University of Arizona Cancer Center
PROCEDURE
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SOP #

Cell Pellet Preparation

Rev 0		Effective Date	Author
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1. PURPOSE

The purpose of this standard operating procedure (SOP) is to establish the format for preparing cell blocks of cultured cells.

2. SCOPE

This standard operating procedure applies to all cell lines brought to the Tissue and Cellular/Molecular Analysis Shared Resources within the Arizona Cancer Center.

3. REFERENCE DOCUMENTS

3.1 Thermoscientific Shandon Cytoblock Kit Ref. 7401150
3.2 TACMASR worksheet

4. RESPONSIBILITIES

4.1 Laboratory personnel making cell blocks are responsible for following the procedures outlined in this SOP and notifying the lab manager when deviations or unexpected events arise.
4.2 Laboratory personnel making the cell blocks are responsible for filling out the TACMASR worksheet upon completion.
4.3 Laboratory manager is responsible for all completed projects.

5. MATERIALS and EQUIPMENT

5.1 Thermoscientific Shandon Cytoblock Kit Ref 7401150
5.2 Centrifuge, Sorvall
5.3 Vortex
5.4 Cotton Q-tip applicatiors
5.5 70% Ethanol
5.6 Tissue-Tek marking pencil

6. SAFETY AND CAUTIONARY NOTES

6.1 Gloves and laboratory coat should be worn while performing this procedure.
6.2 Formalin should be discarded in the chemical waste container located in room 0917.

7. PROCEDURE

7.1 Tissue culture cells should be harvested and fixed in formalin for 1 hour at room temperature or over night at 4°C by the investigator prior to bringing them to TAMASR. Ideally cells should be in a 15ml centrifuge tube labeled with the investigators ID.

7.2 Using a Tissue-Tek marking pencil, label the tissue cassette provided in the cell block with the investigators sample ID.

7.3 Gently vortex the 15ml centrifuge tube containing the cells to resuspend the cells. Sometimes cells are so pelleted that after centrifugation it becomes impossible to thoroughly mix the clotting reagents with all the cells if this step is omitted.

7.4 Centrifuge the tube for 3 minutes @ 1500rpm, (setting #3) in the Sorvall LegenRT centrifuge located in room 0915.

7.5 Pour off and discard the formalin, making sure to get all of the formalin off but do not disturb the cell pellet. Use a Q-tip to soak up any remaining drops on the edge of the tube.

7.6 Depending on the size of the cell pellet, while gently vortexing the tube, add 3-4 drops of reagent #2 (blue color dropper). A large pellet will require 4 drops.

7.7 Continue to vortex and add an equal amount of reagent #1, 3-4 drops (clear color dropper). A gel should immediately form.

7.8 Using the wooden end of the Q-tip, transfer the gel pellet to the circumference of the well in labeled tissue cassette.

7.9 Close the cassette and place in 70% ethanol until processing.

7.10 Cassettes are processed on the standard tissue processor program.