (serum) and 1 lavender-cap tube (K₂-EDTA); maximum volumes when possible.

2. One tissue core (1.5x1.5cm, full thickness excluding the chorionic plate, taken from at least 1.5cm from the edge) is excised, cut into multiple pieces in a 10cm culture dish containing PBS, 4-5 pieces are snap-frozen in liquid N₂.

3. Three cores are excised as described in step 2. Each core is cut in half to generate a maternal zone and a fetal zone (collected in 6-well dishes containing PBS). Each zone is then cut into multiple pieces.

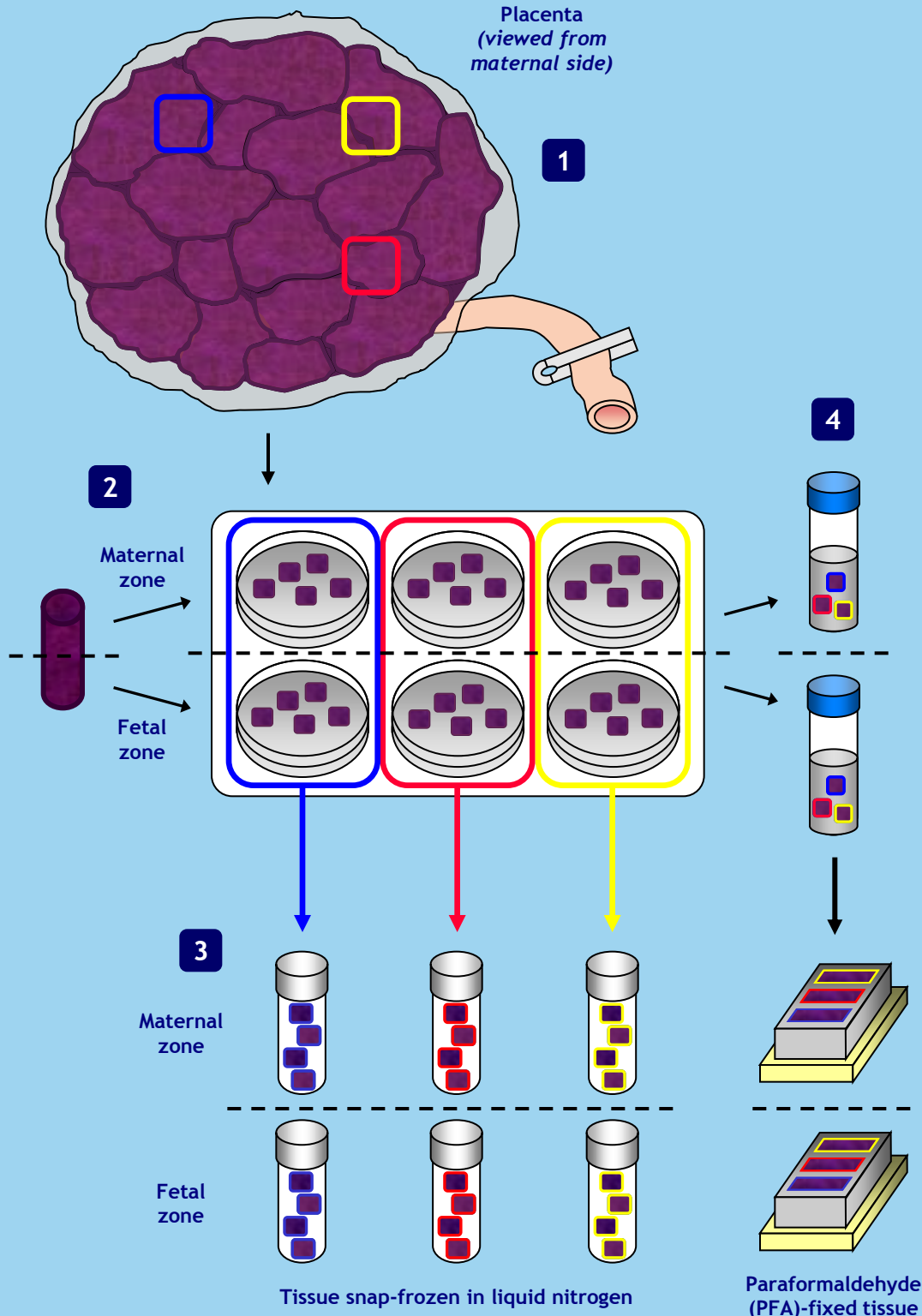
4. 3-4 pieces of tissue from each zone are then placed in a cryomold, covered with OCT medium and frozen on a bath of dry ice. Cryomolds should be labeled so as to allow matching of the maternal and fetal zones from each core.

Tissue snap-frozen in liquid nitrogen

Tissue frozen in Optimal Cutting Temperature (OCT) medium

**Investigator-specified study #3 -
Placenta sampling overview**

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1. Three tissue cores (1.5x1.5cm, full thickness excluding the chorionic plate, taken from at least 1.5cm from the edge) are excised.

2. Each core is cut in half to generate a maternal zone and a fetal zone (collected in 6-well dishes containing PBS). Each zone is then cut into multiple pieces.

3. 3-4 pieces of tissue from each zone are then placed in a cryovial and snap-frozen in liquid nitrogen. Cryovials should be labeled so as to allow matching of the maternal and fetal zones from each core.

4. A piece of tissue from each zone is then fixed in 4% paraformaldehyde for 24h at room temperature, transferred to 70% ethanol and stored until embedding. Maternal and fetal zones are collected in separate tubes to produce representative blocks of maternal and fetal zone tissue.